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ORAL ABSTRACT SESSIONS
FOOD ALLERGY

1

IL-10 gene polymorphism, but not TGF-β1 gene polymorphisms, is associated with food allergy in a Japanese population

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Background: The regulatory IL-10 and TGF-β1 cytokine gene polymorphisms have been associated with allergic diseases in different populations, like Caucasian, Chinese and Indians. However, no associations between IL-10 and TGF-β1 gene polymorphisms and food allergy (FA) in Japanese children have been evaluated so far. To clarify the relationship of polymorphisms of these 2 regulatory cytokine genes with FA, not atopy itself, polymorphisms IL-10 A—1082G, C—819T and TGF-β1 T+869C, G+915C, C—509T in FA patients were compared with those in non-FA atopic controls.

Methods: One hundred-eleven childhood FA patients, with a mean 7.6±4.0 years of age and 115 atopic control children without FA (mean = 8.2±1.5 years of age) were recruited. Most of FA patients and atopic controls were sensitized with house dust mite (92% and 93%, respectively). DNA samples from these subjects were genotyped by using Real Time PCR.

Results: The odds ratio (OR) of IL-10 —1082 AA genotype was 2.5 (95%CI, 1.0–6.4) for food allergy risk when compared with atopic control subjects (p = 0.03). This study had a power of 80% to achieve significance at the 0.05 level for IL-10 —1082A allele when OR is greater than 2.3. Our OR value was 2.4; therefore, we consider this association had enough statistical power to support our finding. There were no significative differences in the frequency of IL-10 C—819T and TGF-β1 gene polymorphisms between both groups.

Conclusion: This result indicates that IL-10 A—1082G gene polymorphism is associated with food allergy susceptibility in atopic Japanese children. Our findings should encourage further studies to elucidate the functional relationship of IL-10 A—1082G gene polymorphism in FA pathogenesis.

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Safety assessment of bacterial choline oxidase protein introduced in transgenic crops

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Background: Previously, transgenic *Brassica juncea* expressing bacterial choline oxidase protein did not show enhanced allergenicity as evaluated by WHO/FAO guidelines. In the present study, choline oxidase protein derived from *Arthrobacter globiformis* was assessed for allergenicity and toxicity.

Methods: In-vitro heat stability of the protein was assessed. Choline oxidase induced allergenicity was assessed in Balb/c mice. IgE reactivity of choline oxidase protein was assessed with atopic patients' sera. Acute toxicity of choline oxidase protein was also assessed in mice model.

Results: Choline oxidase protein was stable at 90°C for 1 hour and reacted with choline oxidase antibodies on immunoblot. Specific IgE levels were low in choline oxidase treated mice comparable to control, while OVA sensitized mice showed high IgE levels. Intravenous challenge with choline oxidase did not induce any adverse reaction but i.v challenge with OVA led to anaphylaxis in OVA sensitized mice. Choline oxidase sensitized mice demonstrated IL-4 release similar to control in splenic culture supernatant however, OVA sensitized mice showed higher IL-4 levels. Histological analysis of lung tissues from choline oxidase sensitized mice showed intact epithelium with normal airways, whereas OVA sensitized mice showed narrowing of airways with increased eosinophilic infiltration. ELISA with allergic patients' sera (n=45) revealed low specific IgE binding (mean OD 0.281) with choline oxidase protein. Acute toxicity studies of choline oxidase protein in mice model showed no significant difference ($p>0.05$) with control in growth, body weight, food consumption and blood biochemical indices. Histopathology of gut tissues of mice fed with choline oxidase showed normal gastric mucosa lining with normal villi in jejunum and ileum sections.

Conclusion: Bacterial choline oxidase protein is heat stable but failed to elicit allergic response in mice model. These data indicates choline oxidase protein may be safe for use in transgenic crops.

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Identification of amino acids critical for IgE-binding to sequential epitopes of bovine κ-casein and the similarity of these epitopes to the corresponding human κ-casein sequence

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Background: The delineation of allergenic (i.e. IgE-binding) epitopes in cow's milk proteins and the amino acids [AAs] critical for IgE binding is necessary to better understand the structural properties of an allergen and to develop more efficacious immunotherapeutic reagents. Furthermore, this information may enable us to better understand cross-sensitivity between different allergens.

Methods: Eleven peptides, 10 to 14 AAs in length, representing the IgE-binding epitopes of κ-casein were synthesized on a derivatized cellulose membrane with single AA substitutions at each position. Membranes were incubated with pooled sera from 15 milk-allergic patients and individual sera from ten of the patients included in the pool.

Results: For 10 of the 11 allergenic peptides, one to five different single AA substitutions resulted in elimination of IgE-binding of pooled patient sera. Overall at least one mutated peptide could be found for these 10 IgE-binding sites that resulted in a reduction of IgE binding in at least 80% of the patients who recognized the native protein. Furthermore, the IgE-binding region at AA104-112 on bovine κ-casein showed a high degree of similarity with the human κ- casein, respectively, including the AAs critical for IgE binding.

Conclusion: This data suggests that critical AAs should be assessed with both pooled and individual patient sera to account for the B-cell epitope heterogeneity between patients with cow's milk allergy. In addition, we identified two potentially cross-reactive peptides between bovine and human caseins of unknown clinical relevance.

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Food allergen absorption kinetics and gastric digestion influence clinical reactivity in allergic patients

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Background: Severe allergic reactions to minute amounts of ingested food proteins represent major problems for both, the allergic patients and the food industry. Even though protein absorption pattern and physiological protein degradation might substantially influence clinical reactivity in allergic patients only limited knowledge is available to date. Thus, in the present study we used fish as a model antigen aiming to analyze protein uptake and the role of gastric digestion in fish allergy.

Methods and Results: Open challenges of healthy subjects with fish followed by protein determination in blood samples revealed the absorption of biologically active fish proteins within 10 minutes after ingestion. Even though maximal serum levels were measured after 1–2 h, a partial pregastric absorption was indicated. Additionally, simulated gastric fluid experiments, RAST inhibition assay, histamine release and skin testing with fish proteins determined physiological gastric digestion to reduce the allergic potential of fish up to 10,000-fold, which was evidenced also by significantly smaller wheal reactions in skin tests. However, protein digestion was abrogated when digestion experiments were repeated imitating hypoacidic conditions in gastric fluids. Fish digested at pH of 2.75 and above revealed comparable reactivity patterns as undigested extracts. Moreover, these test materials triggered severe clinical reactions at 10- to 30-fold lower cumulated challenge doses in fish allergic patients in blinded challenges.

Conclusion: Our data clearly underline the paramount clinical relevance of gastric digestion in fish allergy. Therefore, insufficient gastric digestion represents a risk factor for fish allergic patients to develop anaphylactic reactions at significantly lowered allergen threshold levels.

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Structural, chemical and immunogenic effects of roasting peanut

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Background: In previous studies, we have shown that processes such as roasting, can alter the allergenic properties of peanuts. To understand these observations at a molecular level, the solubility, digestibility, IgE biding and structural characteristics of Ara h 1 and Ara h 2 purified from raw and roasted peanuts were assessed. Also, skin prick test analysis was performed with raw and roasted peanuts to determine altered immunogenicity.

Methods: The solubility and digestibility of Ara h 1 and Ara h 2 within the context of peanuts roasted to different degrees were assessed using SDS-PAGE and allergen specific antibodies. Ara h 1 and Ara h purified from raw, light roast (LR) and dark roast (DR) peanuts were subjected to circular dichroism spectroscopy before and after reduction of disulfide bonds, digestion with trypsin and pepsin followed by SDS-PAGE, and IgE binding with sera from peanut allergic patients using Western blot analysis. Skin prick test (SPT) were also performed on 5 peanut allergic patients using raw and light roast peanuts.

Results: We found that while the secondary structure of Ara h 2 from raw, LR and DR peanut is largely affected with addition of di-thoithreatol (DTT), the roasting process itself only slightly altered the secondary structure and solubility, while rendering Ara h 2 much more resistant to digestion. Minor changes were seen in the secondary structure of Ara h 1 from raw, LR and DR peanut with the addition of DTT, but the protein became less soluble and more resistant to digestion (with changes in the size and number of digestion resistant fragments) with the degree of roasting. The SPT reactivity to roasted peanut was either equal (2/5) or stronger (3/5) than to raw peanut.

Conclusion: The alterations in IgE binding and resistance to digestion of the peanut proteins is most likely due to chemical modifications and, minutely, if at all, to small structural changes. As previously suggested, the Maillard reaction is most likely the predominant contributor to the decreased solubility and digestibility of the proteins in roasted peanuts. This implies that patients with higher skin test reactivity to the roasted peanut have specific IgE to epitopes that are chemically modified by the roasting process rather than to structurally altered epitopes in addition to other IgE epitopes that are recognized by the majority of peanut allergic patients.

INFLAMMATORY MECHANISMS

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Transfer of exosomes between human mast cells

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Background: Exosomes are 30 to 90 nm membrane vesicles of endocytic origin released by many different cells including dendritic cells, lymphocytes, epithelial cells and mast cells. The function of exosomes is not fully known, but they are believed to take part in communication between cells. The aim of this study was to examine the transfer of exosomes between human mast cells. Mast cell derived exosomes have previously been shown to contain a selection of mRNA and miRNA in addition to proteins.

Methods: Exosomes released from the human mast cell line HMC-1 were isolated by repeated centrifugations and filtrations steps. Exosomes were detected using electron microscopy and flow cytometry. For flow cytometric analysis, exosomes were conjugated to anti-CD63 latex beads and immunostained against the tetraspanins CD9, CD63 and CD81 with PE labelled antibodies. Exosomes were labelled with the green fluorescent stain PKH67, washed and co-cultured with HMC-1 cells. The cells were harvested after 0, 3 and 5 h, washed, fixed in formaldehyde and examined by flow cytometric analysis and by fluorescence microscopy. Results are presented as % positive cells and mean fluorescence intensity (MFI) from two independent experiments (n=4).

Results: HMC-1 exosomes were identified by electron microscopy and by flow cytometric analysis. These exosomes were positive for the tetraspanins CD9, CD63 and CD81 and could be transferred to HMC-1 cells. After 3h incubation, 54% of the HMC-1 cells were positive for PKH67 (MFI 243) and after 5h 71% of the cells were positive (MFI 332) compared to the negative control.

Conclusion: The results demonstrate that human mast cells release exosomes that can be transferred to other human mast cells. This exosome-mediated communication between mast cells may be important in allergic diseases.

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Interferon-β augments eosinophil adhesion-inducing activity of endothelial cells

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Background: Viral infections induce exacerbations of asthma. One of the earliest host responses to viral infections is the production of innate cytokines including type I interferons (IFNs), such as IFN-β, which may act to modify airway inflammation. The objective of this study was to investigate whether IFN-β modifies the eosinophil adhesion-inducing activity of endothelial cells.
Methods: Human umbilical vein endothelial cells (HUVECs) were stimulated with IFN-β for 24 h in the presence or absence of TNF-α. Eosinophils were isolated from the peripheral blood of healthy volunteers. The ability of the IFN-β-stimulated HUVEC monolayers to induce eosinophil adhesion was assessed according to the eosinophil peroxidase assay.
Results: Eosinophil adhesion to HUVECs was significantly augmented by IFN-β not in the absence but in the presence of TNF-α. The augmented adhesion was inhibited by anti-α4 integrin monoclonal antibody (mAb) or anti-β2 integrin mAb. IFN-β significantly enhanced the expression of VCAM-1 and ICAM-1 on HUVECs in the presence of TNF-α.
Conclusion: IFN-β can augment the adhesiveness of endothelial cells to eosinophils, mainly through the expressions of VCAM-1 and ICAM-1. This action of IFN-β may contribute to the intensification of airway inflammation in asthma that is associated with exacerbations induced by viral infections.

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CXC and CC chemokines produced by human respiratory epithelium

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Chemokines regulate leukocyte trafficking during their physiological turnover and recruitment to mucosal surfaces in inflammatory reactions. Respiratory epithelium with the ability to respond to locally generated cytokines might be an important source of chemokines attracting diverse cell populations. With this respect, the aim of our study was to compare the capacity of different proinflammatory cytokines to stimulate the gene expression, production and release of multiple CC and CXC chemokines by respiratory epithelial cell line. The chemokine distribution was studied also in bronchoalveolar lavage fluid (BAL) and exhaled breath condensates (EBC) of patients undergoing lung transplantation.

A total of 96 chemokines and chemokine receptor genes has been studied using an oligoarray system (Superarray Inc.) in cultured human alveolar type-II like cells A549 stimulated by multiple concentrations of TNF alpha, IFN gamma, IL-1 beta, IL-18, and IL-33. The chemokine levels in culture supernatants, BAL, and EBC were measured using multiplex immunoluminometric assay (Luminex) or by ELISA.

In repetitive experiments, epithelial cells constitutively expressed mRNA for CXCL1 (Gro-alpha), CXCL2 (Gro-beta), CXCL3 (Gro-gamma), CXCL5 (ENA-78), CXCL6 (GCP-2), and CXCL8 (IL-8), chemokines attracting preferentially neutrophils. Cells stimulated with IL-1 beta or TNF alpha upregulated mRNA expression of chemokines specific for mononuclear cells recruitment such as CCL2 (MCP-1), CCL4 (MIP 1 beta), CCL5 (RANTES), and CCL20 (MIP 3 alpha). For the induction of of CXCL10 (IP-10) and CXCL11 (I-TAC) which attract activated T lymphocytes, IFN gamma was the most potent stimulus. The induction of epithelial cells by IL-1 related cytokines, IL-33 and IL-18, resulted only in a moderate upregulation of few CC or CXC chemokines compared to a potent effect of IL-1 beta stimulation. Epithelium derived chemokines such as CXCL5, CXCL8, CCL2, CCL4 or CCL5 are abundant in BAL fluids but can be only rarely detected in EBC. We conclude from our data that respiratory epithelial cells are involved in regulating the influx of different populations of inflammatory cells by release of multiple CC and CXC chemokines in a highly coordinated manner.

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The influence of maternal allergy on p38-MAPKinase activity in CD14+ monocytes from 2- and 5-year old children following microbial challenge

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Background: Monocytes are members of the innate branch of the immune system and express a wide range of different pattern-recognition receptors called toll-like receptors (TLRs) that recognize conserved pathogen derived motifs. TLR-mediated signalling, which involves p-38 mitogen activated protein kinase (MAPK) activation, ultimately bridges innate- to adaptive immune responses. We showed previously that cord blood mononuclear cells from neonates with maternal allergy had decreased IL-6 levels upon challenge with the bacterial component and TLR-ligand peptidoglycan (PGN), suggesting that the monocyte population in children of allergic mothers has an altered microbial response already at birth. It is possible that such alteration persists during childhood.
Objective: To examine monocytic microbial responses with regards to p-38 MAPK activation in CD14+ cells and to measure IL-6 release from mononuclear cells in 2- and 5-year old children.
Methods: The 2- and 5-year old subjects (n=61 total) were grouped based on having or not having allergic mothers. Peripheral blood mononuclear cells (PBMCs) from the children were stimulated either with medium, LPS or PGN *in vitro*. CD14+ monocytes were analyzed for p-38 MAPK activity by FACS and IL-6 release from PBMC cultures were quantified by either CBA or ELISA.
Results: PBMCs from 2-year old children with allergic mothers release significantly less ($p<0.05$) IL-6 upon PGN stimuli compared to age matched infants with non-allergic mothers. 2-year old infants with allergic mothers also display markedly reduced CD14+ monocyte p38-MAPK phosphorylation after LPS ($p<0.05$) and PGN ($p<0.01$) challenge. This altered microbial response was attributed to maternal allergy rather than to being IgE-sensitized at 2-years of age. At the age of 5 however, these differences could not be seen between the groups on the basis of maternal allergy. However, children with a developed IgE-mediated allergy tended to have higher levels of phosphorylated p38-MAPK upon challenge with LPS and PGN. They also showed significantly increased basal levels of IL-6 in the culture supernatants.
Conclusion: Maternal allergy renders the monocytic population less responsive to microbial challenge to (at least) the age of 2 while this influence is not noted at the age of 5. However, 5-year old infants with IgE-mediated allergy tended to have higher inflammatory activity both at basal IL-6 levels as well as upon challenge with bacterial components.

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EPIDEMIOLOGY

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Prenatal exposure to household pets influences fetal immunoglobulin E production

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Background: Early life pet exposure may protect against allergic sensitization during childhood. Few studies have evaluated the effect of prenatal pet exposure on potential neonatal markers of allergic risk. We investigated whether maternal exposure to pets affects cord blood IgE levels in a population-based, general risk, ethnically mixed birth cohort.

Methods: Pet keeping during pregnancy was ascertained from women residing in a defined area of Wayne County Michigan U.S.A. and recruited from five staff model obstetric clinics. Maternal venous blood was analyzed for total and allergen-specific IgE along with cord blood total IgE from 808 infants.

Results: During pregnancy, 515 households had no indoor pets and 293 had dogs or cats in the home. Of these 293 households, 147 had dogs only, 96 had cats only, and 50 homes kept both dogs and cats in the home during pregnancy. Compared to infants from households with no cats or dogs kept indoors during pregnancy, infants whose homes had either cats or dogs had significantly reduced mean cord IgE levels [0.33 IU/ml (95%CI 0.29–0.37) versus 0.23 IU/ml (95%CI 0.19–0.27) p = 0.03]. This association was most prominent among children of mothers that did not exhibit IgE sensitization to common allergens [0.24 IU/ml (95%CI 0.20–0.30) versus 0.14 IU/ml (95%CI 0.10–0.16) p = 0.02] and those who did not smoke during pregnancy [0.33 IU/ml (95%CI 0.28–0.37) versus 0.22 IU/ml (95%CI 0.18–0.27) p = 0.01]. There was no effect on results when excluding mothers who reported avoiding pets due to allergy-related concerns.

Conclusion: Mothers with either cats or dogs in their home during pregnancy, especially those mothers who are non-sensitized or are non-smokers, deliver children with lower cord blood IgE levels compared to mothers who do not live with these pets, supporting the hypothesis that pet exposure influences immune development in a manner that is protective for atopy and is operant even before birth.

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Gene-environment interactions between CD14 C-260T and endotoxin exposure on Foxp3+ and Foxp3- CD4+ lymphocyte numbers and total serum IgE in early childhood

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Background: Innate immune system stimuli, such as endotoxin, appear to influence allergy risk. Previously we described gene-environment interaction between an endotoxin receptor polymorphism, CD14 C-260T, and endotoxin exposure on total serum IgE; however, the mechanism of this interaction is not known. The objective of this study was to examine whether this gene-environment interaction influences early CD4+Foxp3- and CD4+ Foxp3+ lymphocyte numbers.

Methods: Participating children were part of a birth cohort in the Detroit, metropolitan area. Participants were genotyped for the CD14 C-260T polymorphism. Endotoxin exposure was estimated from dust measured in the home when children were age 6 months. Intracellular Foxp3 protein expression, a regulatory T-cell (Treg-cell) marker, was used to characterize CD4+ lymphocytes in blood collected at age 12 months; total serum IgE was also measured at this time. Since race-ethnicity may confound or modify genetic associations, we stratified all analyses by race-ethnicity.

Results: We observed a significant gene-environment interaction between CD14 C-260T genotype and endotoxin exposure on CD4+ lymphocyte numbers, particularly CD4+Foxp3- lymphocytes. Stratified analyses suggested effect modification by race-ethnicity on CD4+Foxp3+ lymphocyte numbers with a significant interaction in African-American children but not in white children. The interaction between CD14 C-260T genotype and endotoxin exposure on total IgE levels was opposite that observed for CD4+ lymphocyte numbers, suggesting reciprocal relationships.

Conclusion: A gene-environment interaction between endotoxin and CD14 C-260T genotype on IgE levels may be the result of an upstream, opposing effect on CD4+Foxp3+ and CD4+Foxp3- lymphocyte numbers. Race-ethnicity may influence which of these cell populations is affected by this gene-environment interaction.

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Exposure to animals, allergies and parental education

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Background: Studies have yielded different results regarding the associations between animal exposure and atopy. Whether or not these associations are homogeneous across social strata has not yet been investigated. To estimate the association between current animal exposure (cat and dog) and allergic sensitization and manifestations of atopic diseases in 5 to 7 year old school beginners, stratified by parental educational level.

Methods: 30794 six year old children participated in cross-sectional studies between 1991 and 2000 in Germany. Allergic sensitizations to common areoallergens as measured by skin prick and Radio Allergo Sorbent Test and symptoms and diagnoses of atopic diseases (asthma, atopic dermatitis, hay fever) were the dependent variables. Exposure to dog and cat were the independent variables. Logistic regression was used to adjust for confounding.

Analyses were stratified for parental educational level (stratum I, II, III corresponding to <10 years, 10 years and >10 years of schooling).
Results: Prevalences of hay fever, eczema, specific sensitization to pollen and house dust mite and contact to cat or dog were significantly different between educational strata. Globally significant associations between cat contact and sensitization to cat (RAST OR 1.92, 95% CI 1.18–3.11) and hay fever (OR 0.61, 95% CI 0.45–0.82) remained significant after stratification only in the highest educational stratum (OR 3.68, 95% CI 1.90–7.12 and OR 0.55, 95% CI 0.36–0.85). Globally significant associations of contact to dog with sensitization to pollen (OR 0.71, 95% CI 0.56–0.89), wheezing (OR 1.31, 95% CI 1.11–1.55), nose problems (OR 0.72, 95% CI 0.56–0.93) and eczema (OR 0.84, 95% CI 0.77–0.92) remained significant in educational stratum II. Globally significant associations of dog contact with sneezing attacks (OR=0.84, 95% CI 0.71–1.00) and eczema (OR=0.82, 95% CI 0.75–0.91) remained significant in educational stratum III, and in stratum I and II respectively.
Conclusion: In 5 to 7 year old German children, differences across social strata concerning allergic sensitizations, hay fever and eczema may possibly be explained by animal exposure differences. Associations between animal contact and allergic sensitization and manifestations of atopic diseases are not homogeneous across social strata.

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Maternal childhood exposures, previous pregnancies and breast milk characteristics: an influence on offspring's disease risk?

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Background: Populations in high infectious exposure countries are at low risk of some immune-mediated diseases such as Crohn's disease and allergy. This low risk is maintained on immigration to an industrialized country, but the offspring of such immigrants have a higher immune-mediated disease risk than the indigenous population. We hypothesize that early life exposures in a developing country shape the maternal immune system, and that this stimulates development of her offspring in a manner inappropriate for the low infectious load of a developed county. Other maternal exposures may also influence disease risk in offspring.
Objective: To investigate if exposures in childhood (indicated by country of origin) and subsequent exposures influence immunological characteristics relevant to stimulation of offspring.
Methods: ELISA and Cytometric Bead Array examination of breast milk components among 64 mothers resident in Sweden, 32 of whom immigrated from a developing country.
Results: Immigrants from a developing country had statistically significantly higher levels of breast milk IL-6, IL-8 and TGF-β1. A larger number of previous pregnancies was associated with down-regulation of several substances, statistically significant for soluble CD14 and IL-8.
Conclusion: Childhood exposures influence adult immune characteristics potentially relevant to disease risk in offspring. Such a mechanism may explain the higher immune-mediated disease risk among children of migrants from a developing to developed country. Older siblings may influence disease risk through the action of previous pregnancies on maternal immune characteristics.
Clinical Implication: Maternal immune characteristics resulting from childhood exposures and previous pregnancies may be relevant to disease risk in offspring.

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Prevalence of self-reported allergic rhinitis in China

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Background: Allergic rhinitis (AR) is a common disease with an increasing prevalence. However, little is known about the current national prevalence of AR in China, especially the Mainland China. The aim of this study is to investigate the prevalence of self-reported allergic rhinitis; its association with factors of populations, location, socioeconomic status, meteorology and air pollution; the proportion of persistent and intermittent allergic rhinitis, as well as the percentage of undiagnosed subjects from 11 main cities of the mainland of China.
Methods: Telephone interviews were conducted in 11 main cities of the mainland of China after sampling target phone numbers by the approach of random digital dialing (RDD) via computers. And the questionnaires in Chinese for the telephone interviews were mostly based on the well-validated ones.
Results: A total of 38,203 telephone interviews were conducted from September 2004 to May 2005. The response rate is 63.7%. The self-reported prevalence of allergic rhinitis was the lowest in XiAn (8.0%), and the highest is in Urumqi (21.4%). The gender-adjusted prevalence ranged from 8.5% in XiAn to 21.3% in Urumqi; while the age-adjusted prevalence of self-reported allergic rhinitis ranged from 8.7% in Beijing to 24.1% in Urumqi. The adjusted self-reported prevalence of allergic rhinitis was positively correlated with the concentration of SO2.Within the subjects self-reporting allergic rhinitis, about one-fourth (25.6%) were diagnosed as persistent allergic rhinitis, and the other three-fourth (74.4%) were intermittent allergic rhinitis. In addition, the proportion of persistent allergic rhinitis was higher in northern cities than that in southern cities. Less than half of the subjects with self-reported allergic rhinitis had a history of visiting a health clinic. Among them, only 37.3% were previously diagnosed as allergic rhinitis by physicians and 33.1% subjects received surgical and/or medical treatment.
Conclusion: The study demonstrates that the self-reported prevalence of allergic rhinitis in 11 cities throughout the mainland of China has wide variations. Frequently subjects with self-reported allergic rhinitis could not benefit from proper diagnosis or treatment. Subjects with self-reported allergic rhinitis were mostly diagnosed as intermittent allergic rhinitis, and the proportion of persistent allergic rhinitis was higher in northern cities than that in southern cities.

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Detection of specific IgE by proteomic microarray system based on allergenic molecules: a powerful tool for worldwide epidemiology of allergic sensitization

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Background: The availability of highly purified natural and recombinant allergenic molecules and nanotechnology tools applied to IgE detection allows the collection of huge amount of data in a single test for every single subject. The purpose of this study was to analyse the prevalence of allergenic molecule IgE reactivity within an Italian allergic population and to see whether sensitization to rare allergen are detected as well as most common ones.
Methods: Sera from consecutive patients claiming for allergic symptoms caused by exposure on contact, inhalation, or ingestion were studied. Demographic and clinical information were recorded for each of them. A protein microarray having 90 spotted natural or recombinant allergenic molecules (ISAC CRD 90, VBC-Genomics, Austria) has been used for IgE detection under routine testing conditions. Results were collected, stored and analyzed by customized databases and procedures.
Results: 12,058 consecutive individuals (39.2/60.8% male/female, mean age 32.2±19.1 years, range 1–93) having history of urticaria, rhinitis, asthma, or exzema were recruited from March 2006 to June 2007. Patients with at least

one positive molecule (76% subjects, 42.5/57.5% m/f, and mean age 31±30.3 yrs) represented our study group. The allergens most frequently recognized were from the cypress pollen, Cup a 1 (37.3%), timothy grass pollen, Phl p 1 (36%), and house dust mite, Der f 2 (35%) or Der p 2 (29%). Allergen specific IgE to other molecules were detected less frequently (Bos d 7 26%; Der f 1 23.2%; Par j 2 22.8%; Phl p 2 22.5%; Der p 1 22%; Phl p 5 22%, Par j 1 20%; Ole e 1 20%; Fel d 1 19.3%; Phl p 6 15.4%). Allergens as Bla g 2 and Bla g 5 were rarely positive defining a low exposure by the studied cohort.
Conclusion: Allergenic molecules IgE reactivity detected by microarray system represents a powerful, low-invasive and innovative tool for the molecule-based epidemiology and diagnosis of allergic sensitization. This application allows generating a wealth of information otherwise not available with current singleplex diagnostic tests.

AUTOIMMUNITY

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Immunohistochemical staining for differential diagnosis of cutaneous graft versus host disease and drug reactions

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Purpose of the Study: Cutanous Graft-versus-Host-Disease (GvHD) may show maculopapular rash, bullous dematitis and epidermal necrolysis. Differential diagnosis of GvHD from drug reactions is difficult both clinically and histologically. The immunobiology of GvHD suggests a key role of Dendritic cells (DC) in GvHD pathogenesis. The presence of three different dendritic cell subsets, i.e. plasmacytoid dendritic cells (PDC), inflammatory dendritic epidermal cells (IDEC) and Langerhans cells (LC) and other cell types was analyzed by immunohistochemical staining to improve our understanding of GvHD immunobiology and to evaluate immunohistochemistry as a tool for differential diagnosis of GvHD and drug reactions.
Methods: Skin biopsies of patients with clinically diagnosed GvHD (n=27) and drug reactions (n=14) were processed for immunohistochemistry in APAAP technique. Skin sections were stained and semi-quantitatively analyzed for CD1a, CD1b,CD2, CD4, CD8, CD11c, CD20, CD25, CD68, CD206, CD207, CD208 and CD209, BDCA-2 and CCR-7.
Results and Discussion: A relevant dermal infiltration (>20 cells per mm^2) of BDCA-2+ PDC was specific for GvHD. The presence of the PDC correlated significantly with the presence of DC expressing CD1b, CD11c and CCR-7. Hence, the presence of many PDC supports the clinical diagnosis of GvHD. CD1a and CD207 expressing dermal dendritic cells were present in GvHD and drug reactions, whereas high amounts of CD207 expressing epidermal LC were more frequently observed in GvHD. T cells expressing CD2, CD4 or CD8 were equally observed in both patient groups.
Conclusion: The observed differences between a PDC dominated dermal infiltrate in GvHD and a myeloid DC dominated dermal infiltrate in drug reactions should be confirmed by an independent study, as these differences may become clinically useful for differential diagnosis of GvHD from drug reactions.

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Unusual biphasic presentation of intestinal lymphangiectasia: a case report

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Introduction: Chronic diarrhea in adults is a frequent cause of consultation, in the United States, with prevalence between 3–5%. Among the rarest causes of diarrhea are, protein-losing gastroenteropathies. Intestinal lymphangiectasia (InL). Is a rare entity which can be congenital or acquired it is characterized by abnormal, dilated lymphatics throughout the intestinal tract. The disorder leads to leakage of lymph causing hypoproteinemia, lymphopenia and hypogamaglobulinemia, which can leads to depression of both the humoral and cellular immune systems.
Case Report: A 35 year old woman from rural Mexico with a history of asthma for 20 years, presented with a chief complaint of diarrhea for 15 months with up to 10 evacuations a day, she referred colic pain with abdominal distension and flatulence with recurrent symmetrical systemic edema. The patient showed fatigue, weaknesses and muscle spasms as well as myalgias and fasciculations, she lost 22 lbs and developed onychomycosis in all of her toenails. She recalls showing the same clinical signs and symptoms once during childhood. Her medical team did not have a clear diagnosis. Laboratory revealed a lymphopenia, hypoalbuminemia, hypogammaglobulinemia, hypocalcemia and negative HIV by ELISA. The stool Ü1-antitrypsin level was elevated. The CT of chest and abdomen was normal. An esophagogastroduodenoscopy and colonoscopy were performed showing gastritis, a duodenal biopsy demonstrates dilated lymphatics and white villi spots.
Discussion: The lack of elevated levels of protein in the urine along with the depressed serum levels of IgG drew attention to the gastrointestinal tract as the likely portal of protein loss. Detecting an elevated level of α1-antitrypsin in a 24-hour stool collection adds support to the diagnosis of InL. A duodenal biopsy with the characteristic histological findings, demonstrating dilated lymphatics and white spots, confirmed the diagnosis. Treatment of primary InL is symptomatic and only marginally effective. We treat our patient with strict dietary lipid restrictions, pancreatic enzymes, probiotics, diuretics, Medium-chain triglycerides and multivitamins. The patient showed a marked response regained her normal weight, but the diarrhea and edema sporadically presents. To date is the seventh reported case of primary InL in Mexico and to our knowledge the only described in literature of biphasic presentation.

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Atrial natriuretic peptide receptor (NPRA) signaling in human dendritic cells controls treg development

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The natriuretic peptides (NP) are key endogenous factors that control inflammation and immune tolerance, the latter is essential to maintain immune homeostasis, control autoreactive T cells, prevent the onset of autoimmune diseases and achieve tolerance of transplants. The mechanism of regulation of ANP-NPRA signaling in DCs is crucial for the understanding of how ANP regulates innate immunity and how failure of this signaling might contribute to autoimmune disease, chronic inflammation and tissue damage. Herein, the role of the c-terminal NP, ANP and one tolerance inducing NP, NP73-102, in modulating DC function was examined. The results demonstrate that in contrast to ANP, NP73-102 primes DCs to induce regulatory T (Treg) cells. The ANP receptor, NPRA, binds to TLR-2, SOCS3 and STAT3 and affects induction of IL-6, IL-10 and TGF-α, not IL-17. SOCS3 expression is controlled in a MyD88-dependent manner. Also, down-regulation of SOCS3 and TLR2, but not STAT3, affects NPRA expression. These results demonstrate that TLR2, and SOCS3 are key players in integrating ANP-NPRA signaling with innate immunity and provide insight into how inhibition of IRNP affects ANP-NPRA signaling promoting the tolerogenic phenotype of DCs.

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Immune response to Rotavirus in the pathogenesis of celiac disease: molecular effects of anti-VP7 viral protein antibodies on intestinal epithelial cells using a gene array approach

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Background: Celiac disease (CD) is sustained by an inappropriate response against gluten in genetically susceptible individuals. In addiction to genetic and dietary factors, also rotavirus infections have been implicated in CD pathogenesis. Specific recognition of a peptide sharing omology with the VP-7 rotavirus protein by sera of CD patients with active disease has been recently demonstrated. This study evaluated the functional modifications induced by anti-viral peptide antibodies in intestinal epithelial cells.

Methods: Specific antibodies directed against the viral-derived VP7 peptide were affinity-purified from the sera of patients with active CD. The effect of these antibodies cross-reacting with self antigens on T84 intestinal cells were analyzed with the gene array technique. Genes up- and down-regulated were identified by a 2-fold or more change in expression. The analysis was validated by quantitative PCR. The different gene expression patterns were analyzed by using the Array Assist ™ software (Stratagene, La Jolla, California, United States) and a functional clusterization was performed.

Results: Exposure of T84 cells to anti-VP7 antibodies resulted in the upregulation of 697 transcripts with a 2-fold or more increase in expression. Several gene clusters were upregulated including genes encoding for chemokines and molecules involved in inflammation and immune response processes, as well as in apoptosis and cell proliferation regulation.

Conclusion: The exposure of intestinal epithelial cells to anti-VP7 rotavirus antibodies modulates clusters of genes similar to those modulated during CD. These results confirm the important role played by rotavirus infection in the pathogenesis of celiac disease.

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A retrospective study of autoimmune bullous diseases

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Background: Autoimmune Bullous Diseases cause impaired adhesion of epidermis to epidermal basement membrane or impaired adhesion of epidermal cells to each other with characterized by substantial morbidity (pruritus, pain, disfigurement) and in some instance, mortality (secondary to loss of epidermal barrier function).

Objective: The aim of this study is to evaluate the frequency distribution of Autoimmune Bullous Diseases (Pemphigus Vulgaris, Pemphigus Foliaceus, Bullous Pemphigoid, Cicatrical Pemphigoid, Dermatitis Herpetiformis, Epidermolysis Bullosa, Linear IgA Bullous Dermatosis).

Methods: A retrospective study was conducted on the medical record of Autoimmune Bullous Diseases patients that visited to Dr. Wahidin Sudirohusodo General Hospital Makassar, Indonesia for a year (January 2006 to December 2006).

Results: In this retrospective study, there were 6 patients of Pemphigus Vulgaris 4 males and 2 females with ages between 19–35 years old. Three patients of Bullous Pemphigoid 1 male and 2 females, with ages between 23–64 years old. One male patient of Dermatitis Herpetiformis with age 42 years old. There was not patient of Pemphigus Foliaceus, Cicatrical Pemphigoid, Linear IgA Bullous Dermatosis, Epidermolysis Bullosa. All patients recovered from disease.

Conclusion: There has been reported only 10 patients of Autoimmune bullous diseases for a year (January 2006 to December 2006) in Dr. Wahidin Sudirohusodo General Hospital Makassar, Indonesia. This is a rare case. All the patients were treated by systemic corticosteroid and another therapies that suitable for standard therapies and give good result.

Keywords: Autoimmune Bullous Diseases, retrospective study.

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Autoimmunity in families of IgAD individuals

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Background: The prevalence of autoimmune diseases is thought to be increased among IgA deficient individuals (IgAD). The prevalence of autoimmune diseases among IgAD Icelandic individuals and their first degree relatives was investigated. This is a part of a larger ongoing study aimed to evaluate the prevalence, geneaology, complications and the genetic components of IgA deficiency (IgAD) in Iceland.

Materials and Methods: A total of 43 IgAD individuals have been found through the screening of blood donors (5), re-evaluating IgAD individuals from a previous IgAD study from 1977 (10) and by evaluating individuals that were found to have low IgA from 1990 until present at the Institute of Laboratory medicine, Landspitali- University hospital of Iceland (28) Clinical evaluation was done by standard questionnaires that focused on symptoms and signs suggestive of infections, allergies, autoimmunity and cancer.

Results: Among the 43 IgAD individuals, 21% were found to have definite autoimmunity (9/43). Of those, four had two or more autoimmune diseases. This is much higher than has been reported for the general population. Amongst organ specific autoimmunity, thyroid dieases were the most common found (4/9). Whereas, Rheumatoid arthritis was the most common systemic autoimmunity found (2/9). In first degree relatives with IgAD 20/43 or 46.5% had history of autoimmune diseases, RA, DM type1 and autoimmune thyroid diseases being the most common.

Conclusion: Autoimmune diseases are common in patents with selective IgA deficiency. Since the prevalence of various autoimmunity are found to be high amongst IgAD first degree relatives, this suggest that a strong common genetic component could be associated with both conditions. Thus, demonstrating that autoimmunity is an important complications associated with IgA deficiency.

MECHANISMS OF ASTHMA I

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A critical role for atrial natriuretic peptide receptor signaling in allergic disease

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Atrial natriuretic peptide (ANP), which has been extensively studied for cardiovascular effects, has received little attention for its role in immunity and in inflammation. In humans, ANP modifies airway hyperreactivity, however, the precise role of ANP in the lungs and on local inflammationand immunity remains to be established. Herein, we show that mice deficient in the atrial natriuretic peptide (ANP) receptor, NPRA, are resistant to developing allergen induced pulmonary inflammation, T helper type (Th2) response and eosinophilia. Chitosan nanocomplexes of a novel peptide, NP73-102 or a plasmid encoding the peptide, that inhibits NPRA expression, or a short interference RNA (siRNA) for the NPRA induces bronchoprotective and anti-inflammatory activity including decreased eosinophilia and Th2 cytokines in the lung and reverses asthma in mice. In addition, mice deficient in NPRA and mice with decreased expression of NPRA exhibit an increase in FoxP3+T regulatory cells. Together, these results show that NPRA

signalling may provide a novel target for developing new treatment for asthma.

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Up-regulation of epithelial and fibroblast prostaglandin D2 CRTH2-receptors in asthma

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Background: Mast cell-derived prostaglandin D2 (PGD2), now known to act through DP1 and CRTH2 receptors is regarded a central mediator in many allergic diseases including asthma. Blockade of CRTH2 receptors has been shown to inhibit airway eosinophilia and mucus production in allergic mice (Uller et al Respiratory Research 2007:8:16). However, it is not known if airway structural cells express these receptors or produce PGD2. Thus, this study explores whether epithelial cells and fibroblasts obtained from asthmatic and healthy airways express CRTH2 and DP1 receptors in response to factors involved in causing asthma exacerbation.

Methods: Using well-validated cell culture techniques, primary bronchial epithelial cells (PBECs) and primary lung fibroblasts from normal or asthmatic subjects were grown from bronchial brushings and biopsies, respectively. PBECs were seeded in 12 well plates and stimulated with the bacterial product LPS (0.1–10 ?g) or virus analogue PolyIC (0.01–10 ?g). Primary lung fibroblasts were stimulated with the mediators IL-13, TGF-beta and infected with the minor rhinovirus RV1?. Cells were harvested at different time points after stimulation using Trizol extraction for RNA. Then mRNA levels for PGD2 synthase and its receptors were quantified by RT-qPCR.

Results: PBECs stimulated with Poly:IC or LPS resulted in a dose-dependent up-regulation of toll-receptor TLR3 and TLR4 and proinflammatory cytokines IL-8, IFN-beta, and IFN-lambda. PBECs stimulated with Poly:IC exhibited no up-regulation of CRTH2 whereas LPS stimulation dose-dependently up-regulated the CRTH2 receptor. Neither LPS nor Poly:IC stimulation up-regulated DP1-receptors in PBECs. In bronchial fibroblasts the allergic remodelling cytokines (IL-13 and TGF-beta), up-regulated the expression of CRTH2 and, additionally, induced PGD2 synthase production.

Conclusion: We demonstrate that airway structural cells in asthma such as epithelial cells and fibroblasts harbour CRTH2-receptors which are further up-regulated at exposure to bacterial products and allergic cytokines, respectively. Hence, studies on roles of PGD2 and, particularly, CRTH2 receptors in remodelling of asthmatic airways and at exacerbations of the disease are warranted.

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Association of acidic mammalian chitinase (AMCase) with atopic asthma and serum total IgE

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Background: AMCase, an enzyme known to degrade chitin, has been shown to be highly expressed in asthmatic lungs. It has also been shown to contribute towards asthma pathogenesis by affecting the IL-13 downstream pathway. Hence, *AMCase* is a potential candidate for asthma genetic studies.

Methods: To investigate the association of the *AMCase* polymorphisms and haplotypes with atopic asthma and total serum IgE levels in Indian population; Polymorphisms were identified by sequencing 60 unrelated subjects. On the basis of LD and heterozygosity index, a total of six SNPs were short-listed and further genotyped for 270 asthmatic subjects and 292 non-asthmatic unrelated controls and were analyzed for genotype and haplotype association. The results were confirmed using an independent paediatric cohort (Patients=150; Controls=101). Electrophoretic mobility shift assay (EMSA) was performed to check the functional significance of the C–1261T promoter SNP.

Results: The G–1279A promoter polymorphism and the G29968A SNP showed significant association with atopic asthma ($p<0.05$). Moreover, the G–1279A and C–1261T promoter polymorphism was found to be associated with serum total IgE in the patients ($p<0.05$). At the haplotype level, *ATGG* was found to be a major risk ($p<0.05$), while *GTGA* was found to be a protective haplotype ($p<0.05$). Using electrophoretic mobility shift assay, we demonstrated that the C to T substitution at –1261 position abolished transcription factor *Oct-1* binding site.

Conclusion: The results suggest that genetic variation in the *AMCase* gene promoter influences susceptibility to asthma and serum total IgE.

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Direct activation of natural killer T cells induces airway hyperreactivity in a non-human primate model of asthma

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Background: In mouse models of asthma, we and others demonstrated that the presence of natural killer T (NKT) cells is required for the development of airway hyperreactivity (AHR), a cardinal feature of asthma. Thus, AHR failed to occur in the absence of NKT cells. Furthermore, direct activation of NKT cells with glycolipid antigens alone was sufficient to induce AHR, in the complete absence of conventional CD4+T cells and adaptive immunity. These findings indicated that NKT cells are both necessary and sufficient for the induction of AHR in mice. Moreover, NKT cells are increased in number in the lungs of patients with asthma compared to that of healthy control subjects or patients with sarcoidosis, strongly suggesting that NKT cells play an important role in human asthma.

Purpose: To examine the functional role of pulmonary NKT cells in primates, we used a Cynomolgus monkey model of asthma, and determined whether the direct activation of pulmonary NKT cells results in the development of AHR.

Methods: Using a cross-over study design in which each monkey served as its own control, and which minimized the number of monkeys used, we intratracheally challenged four monkeys with vehicle control or a low dose of á-Galactosyl Ceramide (á-GalCer), a glycolipid that specifically activates NKT cells. The monkeys were anesthetized, intubated and intratracheally challenged either with vehicle (two monkeys) or á-GalCer (two monkeys). Twenty-four hours later, AHR was evaluated by measuring airway resistance and dynamic compliance in response to increasing doses of methacholine. Six weeks later, the monkeys were again intubated and challenged with the alternative treatment (á-GalCer or vehicle), followed by assessment of AHR.

Results: All four monkeys tolerated challenge with á-GalCer. Furthermore, after airway challenge with á-GalCer, all four monkeys demonstrated increased AHR, significantly greater than that induced with the vehicle control.

Conclusion: Direct activation of pulmonary NKT cells with á-GalCer induced AHR in a non-human primate model. Therefore, NKT cells play an important functional role in the development of AHR in both mice and in primates.

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Dolichyl phosphate dependent mechanism of steroid resistant asthma development

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Background: In most cases of steroid resistant asthma (SRA) there is a functional hyporesponsiveness of glucocorticoid receptors (GR) on T cells and

disbalance in activity of IL-2, IL-4, IL-10. Dolichyl phosphate (DolP) plays an essential role in cytokine synthesis and in constancy of glycoproteins of the GR and activity of P-glycoprotein. The present study was carried out to estimate the possible role of DolP in mechanism of steroid resistant development.

Methods: The samples obtained from 128 patients with asthma: 60 patients with steroid sensitive asthma (SSA) and 68 patients with SRA and from 44 donors. 1) Dolichyl phosphate was defined in T-cells. 2) Alpha-and beta-GR isoforms expression were measured in patients with SRA and SSA. 3) T-cells taken from SRA were cultured in vitro with DolP 4) P-glycoprotein MDR1 expression was assessed by immunohistochemical technique 5) Dolichyl phosphate N-acetylglucosamine-1-phosphate transferase (GPT) due to DPAGT1 polymorphism was assessed in T-cells.

Results: 1) Blood Dol in patients with SRA was increased up to six times making up 689.2 + 47.9ng/mL and urinary Dol concentration was increased up to 590.9%, making up to 48.8 + 9.7 mg/mmol in comparison with SSA patients. 2) The synthesis of DolP was 8.8-10.5-fold decreased in T-lymphocytes in patients with SRA. 3) SRA T-cells membrans contain 5,6–6,4% of P-glycoprotein-170 (the total protein amount) as a resistance marker. SRA T-cells differ from sensitive ones in Pgp content by 10-12 times. 4) DolP in the concentration 10–6 M aid 7–9-fold reducing P-glycoprotein-170 content in membranes of SRA T-cells to 0,4–0,6%. 5) T-cells from SRA patients cultivated with corticosteroids and DolP (Polyprenols) restore the possibility to induce IL-10 synthesis in vitro. 6) The DolP concentration in SRA T-cells was returned to the normal level. Adding polyprenol to culture of T-cells from SRA patients enhanced the expression of alpha GP isoforms and made these cells more responsive to steroids.

Conclusion: The results show the evidence that DolP is rate limiting mechanism of steroid resistance in asthma, associatedThe situation can be changed by resistant T-cells treatment with DolP substitute polyprenol. with hyperactivity of P-glycoprotein and a marked defect of GR glycosylation in T-cells. It is, also, a hypothesis, which has suggested that there is a genetic polymorphism of DPAGT1 that blunts the response to steroids.

RISK FACTORS FOR ALLERGY

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Total IgE levels and in vitro sensitization to Ascaris spp., dermatophagoides pteronyssinus, Blomia tropicalis and Blatella germanica in the tropical Island of Martinique

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Background: We have previously demonstrated that the presence detectable specific IgE to *Ascaris* spp., increases the prevalence of positive skin tests to common aeroallergens, especially mites and cockroaches, and to food allergens, such as shrimp in Martinique.

Objective: The objective of this study was to analyse the correlation between sensitization to *Ascaris* spp., total IgE levels, and in vitro sensitization to *Dermatophagoides pteronyssinus*, *Blomia tropicalis* and *Blatella germanica* in a population of patients residing in the tropical Island of Martinique.

Materials and Methods: 607 consecutive patients (321 females and 286 males) were evaluated at a local allergy clinic for allergic respiratory complaints from February 2003 to March 2007. Mean age was 20.36 (1–75 years). Specific IgE was determined by the CAP method (Phadia). The patients were divided into 2 groups: Group 1: 333 patients with <15 years of age, and Group 2: 274 patients with >15 years of age.

Results: Mean total IgE levels in the studied population was 752.03 kU/L (2–39.888); 238 patients (39.2%) had a positive specific IgE determination to *Ascaris* spp; 387 (63.76%) to *D. pteronyssinus*; 399 (65.73%) to *B. tropicalis* and 218 to *B. germanica* (35.9%). In Group 1: mean total IgE levels: 987.83 kU/L (2.13–39.888); 147 (44.14%) were positive to *Ascaris* spp.; 234 (70.27%) to *D. pteronyssinus* and *B. tropicalis* and 134 (40.24%) to *B. germanica*. In group 2, mean total IgE: 465.46 kU/L (2–8.453); 91 (33.2%) were positive to *Ascaris* spp; 153 (55.84%) to *D. pteronyssinus*; 165 (60.22%) to *B. tropicalis* and 84 (30.66%) to *B. germanica*. In group 1, 146 of the 147 *Ascaris* spp. positive patients (99.32%) were positive to at least 1 mite and 142 to both species; 186 patients in this group were negative to *Ascaris* spp.; in this group 98 (52.69%) were positive to mites ($p<0.05$). In group 2: 91 (33.21%) were positive to *Ascaris* spp. and among them, 81 were positive to at least 1 mite species (89.01%); in the *Ascaris* spp. negative group (183), 97 (53%) were positive to at least one mite species ($p<0.05$).

Conclusion: Sensitization to *Ascaris* spp. seems to be a risk factor for sensitization to mite allergens in the tropics.

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Study on the FcERIβ gene polymorphism in pollen allergic population of Calcutta city

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Background: IgE dependent activation of mast cells and basophils through the high affinity IgE receptor (FcERIβ) is involved in the pathogenesis of type 1 respiratory allergy. The FcERIβ gene is located on chromosome 11q13, showing linkage to atopy and asthma. Mutations in this gene could alter IL-4 production and thus modify IgE levels. An amino acid substitution at position 237 in this gene has been reported to be associated with atopic asthma phenotypes in Japanese and white population. The present study aims to investigate the relationship between the atopic asthma phenotypes and the FcERIβ gene polymorphism in the population of Calcutta city.

Method: 232 adult pollen allergic patients from Calcutta participated in an asthma and allergy phenotype- genotype study. Phenotypes obtained by studying case history, total and specific IgE, IL-4 level, forced expiratory volume in 1 second (FEV1), forced vital capacity (FVC), etc. Genotyping was done by restriction endonuclease fragment length polymorphism of a polymerase chain reaction product spanning the E237G polymorphism site of FcERIβ gene on the DNA samples isolated from the peripheral blood samples of study and control group.

Results: Among 232 subjects 44.4% were suffering from allergic rhinitis, 55.6% from bronchial asthma and 19.8% from combination of both. The skin reaction diameter showed positive correlation ($p<0.01$) with specific IgE, FEV1, IL-4 level and no correlation with total IgE. The restriction endonuclease fragment length polymorphism study of E237G variant of FcERIβ gene showed no significant difference with control group.

Conclusion: The pollen allergic patients have specific IgE level correlated with skin reaction. In the tested population of Calcutta city it has been found that the E237G variant of FcERIβ gene is not at all involved.

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Deworming improves current wheeze and temporarily deteriorates atopy: longitudinal anthelminthic treatment studies in Cuban schoolchildren

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Background: Although helminth infections have been suggested to protect from atopy and atopic diseases, there is still no consensus on their

relationship. We investigated the effect of deworming and intestinal helminth (re)infections on atopy, asthma, allergic rhinoconjunctivitis and atopic dermatitis.

Methods: We examined 440 4–13 year-old Cuban schoolchildren in six-monthly intervals for 24 months. Intestinal helminth infections were diagnosed by stool examination. Atopic diseases were diagnosed by ISAAC (International Study of Asthma and Allergies in Childhood) questionnaire, asthma additionally by spirometry, and atopy by skin prick testing (SPT).

Results: After deworming the frequency of current wheeze ($p<0.001$) and allergic rhinoconjunctivitis ($p = 0.015$) significantly decreased. The percentage of SPT positives temporarily increased from 9.7% (95% CI: 5.5–16.6%) to 32.7% (95% CI: 24.7–42.9%) ($p<0.001$) and subsequently returned to baseline values (11.9%, 95% CI: 6.9–19.6%). (Re)infection with *A. lumbricoides* and *T. trichuria* was positively and hookworm negatively associated with the development or retention of these atopic diseases, while for atopy an opposite trend was seen.

Conclusion: Our data indicate that atopic diseases improve after anthelminthic treatment. Atopy on the other hand increases after deworming. As this increase appears only temporarily, deworming of schoolchildren does not seem to be a risk factor for the development of atopy, nor for atopic disease. Effects of helminth (re)infections on atopy and atopic diseases appear to be species-specific.

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Association of cord blood cytokine production with wheezy infants in the first year of life

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Background: Previous studies indicate that antenatal factors are important in determining susceptibility to asthma. Since cord-blood cytokine productions may influence the development of atopy-predisposing immune response, cord-blood cytokine productions may be an important predictor for asthma. We investigated antenatal cytokines in a prospective birth cohort, intensively monitored for wheezy infants outcome at 1 year.

Methods: Cord-blood serum samples from 234 children were assayed for interleukin (IL)-1β,-2,-4,-8,-10,-12,-13, and-17, interferon-γ, tumor necrosis factor-α (TNF-α), granulocyte-macrophage colony-stimulating factor (GM-CSF), granulocyte colony-stimulating factor (G-CSF), monocyte chemotactic protein-1 (MCP-1), and macrophage inflammatory protein-1-β (MIP-1β). Associations between family history, antenatal and perinatal factors, cord-blood cytokine concentrations, and wheezy infant outcomes (wheezing more than two times) were analyzed.

Results: Data were obtained for 213, including 33 wheezing, infants. Risk of wheezing was related to gestational age, birth weight, cesarean section, and maternal eczema, but not to bacterial/viral infection during pregnancy, maternal asthma, maternal smoking or paternal history. High level of IL-8 had a significant association with wheezy infant outcome at 1 year ($p = 0.025$). IL-5 and-7, MCP1, and MIP-1β were high with maternal bacterial infection, and IL-13 was high with maternal viral infection during pregnancy. Moreover, there were associations between high levels of IL-5 and GM-CSF, and cesarean section. When we included gestational age, birth weight, cesarean section, maternal eczema and cord-blood IL-8 concentration in multivariate logistic regression analysis, birth weight (odds ratio = 0.998, 95% CI = 0.997–1.000) and maternal eczema (odds ratio = 5.356, 95% CI = 1.340–21.41), but no other factors, were significant predictors of wheezy infants.

Conclusion: Maternal eczema and prematurity were important risk factors for wheezing in the first year of life. Infection during pregnancy and cesarean section was related to cord-blood cytokine concentrations. Cord-blood IL-8 may be a predictor for recurrent wheezing at 1year.

35 Abstract withdrawn

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Atopy in young children with asthma

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Background: A close association between outcome in patients with early-onset asthma and atopy has been reported in several population studies. Allergic sensitization in early life seems to be an important risk factor for subsequently persistent asthma during childhood and adulthood. It might be valuable for evaluation of atopy in patients with early-onset asthma in order to predict prognosis and take early intervention.

Methods: Clinical history data of 62 asthmatic children under 4 years of age were collected. The atopy status of each patient was determined by both personal allergic history (eczema and/or allergic rhinitis) and specific diagnosis of allergens (screening tests of fx5E, mx2 and Phadiatop conducted by fluoroenzyme-immunometric assay using the UniCAP100 system). The total serum IgE level was also measured. Logistic regression was used to analyze the effect of clinical characteristics on allergic sensitization.

Results: In comparison with that 74.2% children reported personal history of atopy and 33.9% reported parents' history of atopy based on the clinical history data, the positive rates of fx5E, mx2, Phadiatop were 40.3%, 14.5%, 14.5% respectively. The total allergic sensitization screening test rate was 46.8% and the sensitization rate to inhalant allergens was 24.2%. The allergic history of parent(s), the sensitization to food allergen, the age of first wheezing attack and total serum IgE level were main factors influencing sensitization to inhalant allergens.

Conclusion: Almost half of asthmatic children under 4 years of age were shown as positive atopy tests and a quarter of those patients were sensitized to inhalant allergens. The asthmatic history of parent(s), the sensitization to food allergens, the age of first wheezing attack greater than 2 years and the significantly higher total serum IgE level may increase the possibility of sensitization to inhalant allergens in asthmatic children under 4 years of age.

ASTHMA EPIDEMIOLOGY

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Do boys do the "atopic march" while girls dawdle?

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Background: The "atopic march" hypothesis suggests that infants with eczema are at increased risk of asthma and allergic rhinitis. Others argue that eczema is not a risk factor for asthma or allergic rhinitis, unless there is also sensitisation or early wheezing. The role of infantile eczema as a predictor of risk of childhood asthma and allergic rhinitis was examined in a prospective birth cohort, while controlling statistically for the effects of early wheeze, sensitisation and gender.

Methods: A birth cohort of 620 infants with an atopic family history was recruited. Presence of eczema and wheeze was prospectively documented up to 2 years of age. Sensitisation was determined by skin prick tests (SPT) at 6, 12 and 24 months to 6 common food and inhalant allergens. Interviews were conducted at 6 and 7 years to ascertain asthma in the previous 12 months.

Results: Eczema within the first two years of life was associated with an increased risk of childhood asthma in boys (Odds Ratio [OR] = 3.05, 95%CI 1.74–5.35) but not in girls (OR = 1.44, 95%CI 0.74–2.78, p for interaction = 0.031). There was a similar pattern for allergic rhinitis, where eczema in boys conveyed increased risk (OR = 2.37, 1.32–4.23) but less so in girls (OR = 1.17, 0.58–2.39, p for interaction = 0.123). Adjusting for allergic sensitisation in early life reduced the strength of both these associations in boys by approximately 20% (adjusted OR 2.45, 1.32–4.56 for risk of asthma, and aOR 1.84, 1.02–3.35 for allergic rhinitis) but adjustment for wheeze in the first two years did not substantially alter these associations. If it is assumed that these relationships are causal, the maximum potential reduction in childhood asthma and allergic rhinitis by an intervention to eliminate eczema in boys is approximately 28%.

Conclusion: Eczema in the first two years of life is associated with an increased risk of childhood asthma and allergic rhinitis in boys but not in girls.

These results suggest that the "atopic march" may be more important in boys, and interventions should be targeted at them.

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New onset of asthma, chronic phlegm and lung function decline in relation to indoor moulds and building dampness: A nine year follow up study within ECRHS-II

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The aim was to study new onset of adult asthma, chronic phlegm, chronic bronchitis and lung function decline in relation to indoor dampness and moulds in a 9-year follow-up study (ECRHSII) A follow-up took place from 1998 to 2002, with 4194 men and 4576 women from 27 centres. New asthma and wheeze at the follow-up was studied, excluding those (N = 1605) with either wheeze without a cold, nocturnal attack of shortness of breath or asthma at baseline. New onset of chronic phlegm at follow up was studied, excluding 1716 subjects with usual cough or phlegm at baseline. Self-reported dampness and indoor moulds in the dwelling (ever) and last 12 months was assessed by yes/no questions. A dampness score (DSC) and a mould score (MSC) ware calculated, by summing up number of signs. Moreover, 3118 homes were inspected for observed dampness (OD) and moulds. Adjusted relative risk (RR) were calculated for those ever exposed, or getting new exposure during the follow up, using never exposed as reference group. Totally 43.0% had ever lived in a home with water damage, 43.3% ever in a home with moulds. There was a significant increase of new asthma among those reporting moulds, water damage, and indoor damp spots at home (RR 1.3–2.3). Moreover, there was an association between observed dampness (OD) and new asthma (RR = 1.5; 95% CI 1.0–2.2), between OD in the bedroom and new wheeze (RR = 1.9; 95% CI 1.2–3.0), and between OD in the bedroom and new asthma with BHR (RR = 2.8; 95% CI 1.0–8.1). The effect of indoor moulds on new asthma was stronger in those sensitised to moulds. There was an increased risk of now chronic phlegm among those reporting new exposure to moulds (RR = 1.47; 95% CI 1.01–2.13) and new water damage (RR = 1.54). Females with any reports of dampness at home had an additional decline in FEV1 of −2.25 ml/year (95% CI–4.25 to −0.25), and there was a significant trend in lung function decline in relation to DSC in females (p = 0.03). Moreover, OD in the bedroom was related to a decline in FEV1 of −7.43 mL/year (95% CI-13.11 to −1.74). Meta-analysis did not reveal any centre-heterogeneity of the findings. In conclusion, dampness and moulds in the dwelling can be a risk factor for new asthma and new chronic phlegm in adults, and for lung function decline in females. Mould allergy seems to be a risk factor for asthma in damp and mouldy dwellings.

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Beta-glucan exposure and allergic disease in infancy

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Background: Early life exposure to microbial agents may protect against the development of allergic disease in infancy. This study assessed whether exposure to beta-glucan (a fungal biomass marker) at age 3 months was associated with allergic disease and asthma symptoms in infancy.

Methods: Living room floor dust was collected at age 3 months and analysed for beta-glucan by a modified Limulus amoebocyte lysate method. Skin prick tests to indoor allergens were performed on 546 infants at age 15 months for whom dust was available for analysis and asthma symptoms were collected by questionnaire then and at age 2, 3 and 4 years. Adjusted odds ratios (aOR) for the association of beta-glucan with allergic disease and asthma symptoms were determined by logistic regression. Adjusted geometric mean ratios were used to show determinants of beta-glucan.

Results: The concentration of beta-glucan at the 3rd tertile was significantly associated with skin prick test positivity, aOR: 1.88 (95% CI: 1.17–3.03; p = 0.0097), but not with wheeze or other asthma symptoms at age 15 months, 2, 3 or 4 years. Beta-glucan levels were higher in spring compared to summer, geometric mean ratios were: 1.22 (95% CI: 1.01–1.48; p = 0.04) and 1.25 (95% CI: 1.09–1.44; p = 0.001) respectively for ng/m2 and ng/g. Beta-glucan levels expressed as ng/g were slightly higher in Christchurch than in Wellington, geometric mean ratio: 1.11 (95% CI: 0.99–1.21; p = 0.06). No significant differences in beta-glucan levels were found between living room floors with carpets/rugs and those with bare floors.

Conclusion: Early life exposure to beta-glucan may lead to allergic sensitisation in infancy.

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Prevalence of asthma and allergy in Swedish school employees 1993 and 2003

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Background: During the last decades asthma and allergy have become more prevalent, but it is not known if the increase continues. Especially among adults, data are scarce.

Methods: In 1993 we conducted a questionnaire study on prevalence of asthma and allergy among employees of 39 randomly chosen schools in the county of Uppsala, Sweden. In total 1410 (85%) of the employees answered the questionnaire. Ten years later we performed a new study in the same schools. The same questionnaire was sent to those who worked in the schools in 2003 and 1194 (67%) responded. The questionnaire included questions on self-reported allergy and the questions on asthmatic symptoms used in the European Community Respiratory Health Survey. Differences in prevalence between the two surveys were calculated by the ÷2-test.

Results: Both years, 62% of the respondents were teachers and 38% had other occupations. The proportion of women was 77% in 1993 and 81% in 2003. Mean age was 45.9 years old in 1993 and 47.7 in 2003. Prevalences of allergy and asthma are presented in Table 1.

	1993	2003	p
Furry pet allergy	9.8	13.0	0.012
Pollen allergy	16.7	19.1	0.109
Ever doctor's diagnosed asthma	6.9	8.8	0.073
Current asthma	5.2	6.9	0.073
Any asthma symptom last year	19.3	16.2	0.023
Present asthma medication	4.6	6.4	0.050

Thus, we found a tendency towards the prevalence of ever having got an asthma diagnosis, and having current asthma being slightly more common in 2003 compared to 10 years earlier. However, using asthma medication had increased and suffering from asthma symptoms had decreased. Furthermore, self-reported allergy had increased, especially towards furry pets.

Discussion: The study population was not a representative sample of the Swedish adult population, and one should be cautious when generalizing from the results. However, apart from gender, school employees should not differ substantially from the general working population. Furthermore, the two study populations were very homogenous to each other, with respect to gender, age, occupation and geographical habitat, and the study methods were identical.

Conclusion: The results imply a small increase of asthma and allergy during the 10-year period 1993–2003 in Swedish adults.

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Asthma management in real life; preliminary data from CPD-ARGA project

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Background: Despite asthma control and treatment adherence have been largely investigated in current literature, the appropriateness of general practitioner in prescribing for patients with asthma is less studied. At the beginning of a continuing professional development (CPD) program on rhinitis and asthma, part of ARGA (Allergopatie Respiratorie studio di monitoraggio linee-guida GINA e ARIA), a study supported by Italian Drug Agency (AIFA), the drugs prescription and the healthcare resource utilization of physicians adhering to the study were performed.

Materials and Methods: We observed 70.147 patients followed by 60 GP during 2006. 709 patients had diagnosis of asthma. 68 patients were hospitalized for asthma exacerbations, mean age 51 yrs. Among these patients only 16 were treated with association of beta 2 agonist/inhaled steroid coupled with antileukotrienes (9/16), antihistamines (8/16), theophylline (4/16). Systemic steroids (prednisone, betamethasone) were prescribed to 10 patients, among them, 3 received no other therapy, other 3 patients received only oral antihistamines. In 4/16 patients were prescribed tiotropium although asthma indication is not approved. The total amount of prescribed asthma drugs for these patients was low, it could last for a maximum of two months. The most prescribed drugs were beta 2 short acting bronchodilators (up to 22 drug boxes for a patient). No patient was evaluated by a specialist (allergist or pneumonologist) before hospitalization; only 6 were visited by a specialist after their discharge from the hospital and 11 underwent a spirometric evaluation. 7 patients were hospitalized twice in a year.

Conclusion: Regardless of the efforts of national and international societies for the diffusion of asthma diagnosis and treatment guidelines, asthma is still undervalued and untreated by general practitioners; this could be the principal cause of hospitalization for asthma exacerbation in the observed group of patients. It could be interesting to evaluate in a CPD blended program (5 residential plus 4 distance learning courses) in which the contents are tailored on physicians knowledge, which healthcare resource utilisation and drugs prescription would be able to improve the asthma management.

PEDIATRIC ALLERGY AND ASTHMA I

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Development and first validation of the Quality of Life in Children with Keratoconjunctivitis (QUICK) questionnaire

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Aim: The aim of our study is to develop and validate a questionnaire to evaluate Health Related Quality of Life (HRQL) in children with severe vernal keratoconjunctivitis (VKC).

Methods: An initial list of 42 items was developed and administered to 30 children with VKC in the active phase (6F, 24M; mean age 9 ± 2 years). The 30 most significant items were selected and converted into questions on a Likert scale of 3 steps for the validation phase. Validation involved 41 children with VKC in the active phase (8F, 33M; mean age 9.4 ± 2 years). Twenty-two children (5F, 17M) also completed the generic KINDL® questionnaire. Clinical signs were evaluated and scored to correlate QUICK scores to clinical findings. Total sign score (TSS) was calculated. Validation was performed by factorial analysis and Pearson's correlation. Internal consistency was calculated by Chronbach's alpha on the extracted factors.

Results: Factorial analysis extracted 2 factors with good internal consistency: Symptoms (á = 0.89), and Daily Activities (á = 0.77). Correlations of the final version of QUICK, (á = 16 items) to KINDL® scores were in the expected direction. Most patients complained of itching (93%), burning (90%), redness (90%), the need to use eye-drops (90%), tearing (83%) and photophobia (80%). The children's biggest concerns were limitations on going to the pool (71%), playing sports (58%), meeting friends (58%) or playing outdoors (42%). QUICK Symptom score was significantly correlated to conjunctival

hyperemia ($p < 0.001$), secretion ($p = 0.042$), chemosis ($p = 0.012$), superficial punctuate keratopathy ($p < 0.001$) and TSS ($p = 0.010$).
Conclusion: QUICK represents a new and simple tool to assess HRQoL in children with severe allergic conjunctivitis, useful for the global evaluation of VKC's impact on children's daily life.

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Depression, anxiety and quality of life in pediatric asthma in a Hungarian pediatric university department

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Aims: The study's objective was to examine depression, anxiety and quality of life according to age and asthma status in pediatric asthma in a pediatric university department.
Methods: 108 patients, age:11.75 ± 3.10 (mean ± SD) years; (boys 11.6 ± 2.8 years and girls 12.1 ± 3.7 years) completed the Child Depression Inventory (CDI), the State Trait Anxiety Inventory for Children (H.STAIC), the Pediatric Asthma Quality of Life Questionnaire (PAQLQ), and a symptom score. FEV1 was also measured.
Results: Mean FEV1% was 97.4 ± 12.8. 23 patients (21%) had intermittent asthma, 40 patients (37%) had mild persistent, 43 patients (40%) had moderate persistent, 2 patients (2%) had severe persistent asthma. The pediatric asthma patients scored 9.36 ± 5.57 points in the CDI. The patients showed as many depressive symptoms as the Hungarian average population, pre-adolescent boys with asthma showed even less. Children with asthma scored 31.16 ± 4.61 points on the H.STAIC questionnaire; (boys 30.64 ± 4.29, girls 32.67 ± 5.27). Children with asthma have the same anxiety level as their healthy peers. On the PAQLQ asthmatic children reached 6.18 ± 1.00 (2.87–7.00); adolescent girls scored the worst (5.62 ± 1.28). Adolescent asthmatic girls have the worst quality of life. Boys reach better quality of life scores as they grow older ($p = 0.02$). Girls with adolescence have a tendency of decreasing quality of life, although the difference is not significant. In adolescence, asthmatic girls experience more QoL deprivation than boys ($p = 0.013$). Depression score, anxiety, or quality of life showed no differences between the intermittent and persistent asthmatic groups. Children in the symptomatic subgroup experienced poorer quality of life. Depression and anxiety were not affected by current asthma symptoms. There was no significant difference in depression, anxiety or quality of life scores according to age.
Conclusion: The psychological status of the asthmatic patients is fairly good. One should concentrate more on the quality of life of girls in adolescence. The good pediatric care of childhood asthmatics helps to avoid the psychological consequences of the disease.

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Polysomnographic findings in atopic and non-atopic children with respiratory sleep disturbances

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Background: Sleep problems are often associated with allergic diseases. The aim of the study was to compare respiratory disturbances (number of apnea and hypopnea) in atopic and non-atopic children with sleep-related breathing disorders.
Methods: In the prospective study a standard polysomnography (EEG, oculogram, chin EMG, nasal and oral airflow, chest and abdominal wall movement, ECG, SaO2, body position) with parallel recording of 24-pH-metry was performed in 27 children (age-and nutrition state-matched): group I–atopic children with chronic bronchial asthma and/or allergic rhinitis (n = 11; mean age-6.9 ± 2.7ys) and group II–non-atopic children with history of sleep apnea and/or snoring (n = 16;mean age-7.3 ± 4.0ys). The Respiratory Disturbances Index (RDI) was defined as a number of sleep apnea (obstructive, central, mixed) and hypopnea per hour of sleep and children with RDI>5/h were included and sleep apnea/hypopnea syndrome was diagnosed. Plasma leptin level was measured with ELISA method.
Results: A mean total IgE and eosinophilia was significantly higher in atopic group than in non-atopic (736.2 ± 1461.1 v 34.3 ± 25.35 IU/ml and 5.05 ± 5.2% v 1.58 ± 1.2%) ($p < 0.005$). Groups were not differ in the nutrition state measured by BMI (18.7(± 5.1) v 18.5 (± 3.9) kg/m2) and by plasma leptin level (5275 ± 8007 v 6729 ± 10829) ($p > 0.05$). Significant elevated RDI was registered in both groups: 11.2(± 9.6)/h v 14.3(± 18.6)/h ($p > 0.05$). There were no differences in the number of sleep apnea in both groups.
Conclusion: In atopic children a basic disease- bronchial asthma or allergic rhinitis- may be associated with sleep apnea / hypopnea syndrome.

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Arts meets science to promote awareness of allergy in a school

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Background: Many adolescents experience allergic diseases but they represent a 'hard to reach' group for the purposes of health education. The objective of this study was work with pupils in a school on a collaborative arts-science focussing on allergy in order to heighten their awareness and knowledge of allergic disease.
Methods: The study particiapnts were fifteen 14-15 year-old pupils who were talented scientists. At the start of the project they participated in three hours of lectures and small group discussion addressing
•What do we mean by allergy? (pathophysiology)
•Who gets allergy? (epidemiology)
•How does it impact on people's lives? (clinical)
•What makes it better? (pharmacology/therapeutics)
They were then challenged with the task of developing art installations for their school that would convey the key messages they had learned about allergy to their peers.
Results: The students worked with a team of four artists (Anna dumitriu, Rachel Cohen, Carol Quinn and Caroline Doyle). They developed five installations around the school and its grounds depicting allergenic items, production of exhibition catalogue, evening private viewing attended by 50 visitors for which the pupils acted as guides and prepared food free from common allergens. The exhibition was then open to the public for the remainder of the term. Installations used a range of media including drawing, textile art, sculpture, videos and performance art.
Conclusion: This activity generated a themed exhibition within the school that promoted discussion about allergy amongst pupils, teachers, parents and the public.

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Asthma and allergy development in mono-zygotic twins discordant for severe respiratory syncytial virus infection in infancy

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Background: Children recovering from severe Respiratory Syncytial Virus (RSV) bronchiolitis are at increased risk of recurrent wheeze, asthma and possible allergy in later childhood. It is difficult to differentiate causes of disease from consequences, especially in complex genetic diseases influenced by environmental exposures.
Aim: To compare the long-term outcome of pulmonary function, sensitization and clinical diagnosis of asthma in monozygotic (MZ) twin pairs discordant for hospitalization with verified severe RSV infection in infancy.
Methods: Clinical examination was conducted on 37 MZ twin pairs (mean age 7.6 years) discordant for hospitalization with severe RSV infection in infancy. Asthma was assessed by interview, lung function (baseline spirometry for schoolchildren and specific airway resistance for preschool children), airway reactivity (responsiveness to metacholine for schoolchildren and dry air hyperventilation for preschool children), fractional exhaled nitric oxide (FeNO), and sensitization (skin testing to common inhalant allergens).
Results: 7 years (mean) after the severe RSV infection there were no differences within the discordant MZ twin pairs with respect to lung function, airway reactivity, FeNO, sensitization or clinical asthma ($p > 0.1$ for all comparisons).
Conclusion: We found no difference on the development of asthma and allergy from having or not having severe RSV infection in infancy within MZ twin pairs. This suggests that severe RSV infection is not causative in the development of asthma or allergy in childhood which is probably caused by other factors in the shared environmental and genetic factors.

AEROBIOLOGY – DETAILED CLINICAL ANALYSIS

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The novel 18 kD allergen major of Catharanthus roseus pollen: Identification, cDNA cloning, cross-reactivity and characterization as a cyclophilin

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Background: Pollen of Catharanthus roseus, a widely distributed plant in India and other sub/tropical countries, has repeatedly been reported to cause allergic reactions, especially among the gardeners. Our objective was to identify and characterize underlying allergens, in order to make them available for immunotherapy.
Methods: The major allergen was identified by western blotting of C. roseus pollen extract using sera from allergic individuals. N-terminal sequencing and screening of a cDNA library of C. roseus were done followed by expression of the recombinant protein in E. coli. Biochemical and bioinformatic analyses were also done to characterize the recombinant allergen at the molecular level.
Results: Western blotting indicated an 18 kD protein (designated Cat r 1) to be a major allergen of this pollen. Its deduced protein sequence showed considerable similarity with cyclophilin family members (Asp f 11, Mal f 6 and human cyclophilin A). The recombinant protein bound to patients' IgE and showed cyclophilin-like enzymatic activity. The structural elements of this protein were estimated by CD spectroscopy. Comparative molecular modelling suggested a considerable 3-D structural similarity with cyclophilins. Inhibition ELISA revealed cross-reactive binding of serum IgE from Cat r 1-allergic individuals to the fungal allergenic cyclophilins Asp f 11 and Mal f 6. The degree of solvent-exposure of different residues was calculated from the modelled structure and B-cell epitopes were predicted. The results support the notion that the allergenic cross-reactivity between Cat r 1 and the other cyclophilins is due to the presence of conserved patches [BBRC. 307(2):422-9] exposed on the surface of these molecules.
Conclusion: Cat r 1 is a major allergen from Catharanthus roseus pollen, indigenous in sub/tropical countries, that both on sequence and functional level belongs to the cyclophilin family. Now also available in recombinant form, Cat r 1 is a candidate molecule for immunotherapy.

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Crystal structure determination of the group 1 allergen from the dust mite blomia tropicalis

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Background: The *Blomia tropicalis* (Blo t) mite species is an important source of allergens associated with rhinitis and asthma in the tropics, where Blo t 1 has been shown to be a major allergen. The group 1 mite allergens are among the clinically most significant indoor allergens worldwide. These proteins are cysteine proteases containing a pro-peptide and a mature region, which can be found in the alimentary canal of the mites. We have previously determined the structure of Der p 1 (from *Dermatophagoides pteronyssinus*), which remains the only member of this allergen group with known structure. Der p 1 serves as a good structural model for the group 1 allergens from the closely related mite species Der f and Eur m (82/84% sequence identity, respectively) but not for Blo t 1 (33% identity). This is reflected in the very low IgE cross-reactivity between Blo t 1 and Der p 1.
Methods: proBlo t 1 was expressed recombinantly in yeast, purified by conventional methods and crystallized using the sitting drop vapour diffusion technique. Single crystals measuring 0.1 mm in the longest dimension were obtained. X-ray diffraction data were collected at Max-Lab and the ESRF. The data processing and structure determination by molecular replacement were performed using the CCP4 program package. Model building was performed in Coot.
Results: Recombinant proBlo t 1 was expressed, purified and crystallized. X-ray diffraction data were collected to a maximum resolution of 2.1 Å. Two molecules were located in the asymmetric unit by molecular replacement using mature Der p 1 as search model. The mature part of proBlo t 1 has been fitted to the resulting electron density. The elucidation of the part of the structure constituting the pro-peptide is ongoing. Only two of the three disulfide bridges found in Der p 1 are conserved in Blo t 1. Furthermore, Blo t 1 has a unique cysteine residue of unknown function.
Conclusion: Like Der p 1, the mature region of Blo t 1 forms a globular protein with two interacting domains that delimit a cleft on the surface where the active site is located. The low sequence identity between Blo t 1 and Der p 1 combined with differences in the structures of several loop regions on the surfaces of these proteins can explain the low IgE cross-reactivity. If the same is true for other Blo t allergens this could explain the differential sensitization to Blo t and Der p observed in several tropical regions.

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Aerobiological and immunological studies on Delonix regia pollen: an aeroallergen from India

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Pollen allergens are reported to be important triggers of IgE mediated allergic disorders. Delonix regia, DR or The Gulmohar tree is an important avenue tree in the tropics, planted for their ornamental value. The aim of this work was to carry out the aerobiological survey of this pollen in Calcutta and to identify their IgE binding proteins.

Seasonal periodicity of airborne DR pollen was recorded using a Burkard sampler. The relationship between the meteorological parameters and monthly pollen concentration was calculated using Spearman correlation

coefficient. The allergenic potential was studied by SPT, IgE ELISA. Protein profile of DR pollen extract was studied by reducing SDS-PAGE. The whole extract was fractioned by DEAE-Sephadex ion exchange column. Allergenicity of the fractions was confirmed by ELISA inhibition and immunoblotting.

Aerobiological survey (2004-2006) revealed the presence of DR pollen from April to July. In the peak month, May, it contributed to about 5.56 % to the total airborne pollen load. Microscopic studies showed that the pollen is 59-60µm in size, has tricolporate aperture, with reticulate ornamentation, prolate-spheroidal in shape and amphiphilous pollination. The variation of the DR pollen load showed significant positive correlation with temperature ($p < 0.05$) and wind speed ($p < 0.01$). A negative correlation was seen with relative humidity. Out of the 500 respiratory allergic patients (mean age-34.7 years) visiting the Allergy Unit, Institute of Child Health, tested with DR allergen extract, 22% showed +1 and 4.8% showed +2/+3 level of reaction in SPT. Among them 33 patients were selected for sera collection on the basis of higher level of SPT reaction and high titre of DR pollen IgE specific ELISA results. On SDS-PAGE, its whole extract resolved in to more than 25 distinct bands between 16 and 109 kD. 3 fractions were obtained by ion exchange chromatography. Fraction I had strong IgE binding reactivity. Immunoblotting with individual patient sera revealed 8 IgE binding protein bands. Among them, 6 were most important as they showed maximum binding. Protein bands of 43 and 32 kD were recognized by 54.5 and 69.7% individual patient sera respectively. Bands identified by lesser number of patients were 96, 28, 25, 23 kD.

The spread of DR tree in Calcutta has lead to a high concentration of its pollen and it contributed significantly to the aeropollen load. They need to be considered as a new potential allergenic source.

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Structure and functional characterization of recombinant house dust mite allergen Der p 2 produced from yeast Pichia pastoris

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Background: Der p 2 of the house dust mite *Dermatophagoides pteronyssynus* is a major allergen causing allergic reactions in many countries including Thailand. Currently, a number of recombinant allergens including Der p 2 have been expressed and increasingly used in various applications due to their homogeneous, high quantity, and a lower cost. Thus characterization of recombinant allergens is an important step to ensure they still retain the properties of the natural allergen.

Objectives: To determine whether determinations of the secondary structure and staining of the hydrophobic cavity of recombinant *Pichia pastoris*-expressed Der p 2.0101 could be used to predict the allergenicity of rDer p 2.

Methods: Circular Dichroism (CD) spectroscopy was performed for determining a composition of secondary structure. Hydrophobic fluorescent probe 1-anilinonaphthalene 8-sulfonic acid (ANS) was used for examining a formation of the hydrophobic cavity. Allergenicity was measured by specific IgE antibody inhibition assay.

Results: Analysis results of CD data showed that secreted rDer p 2 (rDer p 2s), purified from culture media, contained 5% α-helix, 24% β-structure and 74% random coil compared to 14% α-helix, 57% β-structure and 29% random coil of the natural Der p 2 (nDer p 2). After a denaturing step using Tri-Chloroacetic Acid (TCA) precipitation, refolded rDer p 2 (rDer p 2t) contained 7% α-helix, 54% β-structure and 38% random coil. The results from ANS-bound the hydrophobic cavity of both rDer p 2s and rDer p2t showed that, with 50 fold-molar excess of ANS, relative fluorescent intensity (RFI) of both ANS-rDer p 2s and ANS-rDer p2t were 3.1 and 4.9, respectively, compared to RFI 20 of ANS-nDer p 2. The results of inhibition of pooled serum specific IgE binding to Der p 2 confirmed that both rDer p 2s (IC_{50} 42 ng/ml) and rDer p 2t (IC_{50} 38 ng/ml) inhibited specific IgE binding with less affinity than that of nDer p 2 (IC_{50} 5 ng/ml).

Conclusion: This is the first study to report that the *P. pastoris*-expressed rDer p 2 had the different composition of α-helix and random coil, and the ANS-stained hydrophobic cavity when compared to the natural Der p 2. Allergenicity of the rDer p 2 was also decreased possibly as the results of changes in secondary structure and in the hydrophobic cavity.

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Identification of B and T cell epitopes of Cur l 3; a major cross-reactive allergen from Curvularia lunata

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Background: Structure function relationship of major allergens is required to understand molecular mechanism of allergy and to develop suitable modalities for therapy. In the present study, B and T cell epitopes of Cur l 3, a major cross-reactive allergen from fungus *Curvularia lunata* were analyzed using computational and experimental approaches.

Methods: The epitopes were predicted *in silico* and validated experimentally. Five each B cell (peptides 1-5, i.e. P 1-P5) and T cell (peptides 6–10, i.e. P6–P10) epitopes were derived based on sequence homology model. The conserved motif of peptide number 6, i.e. P6 in Cur l 3 present in the core of protein showed cryptic B cell epitope *in silico*. These peptides were chemically synthesized and assessed for their biological activity using ELISA, ELISA inhibition, dot blot, lymphoproliferation and cytokine profiling in hypersensitive patients' samples. Peptides P4, P6 and P 10 were selected for *in vivo* evaluation by skin tests.

Results: P4, P6 and P10 peptides showed higher IgG and IgE reactivity compared to other peptides. ELISA inhibition demonstrated that P1–P6 achieve 50% IgE binding inhibition at 100 ng, whereas P7–P10 could not achieve the same inhibition even at thousand fold higher concentration. In lymphoproliferation assay, peripheral blood mononuclear cells from *C. lunata* patients stimulated with P6–P10 peptides showed at least two-fold proliferation than controls. The levels of IL-4 and IL-5 were elevated in culture supernatants demonstrating Th 2 type of immune response. Among these peptides, P6 showed marked positive skin reactivity in 35/37 Cur l 3 positive patients followed by P4 (30/37) and P10 (10/37).

Conclusion: The B and T cell epitopes spanning the entire region of Cur l 3 were mapped *in silico* and validated experimentally. A cryptic presence of an IgE interacting region was found in predicted T cell epitope i.e. P6. The knowledge regarding T and B cell epitopes of this major allergen can help in developing hypoallergenic variants using mutational strategy.

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Molecular cloning and immunochemical characterization of a new Japanese cedar pollen allergen homologous to aspartyl protease

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Background: Among allergenic pollens, Japanese cedar (*Cryptomeria japonica*) pollen represents the most important aeroallergens in Japan. It

elicits rhinitis and conjunctivitis especially in younger generations. The two-dimensional IgE-binding spectrum of *C. japonica* pollen allergens demonstrated that many allergens remain to be identified. Here we present the molecular cloning and immunochemical characterization of a novel *C. japonica* pollen allergen belonging to aspartyl protease family.

Methods: TOF-MS analysis of a high IgE-binding protein, termed CPA63, revealed its internal amino acid sequences. Based on these sequence information, cDNA-encoding *CPA63* was cloned by RACE-PCR. The allergen was produced as a recombinant protein using baculovirus-insect cell culture system, and purified by double chromatographic technique using HisTrap and HiTrap Q columns. Putative mature recombinant CPA63 (r-CPA63) was produced upon autolysis by incubation in acetate buffer (pH 3.3), and used for ELISA experiment. Its proteolytic activity was tested using FITC-casein as a substrate at different pHs, and substrate specificity was evaluated by using series of protease inhibitors.

Results: *CPA63* cDNA encoded a 472 amino acid polypeptide with calculated molecular weight and isoelectric point of 51.1 kDa and 4.69, respectively. Homology search revealed that CPA63 polypeptide sequence showed about 40% identity with plant aspartyl protease/nucleoid DNA binding protein family members. ELISA demonstrated that purified r-CPA63 was recognized by pollinosis patient IgE at a frequency of 58% (18/31). The r-CPA63 also showed an aspartyl protease-like proteolytic activity, demonstrating its enzymatic maturation upon autolysis.

Conclusion: CPA63 is the first plant aspartyl protease identified as an allergen. That might well open new investigations for other plant aspartyl protease allergens. The availability of CPA63 sequence and recombinant allergen production could be useful to develop future diagnostic technique and therapeutic approaches.

URTICARIA AND ANGIOEDEMA

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Coagulation factor XII (Hageman factor): a new angioedema gene

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Background: Hereditary angioedema (HAE) is characterized by recurrent skin swelling, abdominal pain attacks, and potentially life-threatening upper airway obstruction. The classic HAE types (I and II) are caused by mutations in the complement C1 inhibitor gene, resulting in a quantitatve or qualitative deficiency of C1 inhibitor. In contrast, in a novel HAE type, affecting mainly women, C1 inhibitor concentration and activity in plasma are normal (HAE type III, HAE with normal C1 inhibitor); we hypothesized that an abnormal coagulation factor XII molecule may lead to inappropriate activation of the kinin-forming cascade and, therefore, performed a search for mutations in the F12 gene.

Methods: Twenty unrelated index patients from families with hereditary angioedema and normal C1 inhibitor activity were screened for mutations in the coagulation factor XII gene by sequencing of the 14 exons and splice junctions of the F12 gene; subsequently, beside 235 control individuals, another six such index patients as well as 90 patients with idiopathic angioedema were selectively sequenced for exon 9 of the F12 gene.

Results: Two different non-conservative missense mutations, both located in exactly the same position within exon 9, namely in the second position of the codon (ACG) encoding Thr309 of the mature protein, were identified. Five of the twenty patients screened showed a heterozygous C→A transversion (1032C→A), predicting a threonine-to-lysine substitution (Thr309Lys); one additional patient showed a heterozygous C→G transversion (1032C→G), resulting in a threonine-to-arginine substitution (Thr309Arg). Sequencing of exon 9 in another six index patients revealed one further patient heterozygous for the Thr309Lys mutation. Thus, in 7 of 26 unrelated patients the wild-type threonine is substituted by a basic amino acid residue. The mutations were not found in healthy control individuals (n = 235) and co-segregated with the phenotype in seven families with altogether 23 affected women, providing strong support that they cause disease. Finally, the Thr309Lys mutation was also identified in 2 out of 90 patients with idiopathic angioedema; thus, this mutation may also play a role in a subgroup of these singular angioedema cases with no affected family members.

Conclusion: These findings provide strong support that the coagulation factor XII gene is a new angioedema gene.

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Subcutaneous DX-88 (ecallantide) in the treatment of acute attacks of hereditary angioedema: interim results of EDEMA3®, an open-label, phase 3 trial

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Background: Disinhibition of kallikrein results in excess of bradykinin and is responsible for signs and symptoms of hereditary angioedema (HAE). The highly potent, specific plasma kallikrein inhibitor, DX-88 (ecallantide) has been shown to ameliorate symptoms of HAE.

Methods: Patients 10 years of age or older with documented HAE (12 naïve patients and 48 patients who were treated for a single acute HAE attack in the double-blind, placebo-controlled stage of EDEMA3) participated in this open-label extension. Patients were given 30 mg DX-88 to treat acute HAE attacks at all anatomic site locations- abdominal, peripheral, and laryngeal. Outcome measures at 4 and 24 hours were the HAE-specific Treatment Outcome Score (TOS) and change from baseline in Mean Symptom Complex Severity (MSCS) score. TOS and MSCS are composite scores measuring all attack sites. Time to onset of response and time to significant improvement in overall response were recorded. Safety was assessed by treatment-emergent adverse events (TEAEs) and serious adverse events (SAEs).

Results: Interim data are presented for 59 patients treated for 145 acute HAE attacks (20 men, 39 women, median age 34.0 years, range 12–77 years). Mean TOS was 75.3 ± 39.64 at 4 hours and 81.8 ± 36.55 at 24 hours (≥ 50= improvement). Mean MSCS change from baseline was −2.3 ± 0.6 at both 4 and 24 hours (−1 to −3 = clinically-significant improvement). Median time to onset of overall improvement was 37.5 minutes, and time to onset of significant overall improvement was 165.0 minutes. Safety data for 51 patients included 166 TEAEs (87% mild or moderate in severity and 89% resolved without sequelae), with 30 related TEAEs in 15 patients. Related events experienced by more than 1 patient include diarrhea, nausea, injection site reaction (3 patients each), HAE, myalgia, headache, cough, and pruritus (2 patients each). A total of 7 SAEs occurred in 7 patients; 6 were unrelated to DX-88, and 1 SAE of anaphylactic reaction reported previously, was assessed as related, and resolved without sequelae.

Conclusion: Repeat dosing of DX-88 (ecallantide) for acute HAE attacks resulted in onset of overall improvement in <40 minutes and significant overall improvement in <3 hours. Ecallantide was generally well tolerated with the majority of adverse events being mild or moderate in severity and resolved without sequelae.

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Clinical symptoms and treatment in hereditary angioedema with normal C1 inhibitor

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Background: A new type of hereditary angioedema was described recently. It was characterized by recurrent bouts of angioedema in various organs and normal C1 inhibitor and was observed mainly in women. Our aim was to conduct a detailed study of the clinical features of this condition.
Methods: A total of 138 patients with hereditary angioedema and normal C1 inhibitor who belonged to 43 unrelated families were examined through the use of standardized questionnaires.
Results: A majority of patients with hereditary angioedema and normal C1 inhibitor had skin swellings (92.8%), tongue swellings (53.6%), and abdominal pain attacks (50%). Laryngeal edema (25.4%) and uvular edema (21.7%) also were frequent, whereas edema episodes of other organs were rare (3.6%). Facial swellings and tongue involvement occurred considerably more frequently compared with hereditary angioedema due to C1 inhibitor deficiency. The number of patients with recurrent edema of only one organ was higher than in classic hereditary angioedema. The number of patients with disease onset in adulthood was significantly higher in hereditary angioedema with normal C1 inhibitor compared with classic hereditary angioedema. Erythema marginatum was not observed. A subgroup of patients from families with coagulation factor XII mutations showed the same symptoms as the other patients. Long-term treatment consisted in tranexamic acid and attenuated androgens.
Conclusion: Hereditary angioedema with normal C1 inhibitor levels shows a characteristic pattern of clinical symptoms. The main clinical features include skin swellings, tongue swellings, and abdominal pain attacks. There are many differences in the clinical symptoms and course of disease between this type of hereditary angioedema and classic hereditary angioedema due to a genetic C1 inhibitor deficiency.

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Skin reactivity to autologous serum and safety of COX-2 inhibitors in NSAID-intolerant patients with urticaria and/or angioedema

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Background: Pseudoallergic reactions (PAR) to non-steroidal antiinflammatory drugs (NSAID) are very common in chronic urticaria (CU), as aspirin and related drugs impair symptoms in about 20–30 % of patients. Moreover, a group of otherwise normal subjects who develop urticaria and/or angioedema (UA) only after the intake of NSAIDs also exists. Although the inhibition of cyclo-oxigenase (COX) pathway seems to play a central role in NSAID-intolerance, a very high prevalence of skin reactivity to autologous serum (AS) has also been reported in NSAID-induced UA, pointing to a possible involvement of the immune system in the pathogenesis of these forms.
Methods: 50 patients who had experienced NSAID-induced UA were recruited and gave their informed consent. 28 had a history of CU exacerbated by NSAIDs, whereas the remaining 22 reported acute UA after NSAIDs intake. In 26 cases, UA had been elicited by a single NSAID, whilst 24 patients signalled adverse skin reactions to two or more COX-inhibitors. None of the patients presented nasal polyps, nor had evidence of asthma or sinusitis. Oral tolerance tests were conducted on different days by single-blind, placebo controlled challenge with increasing amounts of both conventional (nimesulide, 100 mg whole dose; meloxicam, 15 mg), and highly selective (celecoxib, 400 mg; eterocoxib, 90 mg) COX-inhibitors. The test was continued until the whole dose was reached, or UA appeared. Furthermore, skin reactivity to AS was assessed in all subjects in basal conditions, as well as whenever challenge-induced adverse reactions occurred.
Results: No patients developed skin symptoms on the placebo day, and 28 tolerated each of the four COX-inhibitors. 16 experienced UA with nimesulide, 8 with meloxicam, 4 with celecoxib and 2 with eterocoxib. Only 4 patients with CU showed a positive cutaneous response to AS, whilst in the other subjects skin tests with both basal serum and samples collected during challenge-induced UA always elicited negative results.
Conclusion: Our findings confirm that highly selective COX-inhibitors represent the safest anti-inflammatory drugs in NSAID-induced UA, although it is well known that coxib intake is not recommended in subjects with renal or cardiovascular diseases. Moreover, these results strengthen the concept that cutaneous reactivity to AS is almost exclusively confined to chronic urticaria, and that autoimmunity is not involved in the pathogenesis of NSAID-induced PAR.

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Treatment of skin swellings with pasteurized C1 inhibitor concentrate in patients with hereditary angioedema

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Background: C1 inhibitor concentrate is highly effective in treating laryngeal edema and abdominal pain attacks in patients with hereditary angioedema due to C1 inhibitor deficiency. Aim was to assess the efficacy and safety of a pasteurized human C1 inhibitor concentrate in skin swellings of hereditary angioedema.
Methods: Between 1976 and 2007, a total of 2,104 skin swellings in 47 patients were treated with 500 (1704 attacks) or 1,000 units (400 attacks) of the C1 inhibitor concentrate. The time to relief, the duration of the swelling, and the severity of symptoms were documented during personal interviews using standardized questionnaires and compared to 9.144 untreated skin swellings in the same patients.
Results: The 1,912 treated skin swellings affected face (483 attacks), hands and arms (831 swellings), feet and legs (638 swellings), genitals (123 swellings), and the trunk (29 swellings). The mean time to relief of symptoms was 1.3 hours (SD 1.4 hours) in all treated skin swellings versus 59.8 hours (SD 27.1 hours) in all untreated swellings. The mean duration of the swellings was shortened from 85.1 hours (SD 41.2 hours) (untreated swellings) to 39.3 hours (SD 30.2 hours) (treated swellings). All patients responded to treatment. In 24 skin swellings of 4 patients the course of the treated swellings was not shortened but symptoms were milder compared to untreated skin swellings. C1 inhibitor concentrate was more effective when injected early in the attacks compared to late injections. There were no drug-related side effects.
Conclusion: The pasteurized C1 inhibitor concentrate is highly effective and safe in treating skin swellings in patients with hereditary angioedema.

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Association of TNF-α promoter polymorphisms with aspirin-induced urticaria

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Background: Although the pathogenesis of aspirin-induced urticaria(AIU) has not been fully understood, mast cell activation was noted in patients with aspirin hypersentivity. Tumor necrosis factor(TNF)-α, a potent pro-inflammatory cytokine is released by human skin mast cells and other inflammatory cells, which was detected on the skin lesion of chronic urticaria. To investigate the role of TNF-α promoter polymorphisms in the development of AIU, we performed association study of the TNF-α promoter polymorphisms with AIU phenotype.

Methods: 239 patients with AIU and 524 normal controls were enrolled from the Ajou University Hospital in Suwon, Korea. 120 patients of the AIU had underlying chronic urticaria with more than 6 weeks duration (AICU). AIU was confirmed by oral aspirin challenge test. Five SNPs in the TNF-α gene (-1031T>C,-863C>A,-857C>T,-308G>A,-238G>A) were genotyped by a single base extension method. Haplotype analyses were done.
Results: The genotype frequencies of TNF-1031T>C and TNF-863C>A were significantly higher in the AIU patients than in the normal controls in co-dominant (p = 0.014, p = 0.007) and dominant (p = 0.007, p = 0.004) models, respectively. The frequency of TNF-ht2[CACGG] was significantly higher in the AIU patients (20.9%) than in the normal controls (14.5%, p = 0.004).
Conclusion: These findings suggest that the TNF-α promoter polymorphisms (TNF-1031T>C and TNF-863C>A) may contribute to the development of AIU.

NEW INSIGHTS IN OCCUPATIONAL ALLERGY AND ASTHMA

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Molecular and immunological characterization of a wheat serine proteinase-inhibitor as a novel allergen in baker's asthma

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Backround: Immunoglobulin E-mediated sensitization to wheat flour belongs to the most frequent causes of occupational asthma.
Methods: In order to identify allergens involved in baker's asthma we have constructed a cDNA library from wheat seeds and screened the library with serum IgE from patients with baker's asthma. The recombinant allergen was expressed in *Escherichia coli* and purified to homogeneity.
Results: One phage clone specifically recognized by baker's asthma patients contained a full length cDNA coding for an allergen with a molecular weight of 9.9 kDa and a pI of 6 which according to sequence analysis could be identified as a member of the potato inhibitor I family, a group of serine proteinase inhibitors conserved throughout the plant and animal kingdom. According to circular dichroism analysis it represented a soluble and folded protein containing mainly β-sheets, random coils and an alpha helical element and high thermal stability. The recombinant allergen showed allergenic activity and reacted specifically with IgE from bakers' asthma patients but not with IgE from grass pollen allergic patients or patients suffering from food allergy to wheat. Using allergen-specific rabbit antibodies it is demonstrated that the protein is mainly expressed in mature wheat seeds and, despite an overall degree of approximately 50% sequence identity, showed no relevant cross-reactivity with plant food sources such as maize, rice, bean or potato.
Conclusion: Recombinant wheat serine proteinase inhibitor may be useful for the diagnosis and therapy of IgE-mediated baker's asthma.

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Gum arabic as a cause of occupational allergy and other allergic symptoms

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Background: Gum arabic is used as an emulsifier, a thickening agent and as stabilizer in foods, in pharmaceutical industry, lithography and cosmetics. Exposure to gum arabic has been reported to cause occasional cases of occupational asthma, but ingestion of it does not commonly cause immediate allergic symptoms.
Methods: In 2005 - 2006 11 workers of a candy factory were examined for suspected occupational allergy. Hard boiled candies were covered with spray dried gum arabic in rotating drums, where the workers poured the gum arabic powder. 7/11 had respiratory and skin symptoms and 4 had skin symptoms. Spirometry, histamine challenge test, exhaled breath NO and PEF measurements, skin prick tests (SPT), gum arabic IgE measurements and cutaneous or bronchial challenge tests with gum arabic were carried out. Patch tests were carried out to a patient with eczematous skin disease. In addition, SPT data from 2997 subjects examined for food allergy symptoms from 1998 to 2006 were evaluated. Subjects with positive SPT to gum arabic received a postal questionnaire.
Results: Four workers had occupational asthma caused by gum arabic. All of them had a positive SPT with gum arabic and specific IgE to gum arabic (0.60-5.6 kU/l). Three patients had a positive bronchial challenge test with gum arabic and one of them had a positive skin challenge test. One patient had significant PEF decrease when exposed to gum arabic at work, a positive skin challenge and immediate oral symptoms associated with ingested gum arabic. None of them had other significant allergies. One worker had contact dermatitis caused by thiuram chemicals and another with positive SPT to house dust mites had work related allergy to carmine red used in candies. Five workers had no occupational disease. Of patients tested for food allergies 17/2997 (0,005%) had positive SPT to gum arabic and most of them also had positive SPT to pollen and foods. 11/17 (65%) returned the questionnaire. At the time of skin testing gum arabic associated allergic symptoms were suspected in three, and two reported gum arabic ingestion associated oral symptoms in the questionnaire. One gum arabic SPT positive patient without respiratory symptoms had an occupational history of work in a candy factory for years ago.
Conclusion: Occupational allergy caused by IgE mediated allergy to gum arabic is a risk in exposure occupations. Ingested gum arabic causes symptoms in few patients.

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Determinants of elevated exhaled nitric oxide (eNO) among bakery workers in South Africa

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Background: Measurement of the concentration of NO in exhaled air (eNO) is a useful non-invasive method of assessing inflammatory airway disease. This has a potential widespread utility as an early marker for occupational allergy and asthma in the surveillance of workers exposed to respiratory sensitisers. The aim of this study was to determine the predictors of elevated exhaled nitric oxide and their correlation with ocular-nasal and asthma symptoms among bakery workers.
Methods: A cross-sectional study of 417 bakers was conducted in 31 supermarket bakeries. Baseline demographic and occupational data as well as work-related respiratory symptoms were collected using an interviewer-based questionnaire. A hand-held portable nitric oxide sampling device (NIOX MINO® Airway Inflammation Monitor; Aerocrine AB, Solna, Sweden) was used to determine eNO concentrations, during the workshift, according to ATS/ERS recommendations (2005).
Results: The mean age of the bakers was 33 years and 44% were current smokers. The prevalence of recent chest infections (past 3 weeks) was 35%, whilst 16% reported wheezing and 29% ocular-nasal symptoms in the past 12 months. The prevalence of work-related symptoms was higher for ocular-nasal (39%) than chest (15%) symptoms. The mean eNO was 25ppb, with 19% demonstrating moderately increased (21–35ppb) and 17% high (>35ppb) eNO

levels. In the univariate analysis, age (OR: 0.97), male gender (OR: 1.80), current cigarette smokers (OR:0.54), exercise (OR:2.56) and recent chest infections (OR:2.29) were significantly ($p<0.05$) associated with high (>35ppm) eNO levels. In the multivariate analysis, after adjusting for potential confounders (age, gender, smoking, recent chest infections), workers with high eNO levels (>35ppb), were twice as likely to report wheezing (OR: 2.54) or ocular-nasal symptoms (OR: 1.98) in the past 12 months and three times (OR:3.45) more likely to report work-related ocular-nasal symptoms.

Conclusion: This is the first study to demonstrate increased eNO levels among supermarket bakery workers. Markedly elevated eNO (>35ppb) could be used as a marker for work-related ocular-nasal symptoms and recent wheeze. Its utility in identifying bakery workers with work-related wheeze needs further investigation.

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An investigation of respiratory hypersensitivity and work related allergens on pig slaughters in Taiwan

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The prevalence of respiratory sensitization among 213 pig slaughters in Taiwan was investigated through questionnaire and health examination. Following the allergic syndromes, lung function, MAST test and ELISA, a work related hypersensitivity was suggested.

It showed about forty percent of the investigated workers contact pig exudates, over four hours long in their workdays. Almost all of the workers were not used to wear masks. About 26% of the workers exhibited bad function of lung and most of them exhibited the restriction type. Among the 35.7% workers having allergic problems, most of that were related to their working places. Possible allergens were postulated to be pig exudates sources mainly. From the results of MAST of eighty workers, the respiratory hypersensitivity are statistically significant with the positive reacted IgE against Candida albicans, cat dander, especially in some special work kinds or areas, such as: main bone cutting, body translocation, bleeding out.

Among the serum samples of sensitized workers, 56% were positively against protein components of pig plasma, and hundred percent to components of pig saliva. Workers who claimed being allergic showed high positive rate to IgE titers specific to animal exudates, otherwise, those workers also exhibited holiday recovery effects mostly.

In SDS-gel electrophoresis, Special components may be evaluated as antibody biomonitoring factor on exposure assessment, such as components with MW 55K, 37K, and 32K Da, that were significantly different between exposed workers and control group.

Two-dimension acrylamide gel electrophoresis was performanced, using immunoblot analysis, specific IgE binding components in pig saliva were found. We postulated that might be animal specific allergens. After excising the reactive spots, we use mass spectrometry to identify those allergenic components in pig saliva.

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Evaluation of sensitization in a population of professional catering students

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Background: The intention of our study is to evaluate the incidence of allergic problems in a population of catering students at the beginning and at the end of their course, with the aim of defining certain clinical parameters which may be considered as risk or prognostic factors for the future emergence of an occupational allergy.

Methods: We enrolled in the study 601 students of a professional catering school: 412 in the first year (14/15 years old) and 189 in the fifth years (18/19 years old). We used a questionnaire for respiratory problems and another for dermatological ones. 424 subjects were also patch tested with 10 aptens. 26 students were retested 2 years later (16/17 years old) their first patch control.

Results: The incidence of reported symptoms of allergy was higher in fifth year than in first one. We noticed an increase of referred contact urticaria reaction due to food contact in fifth year students in comparison to the first year ones. We could observe a rising in positive reaction between first year students and fifth year students to Balsam Perù, Nickel, Fragrance mix and Garlic. In particular we found an higher incidence of positive patch reaction in female population. On the contrary the rising in positive reaction to Garlic is higher in male. About two third of the subjects positive to Garlic present a sensitization to Diallyl Disulphide. The data related to the group of students retested after 2 years reveal an higher incidence of positive patch tests reactions ($p < 0.05$), especially for Garlic ($p < 0.05$).

Conclusion: The rising of positive patch test reactions to Garlic and Balsam Perù should be caused by the contact with cooking spices. We hypothesize that the increase in positive reactions to garlic in the male group could depend on manipulation of garlic in the trainee period; females do not show similar results, perhaps due to a precedent of cooking activity at home. The higher incidence of allergic pathology between students during fifth year of school and students at first year was probably due to scholastic practical activity. To understand if other garlic antigen are involved in sensitization, we want to TLC patch test who were positive to garlic and negative to Diallyl Disulphide. The study will continue following the first year students, repeating the tests in the third and the fifth year of school in order to highlight the variations in positive skin reactions to allergens handled during cooking activities.

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Genetic polymorphisms of ADRB2 and IL10 can be associated with the risk of IgE sensitization to digestive drug powders in exposed medical personnel

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Objective: It has been reported that digestive drug powders could induce IgE mediated respiratory allergy in the medical personnel. The aim of this study was to evaluate genetic susceptibility factors associated with IgE sensitization to digestive drug powders in medical personnel.

Methods: We performed a case-control study involving 38 subjects with digestive allergy, 115 asymptomatic exposed subjects and 123 non-exposed controls working University Hospitals. Digestive allergy was determined by results of serum specific IgE measured by ELISA and skin prick tests with 5 commonly exposed drugs. 10 single nucleotide polymorphism (SNP)s: beta2 adrenergic receptor (ADRB2 46A>G), high-affinity IgE receptor alpha chain (FCERIA-344C > T, FCERIA-95T > C, FCER1B-109T > C, FCER1B 237A > G), transforming growth factor beta1 (TGFb1-509C > T), interleukin 10 (IL10-1082A > G), cysteinyl leukotriene receptor type 1 (CYSLTR1-634C > T), cysteinyl leukotriene receptor type2 (CYSLTR2-2079C > T, CYSLTR2-2534A > G) were genotyped using a single base extension method.

Results: Among the 10 SNPs, we found two distinct significant associations with digestive allergy in exposed medical personnel; the AA genotype of ADRB2 46 A > G ($p = 0.016$, OR = 3.045, CI: 1.229–7.554), and the AG and GG genotype of IL10 -1082 A > G ($p = 0.008$, OR = 5.081, CI: 1.517–17.025, by multiple logistic regression analysis controlling for age, atopy and workplace).

Conclusion: These results suggest that genetic polymorphisms in the ADRB2 46A > G and IL10 -1082 A > G may be genetic factors to increase IgE sensitization to digestive drugs in medical personnel who can be exposed to drug powders occupationally.

SUBLINGUAL IMMUNOTHERAPY–CLINICAL

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Specific immunotherapy for allergic respiratory diseases: evidence based evaluation

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Background: According to Evidence based medicine (EBM), conclusions from meta-analyses of randomized and controlled trials represent the most solid source to assess the efficacy of a treatment. Meta-analysis is a statistical procedure that incorporates the results of different independent studies pooled together, provides a quantitative estimate of treatment effects, and may explain and quantify the heterogeneity between results of individual studies.

Objective: To evaluate the efficacy of allergen specific immunotherapy (SIT) in the treatment of allergic asthma and rhinitis by means of available meta-analyses.

Methods: Electronic databases were searched up to January 31st, 2007, for meta-analysis of randomized, double-blind and placebo-controlled trials assessing specific immunotherapy in respiratory allergy. We looked for studies evaluating the effects on symptom scores and use of rescue medication, using standardized mean differences (SMD) and the random-effect model. The individual meta-analysis quality was evaluated following the Cochrane group and the QUORUM statement recommendations.

Results: Six meta analyses met the inclusion criteria. Four studies evaluated sublingual (SLIT) and two subcutaneous (SCIT) immunotherapy. All of them reported a reduction in both symptom and medication scores; but one study did not find a significant size effect because of studies' inclusion criteria. SMD for symptoms score and rescue medication are shown in table. Heterogeneity was significant in all five included reviews. Studies about SCIT in both allergic rhinitis and asthma are in progress, but results are not available at the moment.

Conclusion: The final conclusion from the present review is a general acceptance of Allergen Specific Immunotherapy efficacy using the criteria of Evidence Based Medicine. Nonetheless, we should look forward, the GRADE

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Meta-analysis	Disease	SIT	Number of patients	Symptoms score SMD (95% CI)	Rescue medication score SMD (95% CI)
Abramson MJ 2003	asthma	SCIT	1064	−0.72 (−0.99 a −0.44), p <0.001	−0.80 (−1.13 to −0.48), p<0.001
Calamita Z 2006	asthma	SLIT	303	−0.38 (−0.79 to 0.03), p = 0.07	−0.82 (−1.25 to −0.39), p=0.0002
Canonica GW 2007	asthma	SLIT	412	−1.42 (−2.25 to − 0.59), p = 0.02	−1.63 (−2.83 to 0.44), p=0.007
Calderon M 2007	rhinitis	SCIT	1063	−0.73 (−0.97 to −0.50), p < 0.001	−0.57 (−0.82 to −0.33), p<0.001
Penagos M 2006	rhinitis	SLIT	484	−0.56 (−1.01 to −0.10), p = 0.02	−0.76 (−1.46 to −0.06), p=0.03
Wilson DR 2005	rhinitis	SLIT	979	−0.42 (−0.69 to −0.15), p = 0.002	−0.43 (−0.63 to −0.23), p=0.0003

evaluation, taking in consideration efficacy but also safety and costs, will probably support even more SLIT as the best option. Specific studies are presently ongoing to apply this new evaluating system to SIT. Finally, the recent SIT and SLIT studies, with a relevant number of patients, further support the conclusions obtained through the meta- analysis evaluation.

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Efficacy and safety of grass allergen tablets in patients with grass pollen rhinoconjunctivitis of differing severity profiles

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Background: The efficacy, safety and optimal dose of grass pollen tablets for sublingual immunotherapy (SLIT) in allergic rhinoconjunctivitis patients were investigated in a multinational, randomised, double-blind, placebo-controlled study in 628 adults. We present the results of further analyses in subgroups of the original study population.

Methods: Patients were randomised to receive once-daily five-grass-pollen extract sublingual tablets (Stallergènes, Antony, France) of 100 IR (n = 157), 300 IR (n = 155) or 500 IR (n = 160), or placebo (n = 156) for 6 months. Four sensitivity subgroups were identified. Group 1: Patients with a baseline immunoglobulin E (IgE) >17.5 kU/L, Group 2: patients with a Retrospective Rhinoconjunctivitis Total Symptom Score (RRTSS) >15 (the third quartile); Group 3: Patients with a baseline skin prick test (SPT) wheal diameter >10.5 mm (the third quartile); Group 4: patients with at least one of the above characteristics. An exploratory analysis was performed on the primary efficacy outcome, the effect of SLIT on the Rhinoconjunctivitis Total Symptom Score (RTSS), and adverse event (AE) data.

Results: Across the four subgroups average RTSS scores were Group 1: 3.91 [SD: 3.157] to 4.86 [SD: 3.252], Group 2: 3.83 [SD: 3.135] to 5.34 [SD: 3.049], Group 3: 2.55 [SD: 2.127] to 5.56 [SD: 3.066] and Group 4: 3.61 [SD: 2.967] to 4.94 [SD: 3.188]). Group 3 patients receiving 100 IR differed from all other treatment groups because rhinorrhoea, rather than sneezing, achieved the highest average score.

The analysis of covariance (ANCOVA) showed that in Group 1 patients the average RTSS did not differ significantly with different doses of SLIT (p = 0.2032). In Groups 2, 3 and 4, doses of 300 IR and 500 IR were more effective than 100 IR and placebo (p < 0.0346).

The relative safety profiles of the four treatments were similar across all four subgroups. All doses of SLIT administered in this study can be considered safe in the patients investigated.

Conclusion: Regardless of severity profile or sensitisation status (high specific IgE, skin sensitisation or severe clinical symptoms), both 300IR and 500IR demonstrated a significantly improved RTSS, compared with placebo. In agreement with previously reported data, the risk-benefit ratio validates the use of 300IR tablets for clinical practice in all these patient sub-groups.

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Sublingual immunotherapy with grass pollen tablets demonstrated clinical efficacy in patients with rhinoconjunctivitis from the first pollen season, at peak season and throughout the season

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Background: The optimal dose (300IR) of a 5 grass-pollen sublingual immunotherapy tablet was previously demonstrated in terms of efficacy and quality of life from the first pollen season. Here we aim to confirm whether this dose remained optimal during the peak of the pollen season by assessing efficacy and quality of life data.

Methods: 628 patients with grass pollen rhinoconjunctivitis were randomised in a double-blind, placebo-controlled, multi-centre, pan-European trial. Patients received once-daily tablets (Stallergenes, Antony, France) of 100IR, 300IR, 500IR or placebo, starting 4 months before and throughout the 2005 grass pollen season. The pollen season was defined as the first day of three consecutive days with a grass pollen count above 30 grains/m^3 of air, recorded using Burkard-type volumetric pollen traps, to the last day before three consecutive days with a pollen count below 30 grains/m^3.

Results: The grass pollen season lasted an average of 30 days, with the peak of the season lasting for 12 days. Of the 42 sites, the Czech sites had the longest pollen season (22 to 44 days) and the Hungarian sites had the shortest season (8 to 36 days). The mean treatment duration before the grass pollen season was similar in the four treatment groups (121.4±31.1 to 128.6±15.4 days in the safety population). Both the 300IR and 500IR groups had highly significant improvements in Rhinoconjunctivitis Total Symptom Score (RTSS) versus placebo at the peak pollen season (p = 0.0005 and p = 0.0014 respectively), which agreed with improvements in RTSS in the primary efficacy evaluations. The average RTSS scores were slightly elevated during the peak pollen season in all treatment groups. Comparisons of efficacy, using only the 10 pooled sites with the highest pollen counts, also showed statistically significant improvements in the 300IR and 500IR groups versus placebo, but no statistically significant difference between the 100IR group and placebo. The overall RQLQ score was highly significantly improved in the 300IR and 500IR groups versus the placebo group at peak (p < 0.0001) and at the end (p > 0.0031) of the pollen season. All doses were well tolerated.

Conclusion: At the peak pollen season, efficacy and quality of life data for both 300 IR and 500 IR groups was significantly improved versus the placebo group. These results confirm the conclusions of the primary evaluations and validate the use of 300IR tablets for clinical practice.

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SQ-standardised grass (phleum pratense) allergen tablet improves disease specific quality of life in adult patients with grass pollen induced rhinoconjunctivitis over 2 years of continuous treatment

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Background: Grass pollen allergy is one of the commonest inhalant allergies leading to impaired quality of life (QoL) and increased health care expenditure, with as many as 50% of respiratory allergic patients being allergic to grass pollen. 70% of allergic patients feel that allergy limits their QoL. Specific immunotherapy (SIT) is currently the only treatment modality able to change the underlying mechanisms of allergic disease and thereby prevent allergic symptoms.
Methods: Disease specific QoL (Juniper's Rhinoconjunctivitis Quality of Life Questionnaire (RQLQ)) data from a randomised, double-blind, placebo controlled, multi-centre trial, including patients with significant rhinoconjunctivitis due to grass pollen, were analysed. Patients were followed for a 2 year period, where they were randomised to receive the SQ-standardised grass allergen tablet (SQsGAT), (ALK-Abelló A/S), or placebo. Both groups had access to "as needed" symptomatic treatments for their rhinoconjunctivitis. Second year data on average weekly RQLQ score was available for 351 patients, SQsGAT (n = 189) and placebo (n = 162).
Results: Overall mean RQLQ score was significantly better in the SQsGAT treated patients compared to placebo (33%, p<0.0001). Analysis of the individual domains within the RQLQ questionnaire showed that the SQsGAT improved all 7 domains compared to placebo.

Domain	Placebo*	SQsGAT*	Difference*	% Diff.
Activities	1.44	1.04	0.40	27.8%
Sleep	0.62	0.20	0.42	67.7%
Non-nose / eye symptoms	0.71	0.32	0.39	54.9%
Practical problems	1.43	0.87	0.56	39.2%
Nasal problems	1.42	1.03	0.39	27.5%
Eye symptoms	1.21	0.61	0.60	49.6%
Emotional	0.61	0.19	0.42	68.9%

*Median value

Conclusion: These results demonstrate that in adult patients with grass pollen induced rhinoconjunctivitis, the SQ-standardised grass *(phleum pratense)* allergen tablet after 2 years of continuous treatment offers improvements in the overall RQLQ score and in all 7 individual RQLQ domains compared to placebo. Resulting in clinically relevant improvements in disease specific QoL compared to placebo.

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The efficacy of coseasonal SLIT over three consecutive seasons is maintained after cessation of treatment in patients with grass pollen allergy

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Background: The data for the carry-over effect of SLIT treatment are still scarce. We therefore evaluated the efficacy, carry-over effect and safety of SLIT in a patient- friendly, seasonal treatment concept.
Methods: A baseline season was followed by seasonal treatment in 3 consecutive years and 1 follow-up season without SLIT. At the beginning of each season, treatment started by a 5-grasses mixture with titration under medical observation within 90 minutes (0-30-90-150-300 IR) followed by a daily intake of 300 IR for the whole season. Symptoms, medication intake and adverse events (AE) were documented daily in a diary for each pollen season of the 5-year observation period. Specific IgE and IgG4 were determined by CAP® and micro-array technique which also allowed possible cross reactivity to other proteins sich as food allergens.
Results: 213 patients (99m, 114f, mean age 32.2 years) with AR against grass pollens (mean disease duration 12.8 years) were randomized. Data from 184 patients were analyzed after the follow-up period. Mean treatment duration varied between seasons (81.8–92.7 days). The combined score (symptoms and medication) improved in the treatment seasons continuously up to 43.4% compared to baseline in the verum group (V) and fluctuated between +8.9 to −8.5% in the placebo group (P). Similar changes were observed for the symptoms score with a consecutive decrease of 39.0% (V), and fluctuations between +7.0 and +17.8% in P. All intergroup differences were significant in all seasons ($p < 0.05$). The improvement for verum continued in the follow-up period in the combined score (−1.76 (V) vs. −1.19 (P), $p = 0.0508$) and the symptoms score (−1.94 (V) vs. −0.30 (P), $p = 0.0144$). Intake of rescue medication was very low (n.s. for V vs. P in all seasons). There was an increase of specific IgG4 in the first season which remained at this higher level during the treatment seasons for V vs. P ($p = 0.0001$). Titration and daily SLIT intake were well tolerated. Most frequent reactions were mild to moderate local itching and burning in the mouth. Serious systemic or anaphylactic reactions were not reported.
Conclusion: This study shows in a large patient cohort that the seasonal sublingual immunotherapy with ultra-rush titration is effective with significant differences versus. placebo from the first treatment season on. It is indicative for a carry-over effect of seasonal SLIT. The treatment was well tolerated.

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Long-lasting effect according to the duration of sublingual immunotherapy: a 15-year prospective study

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Background: There are still few data on the longterm effects of sublingual immunotherapy and it is not known which is the optimal duration of the treatment.
Methods: In this prospective open controlled study we followed up for 15 years patients with respiratory allergy and monosensitized to mites. The subjects were subdivided in 4 groups: only drug therapy and sublingual immunotherapy for 3, 4 or 5 years. Clinical scores, pulmonary function and skin sensitizations were evaluated every year during winter months. The clinical effect was considered significant when clinical scores decreased by at least 50% from baseline. When the clinical scores overcome 50% of the baseline, the patients were vaccinated again.
Results: Seventy-eight patients were enrolled and 59 completed. In the 12 controls no change in clinical scores was seen throughout the study. In the patients receiving immunotherapy for 3 years a significant clinical benefit persisted for 3 years, then a progressive worsening was seen and after 7 years the scores raised significantly. In those receiving immunotherapy for 4 or 5 years, the clinical benefit persisted for 5 years and a significant worsening was seen 8 years after discontinuation. 58% of the controls displayed a reduction of FEV1 below 80% of predicted over 15 years, whereas this percentage was <10% in the immunotherapy groups. The occurrence of new sensitizations over 15 years in the controls and the 3 immunotherapy groups was 100%, 21%, 12% and 11%, respectively ($p < 0.01$). The second course of vaccination achieved a more rapid benefit (1 year).
Conclusion: This observation suggests that a duration of SLIT of at least 4 years can lead to a long-lasting clinical benefit.
Key words: sublingual immunotherapy, long-term effect; house dust mites

DIAGNOSIS OF ASTHMA

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Correlation between FEV1 and asthma control test in newly diagnosed asthma

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Background: Tools reflecting the multidimensional nature of asthma and that can be easily and quickly administered are needed for the assessment of asthma control in daily practice. The Asthma Control Test (ACT) was developed to meet this need. It consists of five questions enquiring about the frequency of symptoms in the last four weeks. Each question has five possible answers scored from 1 (worst) to 5 (best). Aim of this study was to assess whether there is some correlation between ACT and functional measures in outpatients with a new diagnosis of asthma.

Methods: Patients firstly referred for respiratory complaints and without a previous diagnosis of asthma, underwent a detailed clinical history, clinical visit, spirometry and ACT. Asthma was diagnosed on the basis of : (i) history of attacks of chest tightness, cough, wheezing, nocturnal awakenings and (ii) impaired spirometry with a reversibility of at least 12% in the FEV1 after salbutamol or, in alternative, a normal spirometry with a positive methacholine challenge test. Results. From April to June 2006, 32 patients (20 males, mean age 32.3, range 16–57 yrs) had a new diagnosis of asthma. Their severity according to GINA guidelines was: 1 intermittent, 9 mild, 18 moderate and 4 severe. Their FEV1 ranged from 62% to 112%. There was no correlation between ACT and FEV1 ($r = 0.26$; $p = 0.2$) (Fig 1).

Conclusion: Despite FEV1 directly reflects the bronchial obstruction, in our naïve patients it was not correlated with severity as self-assessed by ACT. This probably reflects the well known individual variability in the perception of symptoms. Our data further confirm that the diagnosis and follow-up of asthma must be based on an integrated approach including clinical aspects and clinical parameters.

Key words: FEV1, Asthma control

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Predicting asthma control in children - using exhaled nitric oxide (FeNO)

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Background: The world literature suggests a disappointing lack of asthma control as reflected by ongoing asthma morbidity. One of the problems in assessing asthma control has been reliable strategies to do so. Measurement of exhaled nitric oxide is a useful non-invasive and efficient method of assessing airway inflammation almost directly in children with asthma.

Methods: A random sample of asthmatic children (aged 5–11 years) attending the Childrens Asthma Clinic at Pretoria Academic Hospital and the Private Paediatric Practice of the second author had their asthma control assessed by 4 methods, namely, Doctor assessment, Symptom Score (Paediatric ACT), Lung function (FEV1/FEF25-75) and Exhaled nitric oxide (FeNO).

Results: 80 children (with mild persistent asthma) were studied. There was no significant correlation between assessment variables. Paerson correlation coefficient $r = -0.0179$ for ACT vs FeNO, $r = -0.1365$ for FEV1 vs FeNO. Only FEV1 and FEF25-75% reveal significant correlation. Using a multi-regression analysis all the variables together explain only 16.45% of the variation in FeNO and none of the coefficients have significant p-values.

Conclusion: If FeNO is the 'gold-standard' of assessment of asthma control, as has been suggested, then all the other measures we tested are a weak substitute for this assessment. It may be possible that our assessment methods were incomplete and that some other variable may prove more valuable in this regard. Alternatively It also seems likely that an important conclusion would be that asthma control requires more than one end-pont in assessment.

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Oxygen saturation measurement in obstructive outpatient: an useful tool for exacerbations management in GPs setting?

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Background: Severe asthma and COPD are characterized by clinical exacerbations. GPs are often required to decide between hospitalisation and home treatment for these patients.

Aim: To evaluate if the ambulatory measurement of Oxygen Saturation (SO2) could be safe and effective, to verify the correlation between SO2 measurement during scheduled visits an during exacerbations, to define the patient pattern that requires hospitalisation, to assess the possibility of SO2 database registration in Gps setting.

Methods: 450 GPs and 50 pneumologists have been enrolled in the study with the target to evaluate 16000 ptz (10000 by Gps; 6000 by pneumologists). Each physician has been provided with a pulse oxymeter. The SO2 measurement and the fill in of a specific questionnaire have been required to the outpatients attended for disease exacerbation.

Results: A descriptive analysis of results will be provided. The patients will be divided in two population according to the necessity of hospitalisation and a comparison of all the parameter registered in GPS and specialists setting will be performed. The relative importance of SO2 measurement for GPS and pneumologists will be evaluated.

Discussion: The present ongoing study should provided important information about the utility and safety of SO2 measurement in obstructive patient evaluation during exacerbation. The results should be useful in defining guidelines for exacerbation management.

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Budesonide/formoterol reduces the production of IL-5, RANTES and TNF-α from mononuclear cells but not the adhesion of activated eosinophils

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Background: Budesonide (BUD) / formoterol (FOR) combination (Symbicort®) for both maintenance and reliever therapy (SMART) results in the reduction in asthma exacerbations as compared with traditional fixed dosing regimens. The mechanism by which SMART provides outstanding clinical benefit remains to be elucidated, however, the use of combination therapy during the early stage of asthma exacerbation may intervenes in the development of subsequent airway inflammation. The objective of the study was to evaluate whether the use of BUD/ FOR at the early stage of cell activation can modify the adhesion of eosinophils or production of cytokines from mononuclear cells.

Methods: Eosinophils isolated from human peripheral blood were pretreated with either BUD (0.1 μM), FOR (0.1 μM), or BUD/ FOR for 30 min at 37 °C and stimulated with IL-5, LTD4, or PMA. The adhesion of eosinophils was evaluated using an EPO assay. Peripheral blood mononuclear cells (PBMC) were treated with BUD (0.1 μM), FOR (0.1 μM), or BUD/ FOR either 15 min or 4hr following the stimulation with a combination of ionomycin and PMA. The concentrations of IL-5, RANTES, and TNF-α in the culture supernatants of PBMC were examined by ELISA.

Results: Neither BUD, FOR, nor BUD/ FOR combination modified eosinophil adhesion induced by IL-5, LTD4, or PMA. By contrast, both BUD alone and BUD/ FOR combination significantly reduced the productions of IL-5, RANTES, and TNF-α by PBMC ($N = 6$, $p < 0.001$), despite these compounds were added either 15 min or 4hr following the stimulation of the cells.

Conclusion: Inhalant BUD/ FOR at early stage of exacerbation of asthma may suppress the progression of allergic inflammatory cascade via its inhibitory effect on mononuclear cells and hence prevents the development of severe asthma exacerbations.

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A review of three quality of life measures in asthma

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Background: The measurement of quality of life has become a standard feature in clinical trials, surveys and clinical practice, to capture aspects of asthma that are salient from the patients' perspective. Several disease-specific questionnaires have been developed for asthma. Three are widely used, but have never been compared: the Asthma Quality of Life Questionnaire (Juniper et al.), the Asthma Quality of Life Questionnaire (Marks et al.) and the Living With Asthma Questionnaire (Hyland et al.) We wish to review these instruments.

Methods: We reviewed the questionnaires with respect to number of items, item content areas, response format, item generation technique, item selection technique, and methods by which validity claims were made.

Results: We found variation in the number of items included (20,32,68) and the item content areas in which the items are grouped. The Hyland et al. questionnaire includes positive and negative items, while the other two questionnaires include negative items only. Each questionnaire uses a different response format. There was variation in the item generation methods, and the degree to which clinical experience was used as a source. Items were either selected using standard psychometric methodology (factor analysis, principal components analysis) or impact analysis, i.e. items are being selected which are most frequently perceived as important by respondents. Validity claims were made mainly by weak correlations with clincial markers of asthma (such as FEV1,% predicted or asthma medication) and strong correlations with generic quality of life measures.

Conclusion: The variation in methodology is important because the questionnaires measure different things, depending on the item wording and the domain structure. Guidance is needed to facilitate the choice of instrument for a given purpose, and the performance of the questionnaires should be evaluated against each other, using appropriate methods to show agreement (e.g. Bland-Altman plots).

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Sensitization patterns of allergens among asthmatics in Sri Lanka

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Introduction: The sensitization pattern among asthmatics in Sri Lanka is unknown.

Objectives: To determine the sensitization pattern among asthmatics in Sri Lanka.

Methodology: The study was carried out in forty physician-diagnosed asthmatic patients. Twenty two non-asthmatics were recruited as controls. Sensitization to 12 allergens was assessed by skin prick test using standardized allergen extracts. A skin wheal diameter, 3mm greater than the negative control was considered a positive reaction.

Results: The mean age of asthmatics was 32.4 years (range 9–59) and 57.5% were males. The mean age of controls was 32.2 years (range 14–58) and 59.1% were males. The analysis of the data showed that 97.5% asthmatics had positive skin reaction to at least one allergen compared to 72.7% prevalence in the control group. The common allergens among asthmatics were Dermatophagoides pteronyssinus (77.5%), blomia (65.5%), cockroach (55%), storage mite (35%), latex (20%)), cat fur (17.5%), aspergillus (12.5%), grass pollen (12.5%), cow's milk (12.5%) egg white (10%) cereals (7.5%) and dog hair (5.%). The pattern of sensitization among non-asthmatics was Dermatophagoides pteronyssinus (54.5%), cockroach (50%), blomia (36.4%), aspergillus (36.4%), grass pollen (27.3%), storage mite (22.7%), cow's milk (18.2%), latex (13.6%), egg white (13.6%), cereals (13.6%), dog hair (9.1%) and cat fur (9.1%). Sixty two percent (n = 25) of the asthmatics had at least one other allergic manifestation while non-asthmatics with other allergic manifestation were 36.4% (n = 8). Other allergic manifestations in the asthmatic group were rhinitis alone 22 (55%), eczema alone 1(2.5%) and both rhinitis & eczema 2(5%) whereas in the non-asthmatic group it was rhinitis alone 7 (31%), eczema alone 0% and both rhinitis and eczema 1(4.5%).

The pattern of sensitization for mites alone, cockroaches alone and mites and cockroaches in the asthmatic group with rhinitis (n = 24) was 23 (57%), 11(27%) and 11(27%) and in the non-asthmatic group with rhinitis (n = 7) was 3 (13%), 4 (18%) and 3 (13%) respectively.

Conclusion: The results show that mites, and cockroaches are the most common sensitizing allergens in asthmatics and the commonest association of asthma was rhinitis in Sri Lanka.

INFECTION AND IMMUNODEFICIENCY

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Successful treatment of disseminated BCG infection in a SCID patient with granulocyte colony stimulating factor

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Background: Severe combined immunodeficiencies (SCID) are disorders with impairment of humoral and cellular immune functions. Persistent infections with opportunistic organisms are major causes of death. The prognosis of disseminated bacillus Calmette-Guèrin (BCG) infection in immunocompromised host is unfavorable since response to standard therapy is poor.

Methods: We report a successful treatment of disseminated BCG infection with granulocyte colony stimulating factor (G-CSF) in a 15-month-old girl with SCID. The infection resulted from BCG vaccine administered during the newborn period. The patient presented at 6 months of age with chronic diarrhea, failure to thrive, and generalized candidiasis. She was subsequently diagnosed as a SCID with IL-7Rα deficiency by gene sequencing. HLA-haploidentical bone marrow transplant (BMT) from her father was attempted at the age 8 months. Graft versus host disease soon developed with failure of engraftment. Seven months post-BMT, she developed abscess at right upper arm and multiple subcutaneous nodules on skin all over the body. Numerous acid-fast bacilli (AFB) were demonstrated from smears of the abscess materials. *M. tuberculosis* complex was identified by culture and polymerase chain reaction (PCR). A diagnosis of disseminated BCG infection was made. After 2 months of intensive antituberculous (anti-TB) therapy comprising five agents (isoniazid, rifampicin, ethambutol, levofloxacin and amikacin) guided by in-vitro antimicrobial sensitivities of the culture, failed to control the infections. She subsequently developed hepatosplenomegaly and BCG-osteomyelitis of left femur.

Results: G-CSF was added to her treatment regimen to enhance phagocytosis. She recovered quickly within 2 weeks with G-CSF treatment as evident by significant clinical improvement. G-CSF was continued for 4.5 months while she continued to receive triple anti-TB therapy and monthly intravenous immunoglobulin infusions without relapsing of active tuberculosis infection.

Conclusion: To our knowledge, this is the first case of SCID with disseminated BCG infection which was successfully treated with anti-TB agents in conjunction with G-CSF. Since serious BCG infections in SCID is not

uncommon in developing countries where BCG vaccination is mandatory to all newborns, combination of G-CSF and anti-TB drugs should be considered.

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IL-17F His(161)Arg but not IL-8 A(−251)T is associated with the development of respiratory syncitial virus bronchiolitis in the Japanese population

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Background: The involvement of neutrophil-mediated inflammation may play an important role in the pathogenesis of acute respiratory syncytial virus (RSV) bronchiolitis. Interleukin-8 (IL-8) mediates the activation and migration of neutrophils from peripheral blood to the airway. IL-17F is a recently discovered cytokine that plays a role in tissue inflammation by inducing release of proinflammatory and neutrophil-mobilizing cytokines. The contribution of genetic polymorphisms of IL-8 and IL-17F has not yet been clarified in the Japanese population.

Methods: Forty-eight children who had been hospitalized because of severe RSV bronchiolitis were recruited. The exclusion criteria included cardiac diseases, chronic respiratory diseases, previous wheezing episodes, an age > 24 months and prematurity. Two hundred and three children (age 6–12) who had never had wheezing episode were recruited as control subjects of this study. These control subjects were considered to have suffered from a mild RSV infection but not bronchiolitis during infancy because it is known that almost all children are infected by RSV at some time before reaching three years of age.

We genotyped IL-8 A(−251)T and IL-17F His(161)Arg by modified amplification refractory mutation system polymerase chain reaction (ARMS-PCR) method, and investigated a relation between these SNPs and the development of RSV bronchiolitis.

Results: The distribution of the genotype and allele of IL-8 A(−251)T in children with RSV bronchiolitis was similar to that in controls. On the other hand, the distribution of genotype of IL-17F His(161)Arg in children with RSV bronchiolitis was significantly different from that in controls ($p = 0.034$). In comparison to the controls, the frequency of His/His ($p = 0.011$, odds ratio 0.42) was significantly lower in children with RSV bronchiolitis.

Conclusion: IL-17F His(161)Arg but not IL-8 A(−251)T is associated with the development of RSV bronchiolitis in the Japanese population. Our results suggest that the dysfunction of IL-17F may be involved in the pathogenesis of RSV bronchiolitis.

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Expression of alpha-, beta- and lambda-interferon upon during respiratory virus infection

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Background: Rhinoviruses are a major cause of common colds and asthma exacerbations worldwide. It is known that rhinovirus infects and replicates in respiratory epithelial cells of lower respiratory tract inducing proinflammatory cytokines. Type I interferons such as IFN-alpha, IFN-beta and newly discovered type III interferons - IFN-lambdas play vital role in innate immune response against viruses. The method used.

In this study we investigated the potential of different cell types such as, human bronchial epithelial cells (BEC), BEAS-2B cells and PBMC to express and produce various type I and type III interferons upon respiratory virus infection by ELISA and RT-PCR.

Results: In BECs we detected induction of IFN-a mRNA expression by 8 hours, induction of IFN-lambda1 mRNA by 24, IFN-lambda2/3 was induced by 8 and 24 hours. In BEAS-2B cells we detected induction of IFN-alpha mRNA expression by 8 hours, IFN-lambda1 mRNA from 8-hours with peak at 24 and induction of IFN-beta from 24 hours. By ELISA we also observed production of IFN-lambda1 and IFN-beta protein by 24 hours.

Conclusion: IFN-lambdas were induced either earlier and/or to a greater degree than the type 1 IFNs. In PBMC induction of IFN-alpha, IFN-lambda1 and IFN-beta mRNA expression and protein production were all detected from 8 hours. However induction of IFN-alpha was to a much greater degree than -beta or -lambda. Lambda IFNs appear important in epithelial cell responses to rhinovirus infection, while alpha IFNs are the major IFNs released by macrophages.

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Different kinases (PI3K, p38 MAPK, PKA, and classical, novel, and atypical PKC isoforms) are critical for ICAM-1 upregulation on RSV-infected human endothelial cells

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Background: RSV is the most frequent cause of bronchiolitis and pneumonia in infants requiring hospitalization. Especially children with underlying heart/lung failure show an enhanced mortality and morbidity rate during RSV infection. Thereby, the infection of human lung epithelial cells with respiratory syncytial virus (RSV) induces an intense inflammatory response resulting in a prominent recruitment of leukocytes into the lung. Recently, we showed by in vitro experiments that human primary lung endothelial cells (HMVEC-L, HPAEC) and HUVEC are also target cells for a productive RSV infection. Furthermore, infected endothelial cells upregulated their ICAM-1 cell surface expression resulting in an enhanced adhesion and transmigration rate of cocultered effector cells. However, cellular signaling pathways mediating the NF-κB-dependent RSV-induced ICAM-1 expression are still not known. Therefore, by using HUVEC as a reliably in vitro-RSV-endothelial cell infection model we investigated signal transduction molecules involved in the RSV-induced ICAM-1 cell surface expression.

Methods: The endothelial cells were preincubated with pharmacological inhibitors, infected with highly purified RSV particles, washed, and cultured for 48 h in the presence of freshly added antagonists. The used inhibitors did not interfere with the infection process of the HUVEC. The ICAM-1 cell surface expression was determined by FACS analysis.

Results: We observed a PI3K-, p38 MAPK-, PKA-, and PKC-dependent ICAM-1 expression pattern in RSV-infected endothelial cells. By analyzing the involved PKC isotypes in more detail we determined that the classical α and γ PKC isotypes, the novel isotypes η and θ, and the atypical ζ isotype are activated and significantly involved in the ICAM-1 upregulation on RSV-infected endothelial cells.

Conclusion: Our data suggest that the RSV-induced ICAM-1 expression on infected human endothelial cells, responsible for an enhanced adhesion and transmigration rate of immune effector cells into the RSV-infected lung, might be significantly controlled by inhibition of distinct signal transductiaon molecules.

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Seropositivity of cytomegalovirus in women with unexplained recurrent spontaneous abortions

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Background: Cytomegalovirus (CMV) is an ubiquitous agent and the rate of seropositivity in the worldwide adult populations is 60–100%. Primary CMV infection in pregnancy is a cause of spontaneous abortion, IUFD, …. and congenital illness and disability. To find out about the probability of any relation of recurrent, non-active or active CMV infection with pregnancy loss, in women suffering from recurrent spontaneous abortions (RSA), humeral immunity to CMV has been assessed in these patients.

Materials and Methods: To assess the prevalence of CMV viral infections in women with RSA in a case control study Patients were recruited and age matched at the Women's Clinic, during the period of one year. one group of recurrent aborters (Group 1; n=108) and One group of normally pregnant women (Group 2; n =108) were evaluated, including demographic, medical, clinical, and serological data (anti-CMV IgG , IgM).

Results: The mean age of the two groups was 28.2 and 24.47 years. The normal pregnant women had a significantly higher prevalence of anti-CMV (93.5% IgG, 0% IgM) in contrast to (78.7% IgG, 11.1% IgM) in recurrent aborters respectively.

A significantly lower prevalence of serum anti-CMV antibodies was observed for RSA women compared with age-matched pregnant female controls.

Conclusion: These results indicate that women with unexplained RSA have difficulty in responding to CMV infection and could be important when considering fetal infection due to reactivation of chronic CMV infection in the course of pregnancy.

The role of viruses in induction of autoimmune condition and whether women with unexplained RSA have difficulty in responding to CMV, all needs more future researches.

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B-cell reconstitution with cord blood but not with bone marrow transplantation in two siblings with X-linked SCID

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Introduction: Stem cell transplantation (SCT) is the treatment of choice for X-linked SCID. Successful SCT from matched family donors generally results in T-cell engraftment. Despite this positive outcome, up to 50% of patients do not achieve B-cell engraftment and require monthly IVIG infusion.

Methods: We followed two siblings with IL-2 Receptor (common γ chain) deficiency X-Linked SCID, prior to, during, and post transplantation. The first had haploidentical BMT. The second had unrelated umbilical cord blood transplantation (UCBT).

Results: The sibling 1 received a T cell depleted bone marrow infusion donated from his mother at age 3 months. His pre-transplant conditioning consisted of busulfan, cytoxan, ATG and cyclosporine. He received IVIG and solumedrol post transplantation. He is now 10 years old. He currently has only a small number of non-functional B cells. Thus he continues with hypogammaglobulinemia, requiring monthly IVIG infusions. His half-brother received a 7/8 matched cord blood transplantation at age 2.5 months. His pre-transplant conditioning consisted of busulfan, cytoxan, and cyclosporine. He received IVIG and solumedrol post transplantation. He is now 2 years old. He achieved reconstitution of his B-cells by 3 months post transplantation. Currently his quantitative immunoglobulins are within normal limits, with appropriate antibody function, thus he does not require IVIG.

Conclusion: We concluded that unrelated UCB is an alternative source of stem cells for transplantation in children with severe T-cell immune deficiency disorders when a suitable HLA-matched donor is not available and when a T-depleted haploidentical preparation is not beneficial. Benefits of UCBT include rapid and reliable recovery of immune function, low risk of GvHD, and low viral transmission rate. We present a family with two siblings, both with X-linked SCID. There was discordance in B cell engraftment. The sibling treated with UCBT was able to reconstitute B cell function, with production of appropriate quantitative and functional B cells. Overall data from the National Cord Blood Program documents that the hematopoietic reconstitution after UCBT, is much better in children given transplants of UCB than in both children and adults who received BMT. This suggests that cord blood provides a higher likelihood of B cell function compared to HLA haploidentical transplants and should be considered for patients requiring SCT for Primary immunodeficiency.

DIAGNOSIS AND MANAGEMENT OF ALLERGY DISEASE

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What proportion of allergy referrals could be dealt with in primary care?

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Background: In the UK it has been proposed that a large proportion of allergy referrals to a specialist in secondary care could be dealt with in the community by a GP with special interest (GPwSI) in allergy. This study assesses what proportion of referrals could be dealt with in a GPwSI clinic with a predefined range of facilities including: expertise in accurate diagnosis and management of allergy; skin prick testing for aeroallergens; provision of advice on allergen avoidance; ability to assess suitability for desensitisation.

Methods: Consecutive referrals of patients with suspected allergy to a Respiratory Medicine Clinic were reviewed by 3 allergy specialists independently. The referral letter was reviewed and a judgement made on the appropriateness for consultation with a GPwSI. The case was then re-assessed with the benefit of subsequent correspondence and a further judgement made on the appropriateness for consultation with a GPwSI. The proportion judged suitable was calculated and inter-rater agreement was assessed.

Results: 46% of referrals were judged appropriate for management by a GPwSI and after review of the outcome of referral this reduced to 42%. Only 4% of referral letters contained insufficient evidence to make a decision about appropriateness. Two fifths of patients being referred to a respiratory clinic with allergic problems may be dealt with by a GP with a special interest in allergy. The majority of referral letters contain sufficient detail to accurately predict those patients who are suitable for alternative care.

Discussion: The development of a Primary Care Allergy Service could liberate capacity in the specialist setting and enable the specialist to focus on the more complex allergy problems.

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Assessing skin prick tests reliability in ECRHS-I

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Introduction: Atopy, the clinical definition of an IgE high-responder, can be documented by the detection of IgE antibodies in serum or by a positive skin prick test. Selection of a reaction size at which to declare a skin test reaction as positive was not well assessed in epidemiological studies. The aim of the study was to identify the best cut-off level for skin prick tests.

Methods: Using the data collected during the ECRHS I (European Community Respiratory Health Survey) the association of serum allergen-specific IgE and skin prick tests (Dermatophagoides pteronyssinus, cat, timothy grass and Cladosporium) were assessed.

Results: On the 13,391 subjects (50.4% of men), the proportion of positive skin tests ranged from 9.7% (cut-off >5mm) to 20.4% (cut-off >0mm) for Dermatophagoides pteronyssinus, 3.4 to 10.1% for cat, 9.6 to 17.1% for timothy grass and 0.2 and 2.2 for Cladosporium. The most appropriate cut-off appeared to be over 0mm for Dermatophagoides pteronissinus, cat and timothy grass (Youden Index over 0.76). However, the relationship between serum IgE and skin prick test for Cladosporium was weak (Youden index under 0.55).
Conclusion: In epidemiological studies, a single method may be chosen to assess allergenic sensitivity. A cut-off level of over 0mm for skin prick tests appeared to be the most discriminative.

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Combined use of allergen skin prick test and specific IgE antibody on allergen diagnosis in asthmatic children

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Background: To analyze the advantages of combined analysis of allergen skin prick test "SPT" and Phadiatop/specific IgE antibody on the allergen diagnosis in asthma children.
Methods: Inhalant allergen SPT and Phadiatop test were done in 57 asthma children. 33 cases of those asthma children were measured serum specific IgE antibody against Dermatophagoids pteronyssinus.
Results: Dermatophagoids, molds and pets were the main inhalant allengens in asthma children. The positive rates of SPT and Phadiatop in 57 asthma children were 86.0% and 79.0% respectively, and the consistence rate between SPT and Phadiatop was 86.0%. 5 cases with negative Phadiatop were confirmed to have molds allergy via SPT and molds specific IgE test. The consistence rate of Dermatophagoids pteronyssinus SPT and specific IgE was 97.0%.
Conclusion: It is helped to improve the sensitivity and specificity of allergen diagnosis in asthma children when doctors combined analyze the results of allergen skin prick test and specific IgE test.

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Comparison between skin prick test and a reversed enzyme allergo-sorbent test

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Background: Several methods are available for the quantitative determination of allergen specific immunoglobulin E (sIgE) representing a hallmark in the diagnosis of type I hypersensitivity reactions and allergic diseases. One of these methods, the ALLERG-O-LIQ system (Dr. Fooke Laboratorien, Germany), is based on the reversed enzyme allergo sorbent test protocol using anti-IgE coated microtiterplates combined with biotinylated allergens and streptavidine-HRP conjugate. Objective of our study is the comparison of the ALLERG-O-LIQ and skin prick test (SPT) results.
Methods: Serum samples (n = 96) were collected from undiagnosed individuals and tested for sIgE to ten common inhalant allergens including Timothy Grass (g6), Cultivated Rye (g12), Mugwort (w6), *Alternaria alternata* (m6), *D. pteronyssinus* (d1), *D. farinae* (d2), cat (e1), dog (e2) and *Aspergillus fumigatus* (m3). Further, skin prick tests (SPT, Allergopharma, Germany) to above mentioned allergens were performed. Agreement between the results of ALLERG-O-LIQ and SPT was statistically evaluated using different methods including kappa agreement, positive (PPV) and negative predictive value (NPV) and diagnostic efficiency (DE).
Results: The prevalence (in %) of the individual allergens was found at (SPT / ALLERG-O-LIQ) 38.5 / 33.3 (g6), 33.3 / 26.0 (g12), 16.7 / 8.3 (w6), 6.3 / 7.3 (m6), 33.3 / 24.0 (t3), 27.1 / 16.7 (d1), 27.1 / 17.7 (d2), 21.9 / 10.4 (e1), 6.3 / 7.3 (e2) and 0.0 / 1.0 (m3). The agreement (% / Kappa) between both methods was 88.2 / 0.80 (g6), 90.4 / 0.83 (g12), 86.7 / 0.53 (w6), 95.8 / 0.75 (m6), 87.7 / 0.77 (t3), 83.8 / 0.64 (d1), 87.8 / 0.73 (d2), 84.9 / 0.59 (e1) and 95.8 / 0.75 (e5). Statistics for m3 was not calculated. Overall agreement (% / Kappa / $\times^2$) for seasonal, perennial and all allergens tested was 89.7 / 0.78 / 292.3, 90.1 / 0.68 / 231.7 and 89.9 / 0.74 / 544.0, respectively. Sensitivity, specificity, PPV, NPV and DE (all values in %) were found at 73.2, 98.6, 94.7, 91.4 (seasonal) 59.5, 99.0, 92.2, 92.5, 92.5 (perennial) and 92.1 and 67.8, 98.8, 93.8, 92.0 and 92.3 (combined).
Conclusion: Overall, ALLERG-O-LIQ and SPT results are in good agreement. However, differences in the sensitivity of ALLERG-O-LIQ were observed among individual allergens. Based on our data we conclude that ALLERG-O-LIQ represents a reliable test for the quantitative detection of sIgE to inhalant allergens.

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Total IgE in urban black south african teenagers: the influence of atopy and helminth infection

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Background: IgE levels are usually elevated in allergic diseases, being highest in atopic eczema, followed by atopic asthma and allergic rhinitis. Genetic factors are believed to play a role in total IgE levels, with higher levels seen in Black African subjects. Total IgE is also raised in parasite infection. Thus, the higher total IgE levels in Black Africans could be due to environmental rather than genetic factors. Few studies investigate the usefulness of total IgE levels in the assessment of atopy in Black Africans.
Aim: The objective of this study was to determine the total IgE levels in unselected urban Black African high school children and to correlate this with atopy and ascaris sensitization.
Methods: Two hundred and twenty Urban Xhosa children (mean age 17 years) attending a school in Cape Town were studied. Atopic status was assessed by means of specific allergen sensitization (ALK® skin prick tests to 8 inhalant and 4 food allergens), self reported asthma and bronchial hyperresponsiveness measured by methacholine challenge. Total IgE and Ascaris specific IgE were measured by CAP-RAST (Pharmacia). Total IgE levels were subanalysed according to atopic status and ascaris sensitisation in order to establish ranges of total IgE in atopic subjects, ascaris sensitised subjects, and non-atopic, non ascaris sensitised subjects.
Results: Total IgE levels were markedly skewed toward the left and were not distributed in a Gaussian or a log-normal distribution. Mean total IgE was 307.7 kU/L. The median was 106 kU/l and the interquartile range 50.4 kU/l to 288 kU/l. Skin prick tests were positive for aeroallergens in 32.3 % of subjects. Thirty four percent had elevated ascaris IgE.

Total IgE was higher in atopic versus non-atopic subjects and correlated with the number of positive skin prick tests (Kruskall Wallis ANOVAR $p < 0.0001$), self reported asthma ($p = 0.025$) and bronchial hyperresponsiveness ($p = 0.0002$). In addition total IgE correlated with ascaris IgE ($p < 0.0001$). Subjects with no ascaris sensitisation had median total IgE of 77.1 kU/l, similar to the levels seen in people of other genetic origins.
Conclusion: This study has demonstrated that total IgE is correlated on one hand with atopy, bronchial hyper responsiveness and self reported asthma and on the other hand with ascaris sensitisation. The likelihood of helminthic infection rather than genetic differences, is thus the major factor determining a population's specific IgE range.

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Association between serum total IgE, allergic diseases and geohelminth infections in Sri Lankan children

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Background: Elevated levels of total IgE (tIgE) in serum are characteristic of allergic diseases. Levels of tIgE are influenced by genetic predisposition, age, sex and helminth infections. However, the association between tIgE and allergic diseases in children living in areas endemic for helminth infections is not clear. The aim of our study was to assess the relationship between tIgE, allergic diseases and geohelminth infections in children.

Methods: A total of 640 schoolchildren 9–11 years of age was selected by stratified random sampling. Data regarding allergic diseases (asthma, rhinitis and eczema) were collected by a standard questionnaire given to the parent or guardian. Screening for helminth infections was done by examining their fresh stool samples by modified Kato-Katz technique. Serum tIgE was measured by Fluoroenzymeimmunoassay in 67 geohelminth-positive subjects and in a comparable group of geohelminth-negative subjects.

Results: The mean age in the study population was 10 years (SD±0.3). The prevalence of geohelminth infection was 15.5%. *Trichuris trichiura* (14.3%) was the most common followed by *Ascaris lumbricoides* (4.2%) and hookworm (0.2%). Mixed infection was detected in 20.3% of infected children. Infection intensity was light in 68.9% of infected children while 28.4% and 2.7% showed moderate and heavy infection respectively. The cumulative prevalence of allergic diseases was 33.7%. Prevalence of asthma, rhinitis and eczema was 17%, 21.4% and 5% respectively. Serum tIgE concentrations showed a positively skewed distribution. Geometric mean (GM) for tIgE for the geohelminth infected group (1039.9kU/L) was significantly higher than that of the non-infected group (575.4kU/L) ($p = 0.004$). It was also higher in the allergic group (933.3kU/L) than in the non-allergic group (639.7kU/L) but the difference was not statistically significant ($p = 0.068$). The GM for tIgE for non-allergic children in the study population was much higher than that seen in non-allergic children in developed countries.

Conclusion: Serum tIgE concentration was strongly associated with the presence of geohelminth infections in children. Serum tIgE may not be a useful marker for allergic diseases in children living in areas endemic for geohelminth infections.

PEDIATRIC ALLERGY AND ASTHMA II

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Evidence of eosinophil activation in adenoid and tonsil tissues from atopic children

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Background: Adenoid and tonsil have been considered as immune organs. They may present different cellular and cytokine profiles according to the atopic status. The objective of this study was to find the status of eosinophil activation and compare between atopic and non-atopic children.

Materials and Methods: Adenoid and tonsil tissues obtained from 40 children (18 atopics and 22 non-atopics) were enrolled. Seven atopic and 9 non-atopic children had chronic inflammatory diseases such as chronic rhinosinusitis (CRS) and/or otitis media with effusion (OME). Atopic status was classified as a positive result (>2+) on MAST test to common inhalant allergens. Serum total IgE and eosinophil count were measured. ECP level and cellular activation markers, including sCD23, sIL-2R and IL-6 within the supernatant of adenoid and tonsil tissue were measured by CAP system (Pharmacia, Sweden) or ELISA.

Results: The ECP level in adenoid and tonsil tissue from atopic children was 451.0μg/L, 514.2μg/L, which were significantly higher than those from non-atopic children (353.2 μg/L, 374.4 μg/L, respectively). The ECP level in adenoid and tonsil tissues from atopic children with CRS showed the highest (564.6 μg/L and 718.1 μg/L), followed by those from non-atopic children with CRS (449.3 μg/L and 430.7 μg/L), those from atopic children without CRS (371.5 μg/L and 371.6 μg/L), and those from non-atopic children without CRS (291.5 μg/L and 338.2 μg/L). And the ECP level in adenoid were significantly correlated with locally produced total IgE and cellular activation markers, including sCD23, sIL-2R and IL-6.

Conclusion: This findings suggest that activated eosinophil could be involved in inflammatory response occurring in the adenoid and tonsil from both atopic and non-atopic children. The more activated eosinophils, the more CRS can be combined. The activation of T cell, especially Th2 cell could lead to the activation of eosinophil in adenoid and tonsil tissues.

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Plant homeodomain finger protein gene polymorphisms are associated with plasma total IgE and exhaled nitric oxide levels in Chinese children

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Background: Single-nucleotide polymorphisms (SNPs) of the gene encoding plant homeodomain finger protein (*PHF11*) were shown by positional cloning to be associated with severe asthma and elevated circulating total IgE level in Caucasians. These SNPs were also associated with childhood atopic dermatitis. Such genetic association has not been studied in Chinese. The objective of this study is to investigate the association between *PHF11* SNPs and asthma phenotypes in Chinese children.

Methods: 269 asthmatics and 165 non-allergic children were recruited from our paediatric clinics. Their lung function was assessed by spirometry, and exhaled nitric oxide (FeNO) levels were measured online by chemiluminescence analyser at expiratory flow rate of 50 ml/s. Plasma total and specific IgE were quantified by immunoassays. Ten SNPs in *PHF11* were genotyped by multiplex SNaPshot™ reaction with an ABI-310 Genetic Analyser. The linkage disequilibrium (LD) pattern of these SNPs was analysed by Haploview, and the association between asthma traits and *PHF11* was analysed by multivariate regression.

Results: The mean (SD) log-transformed plasma total IgE and FeNO levels in cases and controls were 2.62 (0.61) and 80.5 (59.2) ppb and 1.81 (0.71) and 40.6 (43.2) ppb, respectively ($P < 0.001$ for both). All 10 SNPs followed Hardy-Weinberg equilibrium ($P > 0.05$). There were significant interethnic variations in *PHF11* minor alleles (up to 19%) in our children when compared with Caucasians. *PHF11* polymorphisms were not associated with asthma diagnosis ($P > 0.5$), asthmatics with $FEV_1 < 65\%$ of predicted ($P > 0.3$), or atopy ($P > 0.09$). Multivariate linear regression showed that *PHF11* T167A and G491A were associated with plasma total IgE ($P = 0.048$ and 0.051, respectively); and C507T and C732T were associated with FeNO ($P = 0.019$ for both). Two haplotype blocks could be constructed - one with T79C, G491A, C507T and T661A ($D' > 0.9$) and the other with T395C and C732T ($D' = 0.95$). Three major haplotypes from these 6 SNPs (CTGA CT [53%], TTAT TT [29%] and TCGT TC [18%]) were identified, but which were not associated with asthma diagnosis ($P>0.3$).

Conclusion: Our results suggest that *PHF11* polymorphisms are associated with plasma total IgE and FeNO levels but not asthma diagnosis in Chinese children.

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Transforming growth factor beta-1 In early childhood wheezers

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Background: Transforming growth factor beta-1 (TGF beta-1) is a multifunctional cytokine involved in pro and anti inflammatory pathways. TGF beta-1 is increased in the lungs of individuals with asthma. This prospective case-control study was carried out to outline the changes in serum TGF beta-1 in children with recurrent wheezing episodes and to differentiate between persistent and transient wheezers.

Methods: Sixty infants and young children with ages ranging from four months to three years presented by history of recurrent wheezing attending the Allergy Clinic, New Children's Hospital, Cairo University were enrolled in the study as well as twenty age and sex matched healthy children that were included as control group. Patients were subdivided into 2 subgroups: 41 early-onset asthmatics and 19 transient wheezers. All patients and controls were subjected to history taking, thorough clinical examination and laboratory investigations including CBC for Hb, total leucocytic count and absolute eosinophilic count, liver and renal functions, total serum IgE and biologically active serum TGF beta-1 by ELISA technique.

Results: 1-Mean serum TGF beta-1 was higher in patients (29.18 ± 14.3 ng/ml) than controls (13.13±4.22 ng/ml) with a statistically significant difference ($P < 0.001$). 2-Mean serum TGF beta-1 was higher in early-onset asthmatics (34.76 ± 13.78 ng/ml) than in transient wheezers (17.68 ± 4.63 ng/ml) with statistically significant difference ($P < 0.001$). 3-There was a significant positive correlation between serum TGF-beta 1 and absolute eosinophilic count in early-onset asthmatics ($r = 0.63$, $P<0.001$) as well as in total patients ($r = 0.59$, $P<0.001$). 4-There was no significant correlation between serum TGF beta-1 and total serum IgE in neither both patients subgroups nor in the patients group as a whole. Also there was no significant correlation between IgE and absolute eosinophilic count in total patients group. 5-Mean total serum IgE level was statistically significant higher in early-onset asthmatics than that in transient wheezers in those aged less than one year ($P=0.003$).

Conclusion: These findings support the hypothesis that serum TGF beta-1 may be a valuable diagnostic and prognostic biomarker in early diagnosis of asthma and in differentiation between early-onset asthma and transient wheezing among infants and young toddlers.

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Prostanoid DP receptor gene is not a major candidate gene for asthma and atopy in Chinese children

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Background: Airway sensitisation requires the expression of prostanoid DP receptor in mice. Functional haplotypes of this gene (*PTGDR*) are associated with asthma but not plasma total IgE in Whites and Black Americans. In contrast, another study in Latinos suggested that these genetic markers were not important determinants of asthma-related traits in this population. This study investigated whether six single-nucleotide polymorphisms (SNPs) of *PTGDR* are associated with asthma-related traits in Chinese children.

Methods: Two independent groups of southern Chinese children were recruited. The primary cohort consisted of 291 asthmatics and 178 non-allergic children from our paediatric clinics. Our replication cohort consisted of 556 schoolchildren from a population-based study that investigated obesity and metabolic syndrome in Chinese children. Plasma total and specific IgE were measured by immunoassays. Six *PTGDR* SNPs were determined by multiplex SNaPshot™ genotyping, and their linkage disequilibrium (LD) pattern was analysed by Haploview. Multivariate regression was performed to analyse the association between asthma and atopy traits and *PTGDR* SNPs.

Results: T-549C and C-441T were in complete LD ($D' = 1.0$ and $r^2 = 0.934$), and T alleles of these SNPs were linked together (with TT frequency being 0.671). *PTGDR* C367A and G894A were monomorphic. There was significant interethnic variations in *PTGDR* minor alleles and haplotypes (up to 41% and 38%, respectively) when compared with Whites or Latinos. Asthma diagnosis and aeroallergen sensitisation did not differ between children with different *PTGDR* genotypes ($P > 0.1$). Linear regression revealed weakly significant associations between total IgE and *PTGDR* T-197C and G1044A. When primary and replication cohorts were combined, plasma total IgE was marginally associated with T-197C ($\beta = 0.061$, $P = 0.049$). None of the SNPs was associated with atopy ($P > 0.2$) and aeroallergen sensitisation ($P > 0.25$). *PTGDR* haplotypes were not associated with any asthma-related trait.

Conclusion: Our results do not support *PTGDR* to be a major candidate gene for asthma diagnosis or severity, or atopy in Chinese children.

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Influence of respiratory syncitial virus in the immunological development of children from 0 to 2 years of age

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Following a bronchiolitis caused by respiratory syncytial virus (RSV), later recurrent wheezing are common.

Objective: The aim of this study was to establish the relationship between repiratory syncytial virus and the modifications in immunity that cause appearance of childhood asthma.

Patients and Methods: The prospective study Two groups of study: CHILDREN WITH BRONCHIOLITIS. (Group A). The problem group of children was originally made up of 65 children (with an average age of 3 months, 1 day to 7 months) who were admitted to this Hospital for their first episode of VRS positive bronchiolitis, from December 1997 to February 1998. 50 of them completed follow-up for 2 years. In the tests done at the moment of admittance :hemogram, and Ig G, Ig M, Ig A, Ig E, lymphocyte count, eosinophil cationic protein,IL-4 IFN-g. were at 6, 12 and 18 months, At 18 months an adhesion molecules count was requested: sVCAM.1, sLselectin. The second group (group B), was made up of 80 children, sub-divided into 4 groups of 20 according to age: IL- 4, IFN-g and adhesion molecules.

Results: In the acute stage of the bronchiolitis, the immunological response to the viral infection was anomalous, in that they presented a humoral immunity response (with an increase of immunoglobulins G, A, M, the later with p = 0,014) and a decrease in T, T4 and T8 cellular immunity. The immunolological response to the viral infection must be of cellular predominance, in order to be effective. Among all children suffering from VRS bronchiolitis, we found a statistically significant increase (p = 0,001) of sL-Selectine at 18 months of age in comparison with healthy children of the same age.

Conclusion: the characteristic patterns that lead us to believe that a child who suffers from bronchiolitis will develop asthma are: Increase in humoral immunity response at the acute stage of the bronchiolitis Decrease in cellular immunity response, from the acute VRS infection until 6 months of age Increased IL-4 values mainly at 6 and 12 months of age. Decrease in INF-g between 6 and 18 months of age.

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Association of polymorphism of the mast cell chymase gene promoter region (-1903 G/A) and (TG)n(GA)m repeat downstream of the gene with bronchial asthma in children

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Background: Mast cell chymase is an important mediator of inflammation and remodeling in the asthmatic lung. Various studies examined the association between -1903 G/A single nucleotide polymorphism (SNP) of the mast cell chymase gene (CMA1) and allergic phenotypes with inconsistent results. The (TG)n(GA)m repeat polymorphism 254 bp downstream of the chymase gene was previously found to associate asthma in adults. We sought to investigate the association of the -1903 CMA1 SNP as well as the (TG)n(GA)m repeat polymorphism with childhood asthma and its associated traits in a case-control study aiming to identify local data from our country.

Methods: The study comprised 15 children (6–10 years old) with bronchial asthma and a family history of atopy enrolled consecutively during asthma exacerbation and 15 age and sex matched children with negative personal or family history of allergy as a control group. They were subjected to clinical evaluation and serum total IgE estimation as well as search for polymorphism in the CMA1 gene promoter region 1903 G/A using PCR restriction fragment length polymorphism based genotyping (RELP) and the (TG)n(GA)m repeat polymorphism 254 bp downstream of the CMA1 gene.

Results: Investigating the -1903 G/A SNP revealed significant difference in the distribution of CMA1 A and G alleles between the patients and controls. Allele G was detected in 70% of patients compared to 16.7% of controls, while allele A was overexpressed in the control group (83.3%). Our data showed for the first time a positive association between -1903 G/A SNP and asthma. Concerning the (TG)n(GA)m repeat, a significant difference was obtained in the allele counts among patients and controls in the current study. The largest difference was observed in allele 39 which was only represented in the asthmatic children and allele 37 which was overexpressed among the controls. A positive association for the allele 39 of the (TG)n(GA)m repeat with serum IgE levels was also detected in our series. The findings are limited by the sample size.

Conclusion: We report the association of -1903 G/A SNP and (TG)n(GA)m repeat polymorphism downstream of the CMA1 gene with bronchial asthma in a group of Egyptian children. The findings suggest that the alleles, genotypes and haplotypes investigated are possible important determinants of asthma susceptibility and are probably involved in regulating IgE levels in atopic asthma.

RHINITIS TREATMENT

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Efficacy of mometasone furoate nasal spray-MFNS- in the treatment of allergic rhinitis. Meta-analysis of randomized controlled trials

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Objective: To assess the efficacy of MFNS in the treatment of allergic rhinitis.

Study Selection: Randomized, placebo-controlled and double blind trials that studied the effects of MFNS in patients with allergic rhinitis. Comprehensive searches of the EMBASE, LILACS, Cochrane Library and MEDLINE databases from 1966 up to January 2007 and references of identified articles and reviews.

Outcomes: Different outcomes measured in the active treatment and control groups were considered. Review Manager 4.2.8 Program (Cochrane Collaboration) was used for data synthesis. Outcomes were extracted from original articles. If information was not available, authors of each trial were contacted. Some graphics were digitalized. The analysis included the calculation of standardized mean difference (SMD). There was significant heterogeneity among the study results, because of differing study methodologies. Random-effects model was used.

Results: The initial scanning identified 91 articles, 26 of which were potentially relevant trials on the use of mometasone for the treatment of allergic rhinitis. 14 studies were randomized controlled trials and met inclusion

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Study	num. of patients	TSS mometasone mean (SD IC 95%)	num. of patients	TSS placebo mean (SD IC 95%)	Weight %	SMD
Meltzer JACI 1998	80	4.36 (0.70)	41	5.99 (1.90)	8.11	−1.31 (−1.72–0.89)
Stuck Allergy 2003	13	8.80 (7.60)	11	20.40 (14.80)	3.82	−0.98 (−1.84–0.12)
Bende Ann Allergy Asthma Immunol 2002	103	2.00 (0.95)	104	3.54 (1.10)	9.81	−0.91 (−1.20–0.62)
Graft JACI 1996	114	1.10 (0.90)	101	2.40 (2.30)	9.93	−0.76 (−1.04–0.48)
Hebert Allergy 1996	122	3.30 (2.85)	110	5.20 (4.65)	10.14	−0.50 (−0.76–0.24)
Bronsky Ann Allergy Asthma Immunol 1997	94	7.00 (5.50)	95	8.90 (6.90)	9.81	−0.30 (−0.59–0.02)
Drouin Ann Allergy Asthma Immunol 1996	129	3.90 (2.45)	124	4.60 (2.29)	10.32	−0.29 (−0.54–0.05)
Mandl Ann Allergy Asthma Immunol 1997	181	4.41 (3.50)	184	5.53 (4.50)	10.84	−0.28 (−0.48–0.07)
Gawchik Ann Allergy Asthma Immunol 2003	122	5.73 (4.55)	123	6.21 (5.25)	10.29	−0.10 (−0.35–0.15)
Ciprandi Ann Allergy Asthma Immunol 2001	9	0.64 (0.44)	11	1.42 (0.56)	2.99	−1.46 (−2.48–0.45)
Frieri Ann Allergy Asthma Immunol 1998	11	0.50 (0.60)	10	1.16 (0.85)	3.55	−0.87 (−1.77–0.04)
Meltzer JACI 1999	135	4.30 (2.08)	136	5.49 (1.92)	10.38	−0.59 (−0.84–0.35)
I2 = 78.3%; Z = 5.76 (p < 0.00001)	1113		1050		100	−0.59 (−0.80–0.39)

criteria for the meta-analysis. All randomized clinical trials included 23 comparison groups and more than 20 different outcomes (individual and global). For efficacy assessment, 2163 patients were analyzed; 1113 received mometasone and 1050 placebo. MFNS was associated with significant reduction in total symptoms scores (SMD −0.59; 95% CI −0.80 to −0.39; P < 0.00001; heterogeneity I2 = 78.3%.).

Safety: For adverse events report, 1758 patients were included. 884 received MFNS and 874 placebo. No significant difference between MFNS and placebo was revealed (SMD 1.02; 95% CI 0.83 to 1.25; P = 0.86; heterogeneity I2 = 1.0%)

Conclusions: First among nasal CSs, MFNS efficacy achieved Evidence Ia in the treatment of allergic rhinitis.The frequency of adverse events is similar in both groups, when it was compared with placebo.

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Studies on the role of tumor necrosis factor alpha gene polymorphisms in the therapeutic response to intranasal UV phototherapy

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Tumor necrosis factor alpha (TNFα) is one of the key cytokines in UV-induced immunosuppression. We have shown that intranasal phototherapy with mixed UVA-UVB-visible light (mUV/VIS) is a new therapeutic tool for the management of allergic rhinitis. The aim of this study was to investigate whether the efficiency of rhinophototherapy is related to specific TNFα polymorphisms. Fifty-nine patients with a history of at least 2 years of ragweed-induced allergic rhinitis were treated during ragweed seasons in Szeged, Hungary. Rhinophototherapy was carried out 3 times a week for 2 weeks and continued by a one-week follow up period, or 3 times a week for 3 weeks with increasing doses of mUV/VIS. The total dose of mUV/VIS therapy was the same at the end of the study in both therapeutic regimens. The patients graded the following 4 symptoms of allergic rhinitis on a standard severity scale: nasal itching, nasal obstruction, rhinorrhea and sneezing referred to as total nasal score (TNS). The average improvement in TNS of all patients was evaluated using repeated measures ANOVA. Patients with more than 50 percent decrease of the average change in the TNS were considered as responders and with less than 50 percent decrease as non-responders. According to these parameters, 38 patients were considered to be responders (64%) and 21 patients were non-responders (36%). Blood sample was taken from all patients for single nucleotide polymorphism (SNP) analysis of TNFα. Three SNPs of the promoter region of TNFα gene were selected for polymerase chain reaction restriction fragment length polymorphism (PCR-RFLP) analysis. The genotypes and allele frequencies of selected SNPs' were compared between the responders and non-responders for rhinophototherapy.

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Allergic rhinitis: occurrence of sinusitis in "sneezers-runners" and "blockers"

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Background: Patients with allergic rhinitis can be categorized as "sneezers-runners" and "blockers". The presence of sinusitis is often overlooked in these patients. The occurrence of sinusitis was assessed in these two clinical presentations of allergic rhinitis.

Methods: Consecutive patients with allergic rhinitis, with skin allergy test positivity were enrolled and categorized into "sneezers-runners" and "blockers" as per their predominant symptoms based on ARIA guidelines. All were subjected to computed tomography (CT) of the paranasal sinuses and extent of sinusitis was staged as per Lund and Kennedy criteria. The presence of sinusitis was then correlated with their clinical symptoms.

Results: Of the 338 patients (220 males, 118 females), 134 (39.6%) were "sneezers-runners" and 204 (60.4%) were "blockers". Sinusitis was present in 257 (76%), 78 (58.2%) were "sneezers-runners" while 179 (87.7 %) were "blockers". Sinusitis was significantly higher in "blockers" (78/134 vs 179/204, P = 0.019). Furthermore, the occurrence of sinusitis was significant in "blockers" (179/204 vs 25/204, P = 0.000) but not so in "sneezers-runners" (78/134 vs 56/134, P = 0.12). The mean sinus CT score was significantly higher in "blockers" as compared to "sneezers-runners" (8.9 ± 7.6 vs 6.1 ± 5.6, P = 0.0014). The mean number of sinuses involved too was significantly more in "blockers" (6.54 ± 4.53 vs 4.5 ± 3.2, P = 0.0001).

Maxillary sinuses were the most commonly involved (214, 83.2 %). This too was significantly higher in "blockers" (168/179 vs 46/78, P = 0.029). Breathlessness (105/204 vs 38/134, P = 0.006) and posterior purulent nasal discharge (144/204 vs 44/134, P = 0.046) were significantly more in "blockers" while nasal/palatal itching (83/134 vs 71/204, P = 0.003) and ocular itching (76/134 vs 64/204, P = 0.003) were significantly higher in "sneezers-runners". "Blockers" with sinusitis had significantly higher nocturnal awakening (156/179 vs 10/25, P = 0.041), posterior purulent post nasal drip (138/179 vs 8/25, P = 0.032) and halitosis (112/179 vs 6/25, P = 0.035).

Conclusion: Patients with allergic rhinitis should be categorized as "sneezers-runners" and "blockers" since they have distinct clinical profiles. "Blockers" had significantly more sinusitis as compared to "sneezers-runners". Radiological severity of sinusitis too was significantly higher in "blockers". Presence of sinusitis can possibly lead to suboptimal control of allergic rhinitis.

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Evidence based treatment of allergic rhinitis

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In clinical practice, caring for patients generates many questions about diagnosis, prognosis, and treatment that challenge health professionals to keep up to date with the medical literature. One approach to meeting these challenges is to learn how to practise evidence based medicine (EBM).

According to EBM, conclusions from meta-analyses of randomized trial represent the most solid evidence source to assess the efficacy of a treatment. Meta-analysis is a statistical procedure that integrates the results of different independent studies pooled together, thus allowing a more objective appraisal of the evidence than traditional narrative reviews can do. Meta-analysis provides a quantitative estimate of treatment effects. Pharmacological treatment of allergic rhinitis represents a bench mark of therapeutic management. Practical guidelines need high quality proves of evidence in order to provide reliable recommendations for therapeutic management. Meta-analysis may represent the answer to this need, as a source of solid evidence. Cochrane Collaboration recommendations and Quorom Statements represent the gold standard of quality assessment.

At present only few systematic reviews of high methodological quality are available or in progress: evidence Ia of efficacy for antihistaminic treatment is provided by desloratadine which on 3108 patients showed a significant reductions in total symptoms scores, total nasal symptoms score and nasal blockage relief when compared with placebo; a meta-analysis confirms that also ebastine induces a greater decrease from baseline in mean rhinitis symptom scores than placebo (749 patients). Mometasone furoate is at present the only one nasal steroid which may benefit an evidence of effect Ia by means of an in-progress meta-analysis, whose preliminary results show in 2163 patients a significant reduction in total symptoms scores. According to a recent systematic review with pooled analysis, leukotriene receptor antagonists produce a small but statistically significant improvement in nasal symptom when compared with placebo (3924 patients). All drugs showed good safety profile.

Antihistamines, nasal steroids and antileukotrienes demonstrated evidence of efficacy in the treatment of allergic rhinitis; these tools should also demonstrate a good benefit/cost ratio in order to satisfy the future methodological approaches of guidelines formulation (GRADE).

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Leukotriene receptor expression in allergic rhinitis: the effect of allergen exposure and treatment

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Background: Cysteinyl leukotrienes are mediators contributing to the symptoms of allergic rhinitis and exerting their effect through specific receptors. We aimed to evaluate the effect of a naturally occurring grass pollen season and the anti-rhinitis treatment on the expression of cysteinyl leukotriene receptor 1 (CysLT(1)R) in the nasal mucosa of allergic rhinitis patients.

Methods: In a double-blind randomised study, 59 patients with allergic rhinitis received treatment with either nasal steroid (fluticasone propionate; FP, 200 microg/day), oral antileukotriene montelukast (ML, 10 mg/day) alone or in combination with antihistamine loratadine (LT, 10 mg/day), or placebo over the grass pollen season. Nasal biopsies for immunohistochemical CysLT(1)R analysis were taken before and during the peak of the grass pollen season.

Results: The grass pollen season induced a significant increase in the CysLT(1)R expression in placebo (from 0.097 ± 0.005 to 0.178 ± 0.012 cells/field), ML (from 0.113 ± 0.011 to 0.148 ± 0.009), and ML + LT (from 0.122 ± 0.008 to 0.154 ± 0.016), but not in FP (from 0.123 ± 0.013 to 0.128 ± 0.016) treated patients. FP treated patients had significantly lower pollen-season induced change in the CysLT(1)R expression compared to other groups. Also, both ML and ML+LT treatment groups had significantly lower increase in the CysLT(1)R expression compared to placebo treatment.

Conclusion: We conclude that the inhibitory effect on CysLT(1)R expression is one of the mechanisms by which nasal steroids and antileukotrienes alone or together with antihistamines exert their anti-inflammatory effect in allergic rhinitis with nasal steroids demonstrating the strongest inhibition.

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Efficacy of desloratadine in the treatment of allergic rhinitis: a meta-analysis of randomized, double-blind, controlled trials	G. W. Canonica. Allergy 2007;62:359–366	symptoms scores (SMD −1.63; 95% CI −2.75 to −0.51; P = 0.004) total nasal symptoms score (SMD −0.66; 95% CI −0.91 to −0.42; P < 0.001) nasal blockage relief (SMD 0.32; 95% CI 0.10 to 0.55; P = 0.005)
Meta-Analysis of the Efficacy of Ebastine 20 mg Compared to Loratadine 10 mg and Placebo in the Symptomatic Treatment of Seasona	Paul Ratner et al. Int Arch Allergy Immunol 2005;138:312–318	LS mean TSS (−1.30 (0.16) −1.61 to −0.99; p < 0.0001)
Efficacy of Mometasone Furoate Nasal Spray-MFNS-in the treatment of allergic rhinitis. Meta-analysis of randomized controlled t	Penagos M. In progress	(SMD −0.59; 95% CI −0.80 to −0.39; P < 0.00001; heterogeneity I2 = 78.3%)
Leukotriene Receptor Antagonists for Allergic Rhinitis: A Systematic Review and Meta-analysis	A. M. Wilson. The american journal of medicine. volume 116; 2004	NS percentage of maximum score (WMD −5%; 95% CI −7% to −3%; P = 0.01 for heterogeneity)

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Sputum eosinophils negatively correlate with lung functions in non-asthmatic rhinitis patients

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Background: Sputum eosinophilia is frequent findings in non-asthmatic rhinitis patients. In asthma, sputum eosinophilia is known to be related to airway remodeling and thus reduced pulmonary functions. However, clinical significance of sputum eosinophilia in non-asthmatic rhinitis patients has not yet been determined.

Objective: To evaluate the clinical significance of sputum eosinophil counts in the pulmonary function of non-asthmatic rhinitis patients.

Methods: A retrospective review of medical records of 89 moderate-to-severe perennial rhinitis patients without respiratory symptoms was performed. All of them underwent lung function tests, skin prick testing to common inhalant allergens, methacholine bronchial challenge tests, and sputum induction with hypertonic saline to examination the eosinophil counts. Patients were divided into two groups according to the presence of sputum eosinophilia (≥2%). Patients who were taking systemic steroid, who had past history of destructive lung disease and who had asthma or airway hyperresponsiveness were excluded. Stored sputum samples were used to determine mRNA levels of TGF-β, MMP-9 and TIMP-1.

Results: There was no significant differences in characteristics including atopy index, smoking and use of nasal corticosteroids between two groups. FEV1/FVC was significantly decreased in the group with sputum eosinophilia (98.5 ± 0.1 for positive and 103.0 ± 0.1 for negative sputum eosinophilia, p = 0.015). Sputum eosinophil counts were negatively correlated with predicted percent of FEV1 and FEV1/FVC (R square = .063, p = .017 for FEV1 and R square = .009, p = .004 for FEV1/FVC). The mRNA levels of TGF-β and TIMP-1 between two groups showed no significant difference (p = .366). However, the mRNA level of MMP-9 was significantly increased in subjects with sputum eosinophilia (p = .035). The ratio of MMP-9 and TIMP-1 mRNAs was also significantly increased in subjects with sputum eosinophilia (p = .013).

Conclusion: Sputum eosinophilia in non-asthmatic moderate-to-severe rhinitis patients may affect lung function via airway remodeling.

MECHANISMS OF ASTHMA II

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Maternal asthma enhances umbilical cord blood leptin expression

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Background: Maternal asthma is a strong risk factor for asthma susceptibility in their children. Although this familial inheritance is a well-known phenomenon, the extent and exact mechanisms leading to this immunomodulation in mother/child pairs are poorly understood. The present cross-sectional study investigated the *in utero* priming of fetal allergic inflammatory responses in relation to the presence of maternal asthma.

Methods: Pregnant women with personal history of asthma and those without any atopic disorder were recruited during antenatal visits. EDTA-anticoagulated venous blood was collected from these mothers intrapartum, and also from umbilical veins of their babies postpartum but before placental separation. The protein signatures of 79 inflammatory mediators from these paired serum samples were analyzed using RayBio® Human Cytokine Antibody Array V. Serum levels of leptin, being differentially expressed in umbilical cord blood (UCB) from those with maternal asthma, and C-reactive protein (CRP) were measured by ELISA assays. The respective lower detection limits for these markers were 125 ng/L and 0.1 mg/L.

Results: Six pregnant mothers with asthma and 6 non-allergic controls were recruited, with respective mean (SD) age being 30.7 (5.9) years and 35.1 (4.9) years. Five of 6 UCB samples from newborns with maternal asthma had increased leptin levels when compared with newborns with non-allergic mothers. Other mediators with altered levels in two or more UCB samples with maternal asthma included IL-3, IL-8, IL-12, IL-15, PDGF-BB, NAP-2, GRO and MIG. Serum leptin and CRP levels from asthmatic mothers were significantly higher than those in UCB from their newborns ($P < 0.001$), so were serum CRP levels in non-allergic mothers as compared with their newborns ($P < 0.01$). Leptin levels showed significant positive correlation with CRP levels ($r = 0.712$, $P < 0.001$) in sera from asthmatic mothers. On the other hand, CRP was below the detectable limit in most UCB serum samples. Serum leptin levels in UCB did not differ in those with and without maternal asthma.

Conclusion: Maternal asthma may upregulate leptin-related pathway in UCB of their high-risk newborns, which may predispose them to develop early allergic manifestations.

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Promoter polymorphisms of cysteinyl leukotriene type 1 receptor in patients with different asthma phenotypes

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Background: Cysteinyl leukotrienes (cysLTs) are potent inflammatory mediators acting through at least two receptors $CysLTR_1$ and $CysLTR_2$. $CysLTR_1$ mRNA and protein expression is upregulated in patients with severe asthma phenotypes: frequent severe exacerbations, chronic airflow limitation or steroid-resistant asthma. Changes in the $CysLTR_1$ promoter might influence its transcriptional activity and therefore might modulate airway inflammation.

Aim: Investigate an association of polymorphisms in the $CysLTR_1$ gene promoter with severe and non-severe asthma phenotype.

Methods: The study groups consist of 100 patients with severe asthma, 70 patients with non-severe asthma, diagnosed according to GINA 2005 criteria and 100 healthy subjects. Genomic DNA was isolated from the peripheral blood leukocytes. Fragments of the $CysLTR_1$ promoter region were amplified by PCR and sequenced directly.

Results: Three single nucleotide polymorphisms (SPSs) -634 T/C (rs321029), -475C/A (rs2637204), -336G/A (rs2806489) were identified in the promoter region of $CysLTR_1$ gene. All SNPs were in strong linkage disequilibrium both in patients and in control group. Two most common haplotypes were identified: ht1 [C-A-A] and ht2 [T-C-G], among which ht1 was observed in 71.43% of male patients and 90.48% of male controls. Significant differences were observed in allele frequencies of -336G/A in male subjects. -336G allele occurred significantly more often in men with severe asthma comparing with male control group ($p_{cor} = 0.04$; OR = 3.800 C.I. = [1.112–12.983]).

Conclusion: Genetic variants of $CysLTR_1$ gene promoter might be associated with severe asthma phenotype in male asthmatic patients. Further elucidation of this association might improve understanding of heterogeneous condition in severe asthma.

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Inflammatory changes in upper respiratory tract in patients with aspirin sensitive asthma

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Background: Patients with aspirin- sensitive asthma often have continious inflammation of their upper sinuses complicated by chronic rhinitis, chronic sinusitis and nasal polyposis which frequently leads to asthma attacks. These patients often are not aware of the problems in their upper respiratory tract. Our goal with this study was to evaluate the inflammatory changes in upper respiratory tract in patients with aspirin sensitive asthma (ASA).

Methods: We followed 88 patients with paranasal x-ray, CTscan, skin prick tests, pulmonary function test, rhynomanometry and chest x-ray. Patients were divided in 2 groups. 39 pts with NSAIDs intolerance of ~15yrs were included in the first group (mean age 45, 8 ≥ 4,7yrs). In the second group were 49 aspirin intolerant asthma pts (mean age 44, 8 ≥ 4, 1yrs). The diagnosis of asthma was made based on clinical evaluation, skin prick test and pulmonary function test including histamine provocation test. Endoscopic methods were used for assesment of paranasal sinuses and nasal passages.

Results: Endoscopic examination showed in group 1 nasal polyposis in 8, deviation of nasal septum in 16, hypertrophy of nasal mucosa in 6, and chronic tonsilitis in 4 pts. In 16 pts we have observed changes in x-ray of paranasal sinuses and in 12 changes in CTscans of PNS. In 13 pts- rhynomanometric evaluation showed decrease in of the nose. pathways Bacteriological analysis of nose and throat showed predominance of bacteria species *Moraxella influenze* in 33,3%, *Moraxella cataralis* in 28,2% and *Stahylococcus aureus* in 15,3%.

Conclusion: Our study and results confirm the coexistence of chronic inflamation in upper respiratory tract in pts with bronchial asthma regardless of it's type.

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Role of the RNA-binding protein HuR in posttranscriptional regulation of IL-13 in T cells

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Rationale: Posttranscriptional regulation (PTR) is a key yet ill-defined area of gene expression in T cells. We investigated the PTR of interleukin (IL)-13 and the role the RNA-binding protein HuR plays in this process. HuR associates with adenylate-uridylate-rich elements (ARE) in the 3'untranslated regions (UTR) of mRNAs, promoting mRNA stability and translation.

Methods: IL-13 mRNA decay was monitored in human peripheral blood-derived Th2 cells using the transcriptional inhibitor Actinomycin D. The IL-13 3'UTR was subcloned into a tetracycline (Tet)-off β-globin reporter construct and transiently expressed in the absence or presence of overexpressed HuR in H2 cells. Association of HuR with IL-13 mRNA was detected by immunoprecipitation (IP) of messenger ribonucleoprotein complexes (mRNPs) and by biotin pull-down.

Results: IL-13 mRNA half-life increased in cells treated with PMA and ionomycin compared to unstimulated cells (6.5 ± 2.0h vs 1.6 ± 0.1h, $p < 0.05$, $n = 3$). The decay of the β-globin transcript was faster in cells transfected with pTet-BBB-IL13 compared to those transfected with the ARE-less pTet-BBB (2.1 h vs 11.4 h, $p < 0.05$, $n = 6$). Overexpression of HuR increased β-globin mRNA half-life in cells transfected with pTet-BBB-IL13 (22.7 h vs 2.3 h, $p < 0.01$, $n = 4$). Enrichment of IL-13 mRNA was detected by PCR following IP of Jurkat cell mRNPs with anti-HuR vs a control antibody. HuR binding to IL-13 3'UTR was confirmed by pull-down assay using biotin-labeled RNA probes spanning the 3'UTR or the coding region of IL-13.

Conclusion: PTR plays a relevant role in expression of IL-13. HuR-mediated stabilization of IL-13 mRNA could be a relevant therapeutic target in asthma.

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Functional study of prostaglandin D2 receptor (chemoattractant receptor molecule expressed in th2 cells, CRTH2) promoter polymorphism with aspirin-intolerant asthma

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Background and Objective: Prostaglandin D2 (PGD2) is an important lipid mediator in the pathogenesis of eosinophilic airway inflammation via its receptor CRTH2. The human CRTH2 gene encodes a G protein-coupled chemoattractant receptor which is expressed on Th2 cell, eosinophil, basophil and monocyte. The main objective of the study is to investigate the association of CRTH2 promoter polymorphism in patients with aspirin intolerant asthma (AIA).

Subjects and Methods: The case control study was performed in three groups of patients, 106 AIA, 115 aspirin tolerant asthma (ATA) and 133 normal controls (NC). Two promoter polymorphism of CRTH2 gene were genotyped by a primer extension method, which was performed with the SNaPshot ddNTP primer extension kit (Applied Biosystems, CA, USA).

Results: Two novel single- nucleotide polymorphisms (SNPs), CRTH2-466T>C and -129 C>A were identified in the promoter region of gene. Among two SNPs, the polymorphism of CRTH2 at positions of -466T>C showed a significant difference in genotype frequency between AIA and ATA; AIA patients showed significantly higher frequency of homozygous TT genotype than combined homozygous CC and heterozygous CT genotype of CRTH2 -466T>C ($p = 0.037$, multiple logistic regression analysis controlling for age and sex, recessive model). The luciferase reporter assay showed that the reporter plasmid carrying the -466C allele showed significantly higher promoter activity than -466T allele in human monocytes cell line (U937) and human mast cell line (HMC-1) ($p < 0.001$and $p < 0.001$ respectively). Similarly, the promoter activity of CRTH2 -466C allele cotransfected with GATA-3 was found to be increased dose dependently from the baseline promoter activity in HMC-1 cells.

Conclusion: These results suggest that GATA-3 may be directly or indirectly associated with CRTH2 -466C allele by stimulating mast cell activation with the release of Th2 cytokines and other proinflammatory mediators contributing the increased susceptibility of AIA in CRTH2 -466T>C promoter polymorphism.

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The relationship between early infections due to RSV and the specificities of subsequent development of asthma

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Clinical studies of RSV- infected patients indicated increased levels of Th2 cytokines and IgE, suggesting that an allergy-like condition developed during infection and skew the Th1/Th2 balance toward Th2 > Th1. Comparative retrospective analysis of 76 children, who been admitted between winter seasons 2001 and 2005 for a first attack of brochiolitis before the age of 12 months, was performed in order to elaborate and correlate possible specificity of asthma in term of pathogenesis (A, NAA), immune response and compliance to treatment. In the 3 groups of children, divided on the basis theirs phenotypes: I group (n = 21) IgE associated persistent wheezier, II group (n = 47) nonatopic recurrent wheeze, III (n = 8) viral associated wheeze, RSV infection was responsible for the first attack of wheezing in 98%, 2% has early adenovirus infections in the II group. Family history of asthma and multiple asthmas triggers was performed in 95% in I (bilateral genetic risk was funded

in 18%) comparing with 3% from II group. In opposite, many risk-factors in prenatal, peripartal and early neonates period of life in the present of serious another illness, was found in the II group ($p < 0,05$): oligohydramnion, placentas abruptions and infarct, small for date with extremly LBW 800 ± 57g, gigant baby with extremely higher birth weight: 5250 ± 50g, low Apgar score and reanimation/adaptation problems after birth. Patients in I group demonstrated higher levels of Neu, Eo, moderate degree of diseases and recurrent wheezing and/or asthma. Severity of the illness, many immune, endocrine, metabolic and genetics deregulations, ly-cytosis and Mo-cytosis were typical for II group. Compliance to ICS was significantly better in the I group. On the basis these data we may indirectly conclude implicate excess type 2 and/or deficient type 1 immune responses in the pathogenesis of RSV bronchiolitis and subsequent asthma development in I group. In contrast, II group had probably higher Th1 immune responses during RSV infections, on the basis maternal and intra-uterine factors, and has more severe disease and had shown higher percentage of immune deregulations. Patients in II group have attributes specific for NAA and complex immunoregulatory pattern followed by inappropriate response to treatment. These results indicate that immunological background via CD8+T cells may play an important role in the regulation of the differentiation and activation of the Th2CD4+Tcells during RSV infection.

CLINICAL IMPLICATIONS OF AEROALLERGENS

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Indoor air quality and dyspnea, mucosal, dermal and general symptoms in relation to room temperature and ventilation in university computer classrooms -an experimental study

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The aim was to study effects of increased ventilation and temperature changes in computer classrooms on measured indoor air quality and medical symptoms in university students. Technical university students in four classrooms participated in a blinded study. Two classrooms had higher air exchange (4.1–5.2 ac/h); two others had lower air exchange (2.3–2.6 ac/h). After one week, ventilation conditions were shifted between the rooms. The students reported medical symptoms last hour, on a seven step rating scale. Room temperature, RH, CO2, PM10, and ultra fine particles were measured simultaneously during 1 hour. In addition, illumination, air velocity, operative temperature, supply air temperature, formaldehyde, NO2, O3 and airborne levels of bacteria, moulds and common allergens was measured. Totally 355 students participated at least once in the study during the two weeks , 121 participated twice in the longitudinal analysis. In total, 31% were females, 2.9% were smokers, 3.8% had asthma, 18.1% pollen allergy (hay fever), and 14.0% furry pet allergy. Mean CO2 was 993 ppm (674–1450 ppm), temperature 22.7 C (20–25 C), and RH 24% (19–35 %). Low and high air exchange rate corresponded to a personal outdoor air flow of 7 L/s*p and 10–13 L/s*p, respectively. Mean PM10 was 20 microgram/m3 at low and 15 microgram/m3 at high ventilation flow. In the crude analysis, ocular, nasal and throat symptoms, dermal symptoms, dyspnea, sinusitis, headache, tiredness and nausea was significantly more common at higher CO2 levels and higher room temperature. At higher air exchange rate dyspnea and dermal symptoms were significantly less common. Similar results were obtained in the multivariate analysis, controlling for potential confounders such as gender, smoking and a history of atopy. In the longitudinal analysis of a subset participating twice, increased temperature was significantly related to increase of tiredness. In conclusion, computer classrooms may have CO2 levels above the current ventilation standards (more than 1000 ppm) and temperatures above 22 C, due to crowdedness and high thermal load. Increased temperature and CO2-levels may affect both dermal and mucosal membrane symptoms, as well as headache and tirdness. It is recommended that the personal ventilation flow is at least 10 L/s.

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Allergenicity of the recombinant and native blomia tropicalis allergens among atopic subjects

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Background: Blomia tropicalis (Bt), a predominant house dust mite (HDM) species in tropical and subtropical regions of the world, is a source of multiple allergens causing allergic sensitization among atopic individuals. Isolation and characterization of the allergenicity these Bt allergens are essential to evaluate the clinical relevance of this HDM species.

Methods: Eight recombinant Bt allergens were expressed as GST-fusion allergens in E. coli or secreted allergen in P. pastoris while two native Bt allergens were purified by immuno-affinity chromatography. Specific-IgE reactivity of the Bt allergens were determined using 192 Bt-allergic and 85 non-atopic sera. The panel of Bt allergens were likewise used for skin prick test (SPT) in 110 Bt-allergic pediatric patients. The ability of the recombinant and native Blo t 11 allergens to inhibit the IgE-binding activity in Bt extract was determined by absorption studies.

Results: Blo t 1, Blo t 3, Blo t 5, Blo t 10, Blo t 11-fD, Blo t 12 and Blo t 19 were expressed as soluble GST-recombinant proteins in E. coli while Blo t 4 was expressed in P. Pastoris. Native Blo t 5 and Blo t 11 were purified from Bt extracts using monoclonal antibodies. Of the 192 Bt-allergic patients' sera tested, 68% and 53% reacted positively to recombinant Blo t 5 and Blo t 11-fD, respectively, while 5-30% IgE reactivity was observed with the other recombinant Bt allergens. Interestingly, the IgE reactivity recombinant Blo t 5 and Blo t 11-fD compares with the IgE reactivity of their native counterparts at 70% and 54%, respectively. SPT results showed comparable results with ELISA. Absorption assays, showed that up to 80% inhibition of IgE reactivity in Bt extract can be obtained with the different Bt allergens.

Conclusion: Results obtained from this study suggest the clinical importance of Blomia tropicalis as major source of allergens causing allergic sensitizations in tropical regions. The incorporation of Bt allergens in the panel of diagnostic and immunotherapeutic allergens for HDM allergy are highly recommended.

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Phleum pratense alone is representative for pooideae grass pollen species due to high immunochemical similarity between homologous grass pollen allergens

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Background: The Pooideae grasses constitute a large taxonomical subfamily with thousands of species and worldwide occurrence. Extensive homology between amino acid sequences of grass pollen allergens occur, and grass pollen counts are published together due to a large immunochemical similarity between the grass pollen allergens.

Objective: Study the immunochemical similarity of grass pollen allergens from different Pooideae species.

Methods: Amino acid sequences representing grass pollen allergens were obtained from the Uniprot database. Structural similarity of the group 1 allergens was illustrated by mapping identical amino acids on the surface of the x-ray structure of Phl p 1, PDB: 1N10. A large number (>13,000 data points) of grass pollen allergic patients' IgE was measured by Magic Lite solid phase immunoassay to eight Pooideae grass pollen extracts, respectively. IgE to eight Pooideae grass pollen extracts was measured by ADVIA Centaur solid phase immunoassay with or without inhibition by 2 mg *Phleum pratense* pollen extract. Standard T cell stimulation assays using T cell lines from grass pollen allergic donors were applied to assess T cell cross-reactivity.

Results: Sequence alignment of Pooideae grass pollen allergens showed high homology. Mapping of identical amino acids on the surface of the x-ray structure of Phl p 1 clearly showed the presence of identical surface structures

large enough to harbor IgE binding epitopes. A high correlation (0.86–0.98 Spearman rank correlation coefficient) was observed when comparing levels of IgE to *Phleum pratense* with those of pollen extracts from individual grass species. *Phleum pratense* pollen extract inhibited IgE to other grass species more than 95% in most patients. T cell lines specific for Phl p 1 and Phl p 5 both showed similar stimulation indices when stimulated with different grass pollen extracts indicating extensive T cell cross-reactivity between individual grass species.

Conclusion: *Phleum pratense* alone covers most of the immunochemical reactivity of all tested Pooideae grass species with respect to allergic patients' IgE. T cell lines specific for the group 1 and 5 major allergens showed extensive cross-reactivity between grass species. One specie alone, e.g. *Phleum pratense*, therefore seems adequate for specific management of allergy to Pooideae grass pollens.

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Gradual shift in the aeroallergen index affecting the allergy patients in recent years in Texas panhandle

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Aeroallergens cause serious allergic and asthmatic reactions. We have been analyzing the Aeroallergen data of the Texas Panhandle region for the last 7 years. We determined the aeroallergen index regularly by analyzing the coated Melinex tape from the Burkard Volumetric Spore Trap. Samples were examined, counted and photographed every 24 hours using a BX-40 Olympus microscope with a DP-70 Olympus Digital Camera. Data were correlated with daily temperature, precipitation, peak wind speed and clinical studies established from Allergy A.R.T.S. Clinical Research Laboratory in an effort to aid in diagnosis of mold and pollen-related allergies. Aeroallergens that we recorded were *Alaternaria* conidia, Pezizales ascospores, *Curvularia, Cladosporium, Dreschlera, Stachybotrys*and pollen from short ragweed (*Ambrosia artemisiifolia*), grass (Poaceae), hairy sunflower (*Helianthus hirsutus*), buffalo bur (*Solanum rostratum*), purple nightshade (Solanum elaeagnifolium) and lamb's quarters (*Chenopodium album*). Due to severe drought conditions in the previous years (2002–5), the pollen concentration was significantly low, that reached the highest peak from April onwards this year (2007) with plenty of rainfall. We observed this pick from April to July in last year 2006. We noticed a gradual shift in the aeroallergen index in the Texas Panhandle in recent years. Global warming with increased CO2 concentration exerted widespread impacts on the biotic system. Many regions are currently experiencing warming effect associated with global climate change including longer growing seasons and early arrival of spring. Short ragweed (*Ambrosia artemisiifolia*) released 54.8% more pollen with an ambient CO2 release (Rogers et al, 2006). In Japan a 21-year study showed a gradual shift in *Cryptomeria japonica* pollen season. These results support the steadily increasing number of reports indicating a global warming trend. The temperature change affecting the start dates of the *C. japonica* pollen season is particularly relevant in the context of human health. From the clinical data from the AARTS clinic it is clearly evident that there were more patients suffering from allergic rhinitis during the months of March to June. 4 years back the peak pollen and mold season was between May to September (Ghosh et al., 2006) that has gradually shifted in 2007 to March-June so as the frequency of the patients visiting the Allergy Clinic.

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Upper body clothing is a source of fungal β-(1,3)-glucan exposure

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Background: β-(1,3)-glucan is pro-inflammatory and has been associated with airway inflammation and respiratory symptoms.

Methods: This study assessed β-(1,3)-glucan levels from upper body clothing (jerseys) of 55 subjects. β-(1,3)-glucan levels were estimated with a modified Limulus amoebocyte lysate kinetic assay.

Results: β-(1,3)-glucan levels ranged widely from 2,697 to 162,690 ng/g. β-(1,3)-glucan levels were significantly lower from cotton jerseys, and from warm water washed jerseys.

Conclusion: Jerseys are potentially a significant exposure source of β-(1,3)-glucan.

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Development of a natural cat allergen challenge room (CACR) model

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Background: We have developed a natural CACR in which subjects will be exposed to consistent levels of Fel d 1, the major cat allergen, in a natural setting, such that there is transient exposure to disturbance of reservoir cat allergen.

Objective: To develop the CACR in which Fel d 1 levels range from approximately 500–2500 ng/m^3. This concentration range of Fel d 1 was selected as it is representative of higher levels of Fel d 1 in homes with cats, and is known to induce an allergic and/or asthmatic response.

Methods: The CACR is fully carpeted, contains a couch covered with a cotton sheet, and a chair. The CACR was preloaded with Fel d 1 using a proprietary aerosol generator and maintained by allergen from two CACR inliving cats. Fel d 1 levels were transiently increased by vigorously shaking the sheet to aerosolize Fel d 1. Fel d 1 airborne concentrations were measured using personal air samplers. Total airborne particulate was collected using a Button Sampler for 1h. Samples were analyzed using ELISA and particle sizes were independently measured with a laser particle counter.

Results: Background measures indicated that Fel d 1 was undetectable in the CACR before aerosolization. Following one hour of 1g cat allergen aerosolization and 11 days cat housing, the average maximum total particle count was maintained over 4 consecutive days to be 9.01 ± 3.65X105 ppm^3 and the Fel d 1 concentration 538 ± 192 ng/m^3. After an additional 2h aerosolization of 3.2g Fel d 1 and a total of 15 cat housing days, the average maximum total particle count maintained over an additional 4 consecutive days was 9.8 ± 3.65X105 ppm^3 and the average Fel d 1 concentration was 621 ± 135 ng/m^3. Following an additional two days the concentration of Fel d 1 was 1046 ± 982 g/m^3. Overall, these two periods of aerosolization of Fel d 1 and a total of 20 cat housing days resulted in the mean concentration of Fel d 1 to be 686 ± 455 ng/m^3.

Conclusion: These data indicate that aerosolized Fel d 1 is effective to obtain natural allergen levels rapidly and that live cats can maintain these levels over time within the targeted range. This work demonstrates that we have developed a CACR model that can be used towards the study of the etiology and putative therapeutics for cat allergy and asthma. Funding provided by Allied Research-Cetero Research, CA.

IMMUNOTHERAPY–MECHANISMS

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Drug-induced T regulatory cells in asthma: Toll like receptors 2 and 9 represent distinct markers of glucocorticoid versus 1?25-dihydroxyvitamin D3 action on human CD4+ T cells

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Background: We previously demonstrated that the active form of Vitamin D, 1?25-dihydroxyvitamin D3 (1?25VitD3) alone, or together with glucocorticoids, induces an IL-10 secreting T regulatory phenotype (IL-10-Treg) in human CD4+ T cells, and these cells inhibit naïve T cell, Th1 and Th2 responses. Glucocorticoid induced IL-10 production is defective in patients with clinically insensitive asthma and 1?25VitD3, either in vitro or following oral ingestion by these patients, restores this impaired IL-10 response.

Aim: To identify the distinct phenotypic and functional effects of the two drugs on human CD4+ T cells.

Methods: In vitro based tissue culture, quantitative RT-PCR, flow cytometry and antibody capture assays (ELISA, cytometric bead array) were used to study human CD4+ T cell phenotype and function following polyclonal activation (anti CD3, IL-2) in the presence or absence of dexamethasone and/or 1?25VitD3. T cell phenotype following 1?25VitD3 ingestion was also analyzed.

Results: We show that 1?25VitD3 alone inhibits Th1 and Th2 cytokine production, enhances the generation of IL-10-Treg, but also the percentage of FoxP3 positive cells. However expression of 1?25VitD3-induced IL-10 and FoxP3 appears mutually exclusive. Furthermore, optimal effects of 1?25VitD3 on IL-10, Foxp3 and effector cytokine production are observed at distinct drug concentrations. Dexamethasone selectively induced expression of TLR2 in vitro whilst 1?25VitD3 selectively induced expression of TLR9 on CD4+ T cells both in vitro and following patient ingestion of 1?25VitD3, in comparison to other T cell populations (naïve, CD25+ Treg, Th1, Th2). Ligation of these TLR on the drug-induced IL-10-Treg resulted in loss of Treg function in both cases, but by distinct mechanisms.

Conclusion: Glucocorticoids and 1?25VitD3 promote IL-10 production and regulatory function in human CD4+ T cells alone and in combination. TLR2 and TLR9 can be used to distinguish glucocorticoid and 1?25VitD3 exposure of human CD4+ T cells and ligation of these receptors downregulates Treg function.

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Sublingual immunotherapy to inhalant allergen sensitisation and the effect of chitosan

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Background: Immunotherapy is an established form of treatment but it is necessary to develop more efficacious methods. We have developed a model for respiratory sensitisation induced by intranasal (i.n.) administration of the cysteine protease papain, without adjuvant. It produces high, boostable IgE titres. Upon challenge pulomonary eosinophilia is evident as is the release of Th2 cytokines into the bronchoalveolar lavage (BAL) fluid. The model has been used to optimise sublingual desensitisation in mucosally sensitised animals. We have previously demonstrated its ability to suppress allergen specific IgE. In these studies lymph nodes were examined and the effect of the excipient chitosan.

Methods: Allergic sensitisation was achieved by the i.n. administration of low doses of papain, a homologue of Der p 1. Desensitisation was examined after the sublingual administration of the antigen with chitosan, antigen alone, or chitosan alone. Serum antibodies were examined and high dose challenge followed by BAL was used to study cellular infiltrates and cytokine production in the lung. Lymph nodes were cultured and proliferation and cytokine production in response to allergen stimulation measured.

Results: All treatments, even chitosan alone, suppressed IgE and IgG1 antibodies. Chitosan delivery at desensitisation resulted in a significant reduction of cells infiltrating the lungs that was not seen in the other treatment groups however both the chitosan alone and chitosan with papain significantly suppressed the neutrophilia seen. The Th2 cytokines in BAL fluid were not reduced in the treatment groups although all groups receiving chitosan had elevated levels of IL-10 and IFN-γ. Cultured lymph node cells had lower levels of proliferation after sublingual therapy with a soluble antigen but when chitosan was used cells proliferated to similar levels to controls. Cytokine levels in lymph node cell supernatants showed significantly increased levels of TGF-β and significantly decreased IL-5, IL-10 and IFN-γ in all treatment groups.

Conclusion: Sublingual immunotherapy decreased IgE and IgG antibodies but not lung inflammatory responses. It is however shown that lymph node proliferation and cytokine production was decreased and this could have a long-term effect. Chitosan caused a non-specific reduction that was not greater than allergen alone.

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Analysis of epitope-specific immune responses induced by vaccination with structurally folded and unfolded recombinant Bet v 1 allergen derivatives in man

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Background: Previously we have constructed recombinant derivatives of the major birch pollen allergen, Bet v 1, with a more than hundredfold reduced ability to induce IgE-mediated allergic reactions. These derivatives differed from each other because the two recombinant Bet v 1 fragments represented unfolded molecules whereas the recombinant trimer resembled most of the structural fold of the Bet v 1 allergen.

Methods: Here we analyzed the antibody (IgE, IgG subclass, IgA, IgM) response to Bet v 1, recombinant and synthetic Bet v 1-derived peptides in birch pollen allergic patients who had been vaccinated with the derivatives or adjuvant alone. Furthermore, we studied the induction of IgE-mediated skin responses in these patients using Bet v 1 and Bet v 1 fragments.

Results: Both types of vaccines induced a comparable IgG1 and IgG4 response against new sequential epitopes which overlap with the conformational IgE epitopes of Bet v 1. This response was much higher than that induced by immunotherapy with birch pollen extract. Trimer more than fragments induced also IgE responses against new epitopes and a transient increase in skin sensitivity to the fragments at the beginning of therapy. However, skin reactions to Bet v 1 were reduced one year after treatment in both actively treated groups.

Conclusion: We demonstrate that vaccination with folded and unfolded recombinant allergen derivatives induces IgG antibodies against new epitopes. These data may be important for the development of therapeutic as well as prophylactic vaccines based on recombinant allergens.

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CD4+ CD25+ regulatory T cells in peripheral blood during the specific immunotherapy to grass pollen

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Background: Specific immunotherapy (SIT) is a well known procedure to achieve a clinical tolerance in IgE-dependent allergy. Recent data supports the concept that SIT restores the peripheral tolerance to allergens by reintroducing T cell tolerance. This study was an attempt to determine the effect of subcutaneous SIT on CD4+ CD25+ regulatory T cells in allergic patients.

Materials: Group of 34 subjects included: 19 individuals with allergy to grass pollen confirmed by medical history, positive skin prick tests (SPT) and specific IgE (sIgE) min. 2 class, treated with SIT before pollen season in 2007 (gr. A), 11 sex and age matched control subjects with confirmed as above allergy to grass pollen untreated with SIT (gr. B) and 4 control healthy nonallergic to grass pollen subjects (gr. C). Methods: i) subcutaneous SIT to grass pollen with allergen extract absorbed on aluminum hydroxide according to manufacturer's quidline in the gr. A; ii) Evaluation with questionnaires of clinical rhinoconjunctivitis symptom score during 2006 and 2007 pollen season (the end-point) in groups A and B; iii) The flow cytometry analysis of blood samples was performed at the beginning of SIT treatment, after reaching a maintenance dose and at the end-point in gr. A, and at the end-point in gr. B and C.
Results: 17 subjects from gr. A reached the end point phase of SIT without significant adverse events. Reduction of symptom score >50% in 6 subjects, 26–50% in 9 subjects and <25% in 3 subjects after SIT treatment was shown. In gr. B the symptom score in grass pollen season was comparing to Group A before the SIT treatment. The flow cytometric analysis showed: i) lower percentage of CD4+ CD25+ regulatory T cells in all allergic patients (gr. A-2,1%, gr. B-1,1%) versus the control (gr. C-3,4%) at the end point; ii) statistically non-significant percentage upward trend in CD4+ CD25+ regulatory T cell population during the course of SIT in treated with SIT groups.
Conclusion: Lower count of CD4+ CD25+ regulatory T cells in both groups of allergic patients suggests a dysfunction within this population might contributes to the pathology of allergy. The percentage upward trend in Treg population during the SIT treatment, accompanied by the improvement in symptom score, suggesting the maintenance of balance between Th1, Th2 and Treg populations is crucial for peripheral tolerance. The project will be continued to cover broader population and investigate other immunological parameters.

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Early suppression of basophil activation during allergen-specific immunotherapy by upregulation of histamine receptor 2

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Background: Most of the primary effects of allergen specific immunotherapy (SIT) are exerted on effector cells. However, there is surprisingly few information about the mechanisms by which SIT modifies and suppresses immune responses of basophils and mast cells in particular during the repetitive administration of increasing allergen doses in the built-up phase. One of the main mediators released by effector cells upon allergen challenge is histamine. The diverse immunoregulatory functions of histamine are based on the differential distribution of histamine receptors on immunocompetent cells. While histamine receptor (HR)1 decreases humoral immunity and increases cellular immunity, HR2 displays opposing functions, decreasing cellular immunity and mediating tolerogenic immune mechanisms. Therefore, the early desensitizing effect of SIT due to reprogramming of effector cell responsiveness to histamine via the modulation of the histamine receptor repertoire represents an exciting and plausible strategy.
Methods: Honey bee and wasp venom SIT was applied to 10 patients according to the rush protocol. The mRNA level of HR1, HR2 and HR4 in PBMC isolated from the peripheral blood of patients during the built-up phase of SIT (5 days) was evaluated by real time PCR. Histamine receptor mRNA levels in basophils enriched from the peripheral blood, which were triggered via IgE receptor were determined. Additionally, the activation state of basophils after IgE receptor cross-linking and treatment with ligands selective for HR2 as well as cAMP inducer forscolin, a second messenger of HR2, was analysed by flow cytometry and sulfido-leukotriene release assays.
Results: Significant increase of the HR2 mRNA level as well as the HR2/HR1 ratio was observed in PBMC of patients from the 3rd day of SIT. IgE receptor cross-linking of basophils from the peripheral blood with increasing anti-IgE doses led to the upregulation of HR2. The selective triggering of HR2 with specific agonist dimaprit or forscolin strongly suppressed IgE-receptor-induced activation of basophils in a dose dependent fashion.
Conclusion: HR2 mediates immunosilencing functions on IgE-receptor activated basophils. The up-regulation of HR2 expression on basophils in response to repetitive IgE-receptor cross linking as well as allergen challenge during the built-up phase of SIT might represent a key mechanism and early desensitization effect of SIT on the level of effector cells. *these authors contributed equally.

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Distribution of langerhans cells and mast cells within the human oral mucosa-new potential application sites of allergens in sublingual immunotherapy

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Background: Sublingual immunotherapy (SLIT) has been proven to be a safe and efficient alternative to subcutaneous immunotherapy (SCIT) in the treatment of allergic rhinitis. While antigen-presenting cells such as Langerhans cells (LC) are thought to play a major role in the effectiveness of SLIT, mast cells (MC) most likely account for the observed adverse reactions such as oral itching and sublingual edema. Since only little is known about LC and MC within the oral cavity, we investigated their distribution in search for alternative mucosal application areas with the highest density of LC and lowest presence of MC.
Methods: Different biopsies were taken simutaneously from mucosal tissue of the vestibular, buccal, palatinal, lingual, sublingual and gingival region (n = 10). Tissue was further processed for immunohistochemistry and flow cytometry. MC were detected by chymase, and LC by CD1a expression.
Results: The highest density of MC could be detected within the gingiva while the lowest density of MC was found within the palatinum and lingua. However, in the sublingual region MC were located within the lobe and duct of sublingual glands in a substantial number of individuals, which might explain swelling of sublingual caruncle in some SLIT patients. Considering LC, we could detect the highest density within the vestibular region followed by the region of bucca, palatinum and lingua. Interestingly, the lowest density of LC was located in the sublingual region. By flow cytometry, we investigated the expression of the high affinity receptor for IgE (FcεRI) which might play a central role in allergen uptake during SLIT. Thereby, we could detect the highest expression of FcεRI on LC of the vestibular region.
Conclusion: In view of our data, different mucosal application sites such as the vestibulum might represent an alternative region with potent allergen uptake especially in SLIT patients suffering from sublingual edema. Furthermore our data might serve as a basis in the development of new application forms for SLIT such as tablets or stripes.

MOLECULAR MECHANISMS OF ALLERGY

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Lactic acid bacteria inhibit IgE-induced mast cell activation

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Because probiotics were proposed to decrease the prevalence of allergy in susceptible individuals, we investigated whether they could affect IgE-induced mast cell secretory responses. To this end, mouse Bone Marrow-derived Mast cells (BMMC) were exposed to lactic acid bacteria. BMMC express high-affinity receptors for IgE (FcεRI). When sensitized with IgE antibodies and challenged with specific antigen, they release and secrete a variety of inflammatory molecules including granular mediators and enzymes (among which β-hexosaminidase), lipid mediators, cytokines (among which TNF-α) and chemokines. We checked first that none of the bacteria tested activate mast cells. Indeed, an incubation of BMMC for 20 min or 3 hr with bacteria (at a ratio of 1000 bacteria/cell) did not induced β-hexosaminidase release or TNF-α secretion, respectively. We then investigated whether a previous exposure of BMMC to bacteria would affect the subsequent IgE-induced biological responses of mast cells. We found that an overnight incubation of BMMC with one strain of bacteria (at a ratio of 1000 bacteria/cell), prior to sensitization with IgE antibodies, inhibited Ag-induced β-hexosaminidase release and TNF-α secretion. Live and gamma-irradiated bacteria were equally inhibitory. These bacteria did not decrease the expression of FcεRI and did not affect sensitization of BMMC with IgE. Inhibition required a direct contact between cells and bacteria. Inhibition were similar in BMMC from TLR-2/4-, MyD88-, and NOD2-deficient mice as in BMMC from WT mice, excluding TLR1, 2, 4, 5, 6, 7, 8 and 9, as well as NOD2 as being the responsible receptors. Preliminary works aiming at analyzing how bacteria interfere with FcεRI signaling indicated that: 1) several intracellular signaling molecules had a reduced expression, 2) both early and late FcεRI-dependent phosphorylation events were decreased, 3) IκB degradation was prevented, thus interfering with the nuclear translocation of NF-κB, 4) the IgE-induced increase of the intracellular concentration of Ca2+ was profoundly inhibited. Altogether, our results indicate that probiotics, especially lactic acid bacteria, can exert direct inhibitory effects on mast cell activation. They support the idea that probiotics can protect from allergies, by preventing IgE-induced mast cell activation.

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Induction of allergic airway inflammation by house dust mite allergen specific Th2 cells in mice

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Background: It is known that allergic inflammatory diseases such as asthma are Th2 cells-mediated, however, the pivotal roles of allergen-specific Th2 cells in the induction of allergic lung inflammation have not been fully elucidated. The aim was to elucidate the imunopathological roles of allergen-specific-Th2 cells in allergic airway inflammation.

Methods: Mice were epicutaneously sensitized with a major dust mite allergen, Blo t 5 and a well-characterized Blo t 5-specific-Th2 cell line was subsequently established from the splenocytes of the sensitized mice. The immunopathological roles of the cell line were assessed *in vivo* by adoptive cell transfer approach. Naïve mice received Blo t 5 specific-Th2 cells intravenously followed by intranasal challenge with Blo t 5. The responses of recipient mice were analysed by immunological and histochemical methods.

Results: A long term TCRVβ3$^+$ Blo t 5 specific Th2 cell line producing high levels of IL-4, IL-5, IL-13 and IL-10 but not IFN-γ was established. These CD44^{high}CD62L$^-$ Th2 cells showed up-regulation of CTLA-4, ICOS, OX40, 4-1BB, CD27 but not CD40L upon stimulating with Blo t 5. After intranasal challenge with Blo t 5, Th2 cells recipient mice developed Blo t 5-specific IgG1 and IgE, airways eosinophilia and mucus production of the Goblet cells. In addition to the donor Th2 cells, the cellular infiltrate consisted of CD4$^+$, CD8$^+$ T cells and NK cells of the recipient mice. Such cellular inflammation could be suppressed by dexamethasone intervention. The pathological results were not observed in the PBS challenged recipient mice.

Conclusion: Blo t 5-specific Th2 cells played a central pathological role in mediating allergic airway inflammatory responses resembling those seen in humans. This animal model is particularly useful for screening of novel therapeutics for asthma and allergy.

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Differences in Foxp3$^+$CD4$^+$ ratio between symptomatic atopic patients and asymptomatic atopic controls with similar levels of Th1/Th2 markers

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Rationale: Foxp3, a responsible gene for IPEX syndrome in which both Th1 and Th2 responses are hyperreactive, is known as a master gene of naturally occurring regulatory T cell and maybe of some other regulatory T cell subsets. Therefore, the regulatory roles of Foxp3$^+$ cells in allergic disorders are highly expected; however, the precise role of circulating Foxp3$^+$ cells in common allergic diseases remains unclear. Then in order to evaluate its diagnostic potential, we examined intracellular Foxp3 protein expression in circulating peripheral blood CD4$^+$ T cells from allergic patients and healthy controls at the single-cell level.

Methods: The ratio of Foxp3$^+$CD4$^+$ cells in the CD4$^+$ cell fraction as well as various laboratory data were measured for 64 donors, including 37 patients with atopic dermatitis and/or bronchial asthma. None of the subjects was treated with oral corticosteroids. All the patients were judged positive for specific IgE antibodies against at least one of *Dermatophagoides pternyssinus*, *Dermatophagoides farinae*, and Japanese cedar pollen using Pharmacia's CAP system fluorescence enzyme-linked immunoassay (CAP-FEIA). Twenty-three of the 27 age-matched healthy volunteers also showed at least one positive reaction to those allergens by CAP-FEIA, although they never had subjective symptoms related to allergic diseases.

Results: Foxp3$^+$CD4$^+$ ratio in the total population was correlated inversely with the levels of total serum IgE ($p = 0.021$), % eosinophil count ($p = 0.022$), and serum IFN-γ ($p = 0.048$). By performing a case-control study by matched-pair analysis to eliminate the influence of the Foxp3$^+$CD4$^+$ level correlations with the total IgE level and eosinophil ratios, Foxp3$^+$CD4$^+$ ratio was significantly lower in active atopic patients compared to asymptomatic donors having similar levels of IgE, eosinophils and IFN-γ.

Conclusion: Our findings imply that circulating Foxp3$^+$CD4$^+$ regulates both Th1 and Th2 responses *in vivo*. Moreover, symptomatic atopic patients had a lower Foxp3$^+$CD4$^+$ ratio than asymptomatic atopic controls having similar levels of Th1/Th2 markers. Measurement of Foxp3$^+$CD4$^+$ ratio has the potential to aid in evaluating the presence of active inflammation in patients with allergic diseases, which can not be evaluated by known Th1 and Th2-related markers in patients with allergic diseases.

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Molecular biomarkers of eosinophil-lineage commitment: Multiplex Q-PCR analysis of GATA-1, MBP and IL-5 receptor mRNA expression kinetics in both peripheral and umbilical cord blood

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Background: Using colony assays and flow cytometry, we have shown that eosinophil/basophil (Eo/B) progenitor phenotype and function are associated

with both atopic risk at birth and early childhood clinical outcomes. These assays are cumbersome, however, and we thus endeavored to develop surrogate molecular markers of eosinophil lineage commitment.

Objective: To utilize Q-PCR to determine the kinetic patterns of expression of Eo/B-lineage specific genes in cord blood (CB) and peripheral blood (PB) in response to IL-5 stimulation.

Methods: CB and PB non-adherent mononuclear cells (NAMNC) were isolated from random fresh (and frozen, for CB) samples, and incubated in the presence of IL-5 (1 ng/mL). At 24, 48, 72h, 96h (CB only) and 1 week post-stimulation (PB only), RNA was isolated, reverse transcribed, and expression of IL-5Rá, GATA-1, and MBP was determined utilizing multiplex Q-PCR. Relative expression ratios of stimulated to un-stimulated cells were calculated using the delta-delta Ct method.

Results: Stimulation of CB NANMC with IL-5 resulted in an up-regulation of GATA-1 expression, peaking at 48h, and decreasing expression by 72 hours. Preliminary evaluation of a 96h time-point suggests down-regulation. PB NAMNC's similarly showed up-regulation peaking at 48h but with a lower overall fold-increase and a slower return to baseline expression than that observed in CB. MBP expression in CB was slowly up-regulated in response to IL-5 stimulation, maximal at 96h; in PB, MBP expression was stable until after a full week of incubation when up-regulation could finally be detected. There was completely stable expression IL-5Rá, in both CB and PB.

Conclusion: Multiplex Q-PCR analysis of mRNA from CB and PB demonstrates expression of critical Eo/B lineage-specific events. Further investigation of the validity and utility of Q-PCR analyses of CB and PB for surrogate, molecular markers of Eo/B differentiation is underway.

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Double-stranded RNA synergistically enhances MUC5AC induction by TGF-α in human bronchial epithelial cells

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Background: Goblet-cell hyperplasia and upregulated epidermal growth factor receptor (EGFR) expression are important features in hypersecretory diseases such as asthma. Since viral infection also induces production of respiratory mucins, we investigated whether co-stimulation of double-stranded RNA (dsRNA) and TGF-α enhances the mucin hyperproduction in human bronchial epithelial cells. Furthermore, we investigated the signaling pathway regulating dsRNA and TGF-α-induced MUC5AC secretion.

Methods: A human pulmonary mucoepidermoid carcinoma cell line, NCI-H292, was incubated with polyI:C, which is an analogue of dsRNA, and TGF-α for an appropriate time. MUC5AC protein, mRNA, and promoter activity was measured by ELISA, RT-PCR, and luciferase assay. In inhibition studies, the cells were pretreated with selective MAPK inhibitors prior to stimulation with polyI:C and TGF-α. The activation of ERK was examined by using Western blot analysis. In order to examine MUC5AC mRNA transcript stability, NCI-H292 cells were cultured with actinomycin D after induction of MUC5AC mRNA by polyI:C and/or TGF-α for 18h. EGFR mRNA expression was measured by RT-PCR to examine whether EGFR mRNA is upregulated by polyI:C. NCI-H292 cells were incubated with anti-IL-8 antibody prior to stimulation with polyI:C and TGF-α to investigate the involvement of IL-8.

Results: PolyI:C and TGF-α synergistically increased gene expression and protein production of MUC5AC in NCI-H292 cells. This increase was dependent on MUC5AC gene transcription. MEK1/2 inhibitor (U0126), but not p38 MAPK inhibitor (SB20358) and JNK inhibitor (SP600125), significantly blocked synergistic induction of MUC5AC mucin, indicating that the ERK pathway is the main stream of polyI:C and TGF-α-induced MUC5AC production. Western blot analysis confirmed this result. Addition of polyI:C neither changed the rate of MUC5AC mRNA turnover nor upregulated EGFR expression. Anti-IL8 antibody did not inhibit MUC5AC mRNA expression indicating that IL-8 had no role.

Conclusion: These results suggest that double-stranded RNA has the potential to synergistically amplify induction of MUC5AC mucin by TGF-α stimuli. The mucus hypersecretion in a viral infection model is not regulated by p38 MAPK and JNK but is regulated by a pathway through ERK.

TREATMENT OF ASTHMA

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Monitoring and management of childhood asthma in southeast asian countries: a questionnaire study

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Background: We examined practices of physicians in India, China, Sri Lanka, Australia, Singapore, Indonesia, Philippines and Taiwan in monitoring and treating childhood asthma.

Methods: Our 6-page standardized questionnaire was sent to doctors via post. The questionnaire is made up by questions on: a) methods of monitoring of childhood asthma, b) practices in managing acute asthma exacerbations, and c) choice of therapy in maintenance treatment.

Results: Our study reflects mostly the practices of doctors who practice in urban regions. Of respondents, 41.4% were general pediatricians, while 26.3% were general practitioners. A small fraction of doctors used score cards or diaries to monitor asthma, ranging from 0% (Philippines and Australia) to 15.9% (India). Only 8.1% (Sri Lanka) to 51.7% (Australia) use either a peak flow meter and/or spirometry to monitor asthma. However, for frequency of usage, 35% (China) to 94% (Indonesia) never or seldom make use of a peak flow meter, and 33% (China) to 97.6% (Indonesia) never or seldom use spirometry for monitoring. Nebulised salbutamol given every 20 minutes was the treatment of choice in treating acute asthma attacks by the majority of doctors in each country, ranging from 29% (China) to 85.7% (Sri Lanka). A minority (4% (China) to 18.8% (Taiwan)) administer systemic corticosteroids in an outpatient setting (ER or polyclinic) in acute asthma. 53.6% (India) to 93.9% (Sri Lanka) of doctors also indicated that their duration for treatment with corticosteroids would be for 3–5 days, and at the same dosage throughout the duration. A good number (32.7% (Sri Lanka) to 80.2% (Taiwan)) use antibiotics in the treatment of acute asthma, but only when pneumonia, otitis media or sinusitis was likely. Few doctors favoured high-dose inhaled corticosteroids in acute management, ranging from 0% (Indonesia and Sri Lanka) to 8.4% (Philippines). For maintenance treatment, a significant fraction of doctors chose a long-acting beta agonist (LABA) monotherapy as a first-choice treatment for asthma maintenance. For infants the percentage ranged from 1.3% (Australia) to 76.3% (Indonesia),in preschoolers 0%(Australia) to 61% (China), and in older children, 0% (Philippines) to 61% (China).

Conclusion: There is much room for improvement in increasing doctors' awareness to guidelines for more effective management of paediatric asthma in Southeast Asia, especially regarding the use of LABA monotherapy.

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Efficacy of inhaled heparin is effective in the treatment of acute exacerbation of asthma

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Background: Inhaled Heparin was found to be highly protective against methacholine induced bronchospasm in bronchial asthma possibly via a direct effect on smooth muscle or may have potential as anti-inflammatory activity.

Objective: The aim of the study was to determine the additional therapeutic benefit of inhaled heparin in the treatment of hospitalized patients for acute asthmatic exacerbation and treated with inhaled bronchodilators and glucocorticoid therapy.

Methods: Thirty patients (20 male, 10 female), mean age(31 +/− 10 years), admitted for acute exacerbation of asthma, participated in a prospective, randomized, double-blind, placebo-controlled study. All the subjects received hydrocortisone, administered intravenousl, and nebulized salbutamol. The treatment group received inhaled heparin therapy (20,000 U in 4 mL) every 4 hr. and the placebo group received 0.9% saline solution for 24 hours. Baseline respiratory parameters such as oxygen saturations, respiratory rates, and peak

flow rates were measured and repeated after 24 hours treatment. The number of salbutamol nebulizations needed were recorded at the end of 24 hours.
Results: Most of the patients were in moderate severity of asthma exacerbation. All patients in both groups showed improvement in oxygen saturations, respiratory rates, and peak flow rates. Statistically significant difference was observed between the 2 groups regarding both the respiratory parameters and the mean number of salbutamol nebulizations needed ($P > 0.05$, $P > 0.01$).
Conclusion: Inhaled Heparin demonstrated additional beneficial effect to the combination of beta adrenergic agonists and glucocorticoid treatment in acute asthma attack.

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A descriptive non inferiority comparative analysis of single dose nebulized formoterol powder (Foradil 24 mcg) in isotonic saline vs three doses of albuterol (2.5 mgs ampules, GSK) every 20 min for childrens acute asthma: a cost effective approach for developing countries

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Asthma greater cost expenditures comes from acute care and hospitalizations. In Venezuela's Ministry of Health ambulatory facilities (primary care for majority of 26 millions inhabitants), acute asthma represents a rising and significant burden of care close to a million asthma crisis / year; for the last quarter of a century standard emergency acute asthma care has been delivered in nebulized form with a fixed fenoterol (0.5 mg/ml) - ipatropium bromide (0.25 mg/ml) combination in repeated dosing (Berodual, Boehringer-Ingelheim). We and others (1, 2) demonstrated formoterol powder to be highly effective in relieving bronchospasm when used in nebulized form. This study blindly compares at random a nebulized *single 24 mcg dose (Foradil ® 2 capsules in 2.5 cc / saline) VS three nebulized * doses every 20 minutes of albuterol (GSK) 0.15 mg/kg/dose (maximum 2.5 mgs/dose) in fifty acute asthmatic children (6–12 years, mean 9 y/o) attending an emergency care setting serving an impoverished area of metropolitan Caracas. Clinical scoring, oxygen saturation and pulmonary function, were measured before and one hour after treatment.

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Aleuria aurantia lectin coated microspheres for oral immunotherapy of grass pollen allergy

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Background and Aims: Recently, we could demonstrate that mucosal targeting with Aleuria aurantia lectin (AAL) coated Poly(D,L-lactide-co-glycolic acid, PLGA) microspheres modulated a specific immune response in birch pollen allergy. Using grass pollen as a novel allergen encapsulated in AAL-coated particles we aimed to further evaluate the immune modulating functionality of microspheres in a murine asthma model.
Methods: BALB/c mice were sensitized intraperitoneally with grass pollen extract followed by aerosol challenges inducing acute allergic asthma. Thereafter, animals were treated repeatedly with grass pollen loaded PLGA microspheres coated with AAL for M-cell targeting. Mice immunized with microspheres coated with glycine, with further intraperitoneal allergen injections and naïve animals served as control. The outcome of the treatment was evaluated by measurements of grass pollen specific antibodies (IgG1, IgG2a, IgE and IgA) in sera and bronchio-alveolar lavage (BAL) and by determination of cytokine profiles (IL-5, IL-10, TGF-β) in spleen supernatants.
Results: In the animals treated with AAL-coated mircrospheres allergen-specific IgG1, IgG2a and IgE levels were unaffected by treatment. However, allergen-specific IgA levels increased during therapy. Interestingly, in cytokine evaluations IL-10 levels did not change, whereas reduced amounts of IL-5 and high levels of TGF-β were measured in the mice treated with AAL-functionalized microspheres compared to the other groups.
Conclusion: These data indicated that in the murine model grass pollen allergy was beneficially influenced by targeting M-cells via AAL-coated, allergen-loaded microspheres. Thus, the potential of functionalized microparticles for specific immunotherapy for acute respiratory allergy was confirmed.

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Oral sulforaphane safely and effectively induces antioxidant phase II enzymes in the human airway

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Substantial evidence implicates particulate air pollution exposure as an important factor for asthma exacerbations and the increasing prevalence of allergic respiratory disease. As the inflammatory effects of particulate air pollution are mediated by the induction of cellular oxidative stress, strategies to reduce oxidative stress may potentially reduce the harmful effects of particulate air pollution. Endogenous Phase II enzymes abrogate oxidative stress through metabolism of particulate-associated reactive chemicals and the scavenging of reactive oxygen species. We conducted a placebo-controlled dose escalation trial to investigate the in vivo effects of sulforaphane, a naturally occuring potent inducer of Phase II enzymes, on the expression of GSTM1, GSTP1, NQO1, and HO-1 in the upper airway of human subjects. Sixty human study subjects consumed oral standardized broccoli sprout homogenate (BSH) doses containing sulforaphane once daily for 3 consecutive days. An escalating block design was used with 25, 50, 75, 100, 125, 150, 175, and 200 grams (g) of BSH to ensure safety and tolerability. Additional subjects were subsequently enrolled at doses of 125, 150, 175, and 200 g to examine dose-response effect. Five subjects completed the protocol with non-sulforaphane containing alfalfa sprout homogenate dosing at 200 g as a control group. RNA expression for selected Phase II enzymes was measured in nasal lavage cells by RT-PCR

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	n	Clinical Score pre	Clinical Score post	FEV1 pre (%)	FEV1 post (%)	Sat O2 pre	Sat O2 post
ALBUTEROL	25	10.12	8.20 (p 0,001) †	55.80 %	56.09 % (p 0,749 †	95.22 %	96.86 % (p < 0,001)†
FORMOTEROL	25	10.56	7.04 (p <0,001) †	58.08 %	67.84 % (p 0,001) †	94.56 %	96.92 % (p < 0,001) †

(† Wilcoxon) Nebulized single dose formoterol powder (24 mcg) is as effective as three doses of albuterol and at one fifth the cost in children's acute asthma; is well tolerated and patients of all ages can benefit. Nebulized Formoterol single dose is non - inferior to three doses of albuterol and underscores many possibly substantial care savings, given the heavy load that acute asthma care represents in our country and many other developing nations (3). * PARI ® nebulizer (Pronet Turbo) 1. Perez Puigbo A et al. Nebulized saline solution of dry powder formoterol is useful for acute bronchospasm. Arch Venez Pharmacol Therapeutics 2001; 2:128 2. Ting S et al. Nebulized combined formoterol and budesonide (NCFB) for children younger than 5 years of age with persistent asthma. JACI 2003; vol11,pS148. 3. Watson JP, Lewis RA. Is asthma treatment affordable in developing countries? Thorax 1997;52:605–7.

before and after BSH dosing. All subjects tolerated oral BSH dosing without significant adverse events. Increased Phase II enzyme expression in nasal lavage cells occurred in a dose dependent manner with maximal enzyme induction observed at the highest dose of 200 g BSH. At 200 g BSH daily GSTM1, GSTP1, NQO1, and HO-1 expression was 219, 201, 299, and 221% of baseline expression respectively ($p \leq 0.001$ for all enzymes). Phase II enzyme induction was not seen with ingestion of non-sulforaphane containing alfalfa sprouts. Thus, oral sulforaphane safely and effectively induces mucosal Phase II enzyme expression in the upper airway of human subjects. Based on this work and previous in vitro/animal studies, sulforaphane may represent a novel therapeutic strategy for the treatment of allergic respiratory conditions. Additional studies are underway to determine whether Phase II enzyme induction is effective in reducing the inflammatory effects of particulate-induced oxidative stress in the human airway.

IgE SPECIFICITY AND DIAGNOSIS

133 Abstract withdrawn

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Immunoproteomics approach for identification of novel allergens of aspergillus fumigatus

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Background: Increasing incidences of *Aspergillus fumigaus*-induced allergic asthmatics all over the world necessitate identification of a catalogue of allergens of clinical importance. In the present study, immunoproteomic approach was used to establish a set of candidate allergens, which could be explored further for diagnostic application in allergic aspergillosis and asthmatics including ABPA.

Methods: Immunoproteomics combined with mass spectrometric analysis was used to identify proteins of three-week culture filtrate (3wcf) responsible for inducing *A. fumigatus*-specific IgE immunoreactivity, using pooled sera from *A. fumigatus*-sensitised asthmatics. Their diagnostic potential was also examined against patients with allergic bronchopulmonary aspergillosis (ABPA), by 2-DE immunoblotting with individual sera from such patients.

Results: Peptide mass fingerprint (PMF) using matrix-assisted laser desorption/ionization-time of flight-mass spectrometry (MALDI-TOF-MS) and/or *de novo* sequencing by MS/MS analysis of the protein spots from 2-D gels led to identification of a total of sixteen allergens of *A. fumigatus*. Eleven of them are being reported as allergens for the first time and five were reported earlier. Putative isoforms of the proteins Asp f 13 and chitosanase have been observed for the first time. When studied for immunoreactivity of these proteins among patients with ABPA using their individual sera, these patients exhibited sensitisation although the pattern was varying. Taken together, these proteins could thus be considered as potential allergens even among patients with ABPA. Three of these proteins viz. the hypothetical protein, extracellular arabinase and chitosanase could be major allergens.

Conclusion: The immunoproteomic approach applied to the analysis of culture filtrate proteins resulted in identification of several candidate allergens, many of them novel, contributing to the catalogue of *A. fumigatus* allergenic proteins. These allergens may facilitate improved serodiagnosis for allergic aspergillosis. In addition, the immunoreactivity of these proteins observed among the patients with ABPA may have potential for serodiagnosis and opens up scope for evaluation and development of personalized immunotherapeutics.

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Laboratory testing and evaluation of olive pollen allergy and cross reactivity in Jordan

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Background: The extensive and unorganized planting and cultivation of the holly olive trees in our country is the first cause of allergy. Olive tree (olea europaea) pollen is a major cause of seasonal allergy-making it the first outdoor pollen calendar in Jordan. To date, ten allergens, named Ole e 1 to Ole e 10, have been isolated and characterized from this pollen Ole e 10, a small protein, has been recently described as a major allergen from this pollen, since it affects more than 55% of allergic patients. Few studies regarding olive allergy have been reported in our area. The olive pollination season lasts two months approximately from the middle of May to the middle of July.

Objective: The main aim of this study was to address the performance of olive screen testing by total and specific IgE and the cross reaction with other pollens.

Methods: Total and specific serum IgE to olive pollen was tested and evaluated in 400 allergic patients before, during and after the peak pollen season using immunoblot Allergy Screen-Respiratory pannel.

Results: Out of the 400 patients serum specimens 132 (30%) had allergy to IgE olive pollens. 125(94%) of the allergic patients had elevated total serum IgE >100 IU/ml). 10 samples from healthy subjects were used as negative controls. Cross-reactivity with orange, Banana, Grass pollens and Ambrosia has been noticed in 10 cases (15%).

Conclusion: Olive tree pollen is one of the main causes of allergy in Mediterranean region included Jordan.

It is worth to mention the importance of the total and specific IgE testing for olive pollen allergy detection and cross-reaction with other pollens and fruits Suggesting the presence of Ole e 10-like proteins in many sources which may be related to the development and exacerbation of allergic and asthmatic process, but these results could be interpreted with caution regarding clinical symptoms, signs, and skin prick testing.

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In vivo reactivity to grass pollen in correlation to specific IgE antibody levels in patients with allergic rhinoconjunctivitis

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Background: Previous investigations established dose-response curves between the severity of clinical symptoms and specific IgE antibody levels to both indoor and food allergens in allergic individuals. We questioned whether this also holds true for outdoor allergens e.g. grass pollen allergens.

Methods: 100 patients with allergic rhinoconjunctivitis (ARC) and allergy to timothy grass pollen as well as 40 healthy controls were recruited during the grass pollen season in 2006 and 2007. To investigate the correlation between IgE antibody levels and threshold in vivo reactivity we determined specific serum IgE antibodies to timothy grass pollen (ImmunoCAP™, Phadia AB,

Uppsala) at three time points during one year (June/July; October; December) and performed conjunctival and nasal provocation tests with grass pollen extracts at different concentrations (Phleum pratense, HAL Allergy, Duesseldorf, Germany). In addition, skin prick tests were performed. The investigations were paralleled by grass pollen counts using a Burkard trap performed at the University Campus Biederstein, Munich (<50 m distant from the laboratory, 1.80 m above the ground). Additionally, the patients scored their symptoms with visual analog scales and exhaled nitric oxide was analysed as measure for inflammatory lung reaction.

Results: Preliminary results from 41 patients with ARC revealed a significant relationship between specific IgE concentration in serum and the titrated level of the skin prick test ($p < 0.0096$, Jonckheere-Terpstas trend test). However, no correlation between specific serum IgE to timothy grass and thresholds of nasal and conjunctival provocation tests were found.

Conclusion: Measurement of specific serum IgE levels seems to accurately predict the in vivo reactivity with regard to skin prick test in patients with ARC to grass pollen allergen. The preliminary results indicate a good correlation between indicators of sensitization, but not between serum IgE levels and threshold allergen concentrations that induce symptoms Further data analysis concerning conjunctival and nasal provocation tests and symptom scores in a greater number of patients is in progress.

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The diagnosis of Pollen-Food Syndrome (PFS) through the use of a structured questionnaire

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PFS, a manifestation of Oral Allergy Syndrome (OAS), is a common food allergy. The characteristic symptoms, speed of onset and the typical foods involved suggest PFS could be diagnosed though clinical history alone.

Subjects with reported springtime hay fever completed a PFS diagnostic questionnaire (PFSDQ) and were allocated to either Group 1 (has PFS), Group 2 (No PFS, ? food allergy) or Group 3 (no PFS or food allergy). All subjects then had a consultation with an allergy specialist who made a provisional PFS diagnosis. This was followed by skin prick testing to fresh foods using the prick by prick test method (PPT) and oral food challenge (OFC). The PFSDQ was measured against the gold standard of positive OFC, and a second standard, the final diagnosis accorded to each subject by the medical allergy specialist, using PPT and OFC results to modify his original diagnosis.

119 subjects completed the study; 58 (49%) were allocated to Group 1 (PFS+ve), and 61 (51%) to Groups 2 (30) and 3 (31) (PFS-ve). 47 subjects had a positive OFC, 41 of whom were from Group 1. Although there was a significant difference between the PFSDQ and OFC results ($p < 0.05$), this was not the case when the PFSDQ was compared to the final diagnosis standard ($p = 0.5$). The sensitivity, specificity, positive and negative predictive values of the PFSDQ were all $\geq 90\%$ when tested against the final diagnosis standard. The reliability of the PFS-DQ was high with a standardised alpha score of 0.819 (Chronbachs Alpha Test). Factor analysis showed the key components of the PFSDQ to be symptom type, raw or cooked foods, speed of onset of symptoms and number of reported symptoms (Varimix rotation). Logistic regression analysis showed the only predictive factors of PFS diagnosis were reactions to raw plant foods ($p < 0.01$), onset of symptoms within 5 minutes of eating ($p < 0.01$) and number of reported symptoms to foods ($p < 0.05$).

Diagnosis of PFS using the PFSDQ was not significantly different to that made by a specialist allergist using clinical history, PPT and OFC. The most important predictive variables for PFS were reported reactions to raw plant foods, rapid onset of symptoms and multiple symptoms. The PFSDQ could be a useful diagnostic aid for those working in primary care to screen adults with reported food symptoms for the presence or absence of PFS.

DRUG ALLERGY

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Importance of benzylpenicillin in beta-lactam hypersensitivity diagnosis

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Introduction: Drug hypersensitivity to beta-lactam was often skin tested using benzyl-penicillin, major (PPL) and minor (MDM) penicillin determinants, amoxicillin, ampicillin and any other culprit beta-lactam. However, PPL and MDM were removed from the marked. The aim of the study was to assess the importance of benzylpenicillin before and after the withdrawal.

Methods: Using our Drug Allergy and Hypersensitivity Database (DAHD) and data coming from an Italian centre, we conducted an historico-prospective cohort study. All patients who consulted between 1996 and 2007 for a suspected beta-lactam hypersensitivity reaction and who had at least positive skin test to benzylpenicillin were included. Diagnosis and skin tests followed the European Network on Drug allergy (ENDA) recommendations. Benzylpenicillin, PPL, MDM, Ampicillin, Amoxicillin were always skin tested.

Results: 133 patients (48–36.1% men), 15 (11.3%) asthmatics and 41 (30.8%) atopics were included. 13 (9.8%) were only positive to benzylpenicillin, 32 (26.5%) were also positive for PPL and 66 (55.5%) for MDM. Without skin testing for PPL and MDM, the number of positive to benzylpenicillin increased to 20 (15.0%). No difference was observed for asthma and atopic between subjects only positive to benzylpencillin and those also positive for another penicillin ($p = 0.84$ and $p = 0.34$ respectively). Anaphylaxis and anaphylactic shock were more common in subjects positive to several penicillins (76–67.3% vs 7–35%, $p = 0.001$). Conversely, only 26 (23.0%) vs 8 (40.0%) presented an urticaria / angiodema.

Conclusion: Since the withdrawal of PPL and MDM, benzylpenicillin skin test appeared to be mandatory, 15% of the patients being only positive to this drug. This should significantly reduce oral challenges.

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Drug-induced hypersensitivity syndrome and herpes virus infection

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Background: (BDrug-induced hypersensitivity syndrome (DIHS) is a unique severe adverse drug reaction, which is well known to be accompanied with reactivation of human herpesvirus 6 (HHV-6) in many cases 2–3 weeks after development of drug reactions. The clinical features of this syndrome are acute widespread maculopapular, polymorphous, eczematous and/or erythrodermic erythema, fever, lymph node swelling, liver dysfunction, eosinophilia and leukocytosis with atypical lymphocytes. After first reports of DIHS with HHV-6 reactivation in Japan in 1998, 118 cases have been reported with data of HHV-6 reactivation up to now.

Methods: We retrospectively analyzed recent reports of suspected adverse drug reactions submitted to medical journals from Japan, in order to define the presenting characteristics of these diseases in Japan.

Results: Ages ranged from 0 to 89 years old with a mean of 48.6 years old and the ratio of M/F was 64/54. 63.7 % of causative drugs were anticonvulsants (carbamazepine, 63.3% of the anticonvulsant drugs). The other common causative drugs were sulfonamides, mexilletine (an antiarrhythmic drug) and allopurinol. The symptoms developed 3 weeks or more after the beginning of

causative drug administration in 80.5% of the patients. Recurrence of the symptoms was observed in 40% of the patients. Four of them died and the mortality rate was 3.6%. Reactivation of HHV-6 was detected in 83.9% of the patients by the increase of IgG against HHV-6 and/or increase of HHV-6 DNA in the peripheral blood and sera. On the other hand, reactivation of cytomegalovirus was observed in 4 patients without HHV-6 reactivation. In treatment of 118 patients with DIHS, steroid without puls was in 71 cases (60.2%), steroid puls with mPSL in 15 cases (12.7%; 1g/day in 40.0% of 6/15, under 1g/day in 33.3% of 5/15, unclear in 26.7% of 4/15), intravenous injection of high dose immunoglobulin (IVIG) with steroid in 5 cases (4.2%), and plasmaapheresis (PA) with steroid in 3 cases (2.5%). The mortality in DIHS was unexpectedly high as 3.4% (4/118), as compared to that in SJS.
Discussion: A probable role of HHV-6 in DIHS was discussed in comparison of that of Epstein-Barr (EB) virus in infectious mononucleosis and Mosquito bite hypersensitivity.

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Drug provocation test in patients' \drug hypersensitivity reaction

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Background: It has bndeen confirmed that, drug hypersensitivity are common and life threatening. This diagnosis can be vigorous and based on clinical history and a physical examination, possibly under skin tests and drug provocation test. We intend to describe the out come of drug provocation test in analyzing patients with histories suggesting drug allergy, using retrospective analysis of clinic case series.
Methods: Some 416 consecutive patients with suspected immediate drug allergy referred to the hospital between, January 1999 to February 2005. Patients with served skin reaction and those with positive result on skin test for B-lactams were excluded. it should be noted that single- blinded administration of increasing doses of the suspected drug up to the usual daily dose, under struck hospital surveillance.
Results: 920 drug provocation tests were performing using various drugs, including aspirin (11.4%) nonsteroidel anti-inflammatory drugs (7.5%) macrolides (8.5%) paracetamole (10.1%) and other non-steroidal anti-inflammatory drugs (12.2%). We had 192 (15.6%) positive provocation test results which also reproduce the same symptoms, albeit milder and of a shorter duration - in the following patients.10 (4.2%) with history of anaphylactic shock, 15(5.5%) the history of anaphylaxis without shock. 70 (46.4%) with a history of urticaria and 5(2.4%) with a history maculopapular eruption. All reactions were completely refers by prednisone and epinephrine. However, we assume or accept that false negative results on drug provocation test may have occurred because of sanitation, rare cofactors not included in the diagnostic procedure and tolerance introduction during provocation.
Conclusion: It can be certain that drug provocation tests in individuals with suspected drug allergy performed in carefully control settings can confirm drug hypersensitivity.

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Antituberculous drug rechallenge: success rate among patients with cutaneous hypersensitivity reactions

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Background: This is the first study to systematically document the proportion of cutaneous hypersensitivity reaction (CHR) in patients with adverse drug reactions (ADR) to antituberculous medications and their excipients.
Objective: 1.To determine the proportion of CHR due to specific antituberculous medications; 2. To investigate the success rate of rechallenging by shifting to a different brand of antituberculous medication.
Methodology: All patients with CHR to antituberculous medications from January 1 to August 31, 2001 were included. After at least 48 hours off antihistamines, the patient was rechalleged with white or light yellow colored drugs to minimize excipients the patient may be sensitized to. Rechallenge was considered successful if there was no recurrence of symptoms while on the multiple antituberculous drug regimen.
Results: Sixty-six patients were referred for ADR to antituberculous medications. Forty-five had CHR. Forty-two patients were rechallenged. Fourteen were successful. Sixteen had documented cutaneous adverse reactions to one or more drugs with the highest incidence occurring with ethambutol and pyrazinamide. The success rate of rechallenge was 14/42.
Conclusion: 1. Skin allergy is common (68%) with first line anti-TB drugs. The most common drugs associated with CHR were ethambutol and pyrazinamide; 2. The success rate of drug rechallenge when shifting to a different brand of anti-TB medication is 14/42 (33%)

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Meta-analysis: Risk of angioedema with angiotensin receptor blockers (ARBs) in patients with prior angioedema associated with angiotensin converting enzyme inhibitor (ACE-I)

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Background: Patients who experience angioedema (AE) after taking ACE-I have been reported to develop AE when taking an ARB, but few studies describe the risk. We sought to answer this question by performing a systematic review of the medical literature.
Methods: A literature search was performed in MEDLINE, EMBASE, BIOSIS, and Current Contents with no limitations from Jan 1990-May 2007. SCISEARCH was also used to identify additional citations of key articles. Reference lists of retrieved studies and review articles were evaluated for additional citations. Two authors independently evaluated studies for inclusion and abstracted relevant data according to pre-defined parameters. Any article that described a cohort of patients who had experienced AE after taking an ACE-I, were subsequently exposed to an ARB and followed for at least one month were included. The percentage of patients experiencing AE was abstracted from each article and confidence intervals were calculated using the exact binomial method. The pooled percentage was calculated using the inverse variance method.
Results: From the 238 unique articles initially identified, three articles describing 71 patients met inclusion criteria. One was a randomized controlled trial and two were retrospective cohorts. The mean age of patients was 63 years with 56% male and 44% female. Two of the studies were predominantly Caucasian subjects with the third study being 69% African-American. The mean time of follow-up was 20.3 months. These articles described both confirmed and possible cases of AE secondary to ARB. For possible cases the risk of AE was 9.4% (95%CI 1.6–17); for confirmed cases it was 3.5% (95% CI 0–9.2). There were no fatal events. There was no statistical heterogeneity between trials ($p > 0.3$).
Conclusion: Limited evidence suggests that for patients who developed AE when taking an ACE-I, the risk of developing any AE when taking an ARB is between 2–17% and for confirmed AE the risk is 0–9.2%. This information will aid clinicians in counseling patients regarding therapy options after developing AE due to ACEI.

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Drug allergy and quality of life

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Background: Despite clinical experience suggests that drug hypersensitivity may have a detrimental impact on patients' life, no data are now available about this allergic reaction from a subjective viewpoint.

Aim: The aim of our study was to develop and evaluate the psychometric properties of a questionnaire that assesses the drug hypersensitivity burden from the patient's perspective.

Methods: 172 consecutive patients (69.2% female, 30.8% male, mean age: 41.16 years +14.69) were asked to fill in a 20-item questionnaire to evaluate the impact of drug allergy on QoL.

Results: The different reaction patterns were urticaria (77 patients), anaphylactic shock (24), angio-oedema (20), exanthema (16), others cutaneous eruptions (11), bronchospasm (11), other manifestations (7) The preliminary analysis of patients' answers shows a worse QoL in the elderly ($p < 05$), in women ($p < 01$) and in patients with anaphylaxis ($p < 005$).

Discussion: The use a of a simple tool permits to capture the burden of drug hypersensitivity on patient's life, providing data for a better management of patients

DERMATOLOGY

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Expression of IL-31-producing cells in children with atopic dermatitis complicated by hen-egg allergy

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Background: Cytokines are essential for the development and function of the nervous systems as well as the immune and hematopoietic systems. IL-31 was secreted in larger amounts preferentially in Th2 than in Th1 cells. IL-31 functiones by the heterodimeric receptor composed of IL-31RA and OSMR that is expressed constitutively on epithelial cells and keratinocytes. These cells are likely to be involved in the dermatitis and pruritis of patients upregulating IL-31.

Objective and Methods: Peripheral blood mononuclear cells were obtained from 22 patients with atopic dermatitis allergic to hen-egg ranging from 1 to 13 years in age, as well as from 9 healthy individuals who had no allergic symptoms. The patients had recurrent eczema, pruritis and positive skin reactions to egg white. Patients also had positive responses to the oral provocation test to raw hen egg. Diagnostic criteria for atopic dermatitis was based on the criteria of Hanifin and Rajk. We evaluated expression of IL-31-pruducing cells by the methods of newly developed FCAS analysis.

Results and Discussion: IL-31 was preferentially expressed in CLA (skin-horming cutaneous lymphocyte antigen) as well as CD45RO-positive cells in atopic children complaining of skin lesions and pruritis compared to normal subjects. Severity of these symptoms were correlated with amount of IL-31 in the cells.

Conclusion: These result suggests larger amounts of expression of IL-31 is likely to be involved in the pruritis as well as skin lesions in atopic children.

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Dysregulation of TLR-2 induced effects in monocytes from patients with atopic dermatitis: Impact of the TLR-2 R753Q polymorphism

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Background: Atopic dermatitis (AD) is often complicated by an enhanced susceptibility to bacterial skin infections, especially with Staphylococcus aureus (S. aureus). Toll-like receptors, especially (TLR)-2 recognizes cell wall components of S. aureus, e.g. lipoteichoic acid (LTA) and peptidoglycan (PGN). A heterozygous TLR-2 R753Q polymorphism occurs in a frequency of 11.5% in adult AD patients and has been shown to be associated with a severe phenotype.

Objective and Methods: The aim of this study was to investigate the impact of TLR-2 agonists (LTA, PGN and Pam3Cys) on cytokine production and cell surface marker expression in human monocytes from AD patients with the TLR-2 R753Q polymorphism compared to AD patients with "wild type" TLR-2 and control individuals to elucidate the functional role of the TLR-2 R753Q polymorphism.

Results: We could show that AD patients with the TLR-2 R753Q mutation produced significantly more IL-6 and IL-12 and significantly less IL-8 compared to AD patients with non-mutated TLR-2 upon stimulation with TLR-2 agonists. Expression of CD86 was significantly higher upon Pam3Cys stimulation in TLR-2 R753Q polymorph patients. Expression of CD80 was unaffected after stimulation with TLR2 agonists.

Conclusion: We show for the first time functional differences in TLR-2 responsiveness of monocytes from AD patients with the TLR-2 R753Q mutation compared to wild type AD patients in a ligand dependant manner. Clinical implication: Our data stratify the emerging concept that AD patients have a dysbalance in innate as well as aquired immunity. TLR-2 might be essential in the pathogenesis of AD and involved both in the enhanced susceptibility to skin infections with S. aureus and in a higher inflammatory response in patients with the TLR-2 polymorphism.

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Impact of atopic dermatitis on the quality of life in Japanese patients

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Background: Prevalence of atopic dermatitis (AD) in Japanese population has been rapidly increased for these decades. Impact of AD on their quality of life (QOL) is difficult to be assessed by using a translated QOL scale developed in western nations because of the influence of different culture. The aim of this study is to develop an original QOL assessment scale for Japanese patient with atopic dermatitis.

Methods: We have collected our own data from patients who visited Japanese hospitals and clinics by using an anonymous open questionnaire and some by interviews. One hundred and eighty items were extracted from whole data and based on them, secondary questionnaire comprising 68 items was made by expert's discussion. It was applied to 200 patients with atopic dermatitis for psychometric item reduction.

Results: Through the process of factor analysis and item analysis, sixty eight items were converged to 9 items comprising of four factors such as influence to daily activities, burden of medication, itchiness generated annoyance, anxiety about future. Internal consistency of each factor was excellent such that Cronbachα of each factor was 0.0772, 0.843, 0.865 and 0.789. Item content and factor loading score of each item is weak enthusiasm o.818, influence of sleep disturbance 0.617, burden of house keeping 0.589, burden of topical therapy 0.824, unwillingness of topical application 0.817, annoyance to itchiness 0.821, annoyance to scratching 0.756, anxiety about remission 0.824, and anxiety of fluctuating symptoms 0.684.

Conclusion: This study has revealed impact of atopic dermatitis on the QOL of patient is not only associated with dermatological symptoms but also adherence burden and anxiety about prognosis. The compact 9 time questionnaire developed in Japan this time would be useful to detect compre-

hensive quality of life of patients with atopic dermatitis even in a busy situation of our outpatient clinics.

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Eosinophil proteins in serum and urine of children with atopic dermatitis

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Atopic dermatitis (AD) is an inflammatory skin disease characterized by chronic and recurrent course, beginning primarily in an early childhood. The aim of the project was an evaluation of the usefulness of selected eosinophil proteins in serum (ECP, EPX) and urine (EPX) of children suffering from AD, as markers of disease severity. The study also aimed to analyze correlations between the level of examined proteins and such parameters as: skin prick tests (SPT) results, serum concentration of total IgE and coexistance of symptoms of other atopic diseases. We examined 40 AD children attending Allergic Diseases Diagnostic Center and hospitalized in the Department of Dermatology. As control group we selected 23 healthy children without any symptoms of allergic diseases. Mean level of eosinophil proteins measured in serum (ECP, EPX) and urine (EPX) of children suffering from AD was higher than in controls and statistically significant difference was detected for serum level of EPX. Patients presenting with very severe/severe AD had higher level of eosinophil proteins than patients presenting with mild/moderate AD, although no significant difference was found. AD children with positive SPT results and detectable serum specific IgE presented with higher mean level of serum and urine eosinophil proteins than in compared groups of patients with negative SPT results and undetectable serum specific IgE, although with no statistically significant difference. Total IgE level in AD children representing allergic form of the disease was statistically significantly higher than in children with non-allergic AD. In children with very severe/severe AD, total IgE level was significantly higher than in children with moderate/mild disease. Results presented above indicate singnificant role of eosinophils in etiopathogenesis of AD. Measurements of serum and urine level of selected eosinophil proteins may be considered as an important part of diagnostic approach in children suffering from AD, especially in differentiating of allergic and non-allergic form of disease. Among eosinophil proteins, EPX has always been the subject of particularly thorough investigations, because of easy and non-invasive way of measurement in urine, which is an important feature in diagnostics of children. EPX is a marker significantly better differentiating AD patients from healthy individuals in comparison to other factors, such as ECP and peripheral blood eosinophilia.

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Quantitative analysis of nerve growth factor (NGF) in the horny layer of atopic dermatitis and effect of treatment on NGF

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The expression of nerve growth factor (NGF) is known to increase in the skin of patients with atopic dermatitis (AD) and to be related to disease aggravation. In the present study, we measured skin NGF levels in AD patients and assessed the possibility of a relationship to AD severity, as well as effects of treatments.

Methods: NGF in the horny layer (horn NGF) of skin lesions on the cubital fossa was collected via tape stripping and measured using ELISA in AD patients before and after 2 and 4 week treatments. Itching and eruptions on the whole body and on lesions measured for NGF were evaluated. Peripheral blood eosinophil count, serum LDH level and serum total IgE level were also examined.

Results: The level of NGF was significantly higher in AD patients than in healthy controls, and correlated with the severity of itch, erythema, scale/xerosis, eosinophil count, and LDH level. The NGF level decreased significantly after treatments with olopatadine and/or topical steroid for 2 and 4 weeks. The decrease of NGF correlated with the decrease in the severity of itching, severity of AD on the whole body, erythema, papule, scale/xerosis and lichenification of the lesion, eosinophil count, and LDH level.

Conclusion: The level of the horn NGF was found to reflect the severity of itching and eruptions in AD. Therefore, measurement of the NGF via this harmless method appears to be useful in assessing severity and therapeutic effects in AD.

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Association between the allelic variant delta 32 of CCR5 receptor and the prevalence of urticaria, angioedema and atopic dermatitis symptoms

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Introduction: C-C chemokines, including RANTES, MIP-1, MCP-2, and the CCR5 specific receptor play an important role in the genesis of allergic inflammation. The presence of homozygosis for the delta32 (d32) polymorphism of CCR5 produces the absence of this receptor from the cell surface and heterozygosis promotes a very low expression of this receptor, hence reducing the availability of receptors capable of interacting with chemokines to induce the inflammatory response. We have previously informed a reduced risk of developing asthma and rhinitis in adults that present the d32 allelic variant. We could not find any report about the relationship between d32 and atopic dermatitis (AD), urticaria (URT) or angioedema (ANG).

Objective: To evaluate association between d32 polymorphism and AD, URT and ANG in adults from Rosario city.

Materials and Methods: 111 caucasian subjects, 36 males (32.7%) and 74 females (67.3%), aged 19 to 46 years old ($x = 23.88 \pm 4.45$), answered questionnaires for AD (ISAAC), URT and ANG. DNA was extracted from blood samples and d32 polymorphism was determined by PCR. Oligonucleotide sequences were: sense: 5′-GTCTTCCATTACACCTGCAGCTCT-3′, antisense: 5′-CACAGCCCTGTGCCTCTT-3′.

Results: 6.3% of the subjects presented the heterozygotic polymorphism and 0.9% the homozygotic polymorphism. The risk of suffering from AE, URT or ANG was significantly lower in the subjects that presented the d32 polymorphism. Among the subjects that referred for AD symptoms at least once in their life (15.3%), no one presented heterozygosis or homozygosis for d32, while in the subjects that did not refer these symptoms the frequency of the same genotype was 8.5% (RR = 0.83 IC:0.77–0.91; $p < 0.05$). Among the subjects that referred for URT symptoms at least once in their life (36.9%), 2.4% presented heterozygosis or homozygosis for d32, while in the subjects that did not refer these symptoms the frequency of the same genotype was 11.1% (RR = 0.70 IC:0.52–0.95; $p < 0.05$). Similar results were obtained in subjects that referred for ANG symptoms at least once in their life (13.6%), since no one presented heterozygosis or homozygosis for this polymorphism, while in the individuals that did not refer these symptoms the frequency of the mentioned allele was 8.4% (RR= 0.85 IC:0.79–0.92; $p < 0.05$).

Conclusion: The results obtained in this study allow us to suggest that the reduced risk to develop AD, URT and ANG could be influenced by the presence of the CCR5-d32 allelic variant.

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Prevalence of atopic dermatitis in infants

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Background: It is generally believed that atopic dermatitis (AD) patients have been gradually increased compared with the past. But, the prevalence of AD in infants has not been precisely investigated anywhere. The object of this work was to evaluate the exact prevalence of AD in infants and the relationship between induction/exacerbation of AD and skin barrier dysfunction.

Methods: Four-month-old and eighteen-month-old infants were examined by dermatologists at regular health check-ups in the public health center in Yokohama city throughout the year. Diagnosis of AD was based on the Japanese Dermatological Association criteria for the disease by the dermatologists. Skin barrier dysfunction was evaluated by measuring transepidermal water loss (TEWL) on face, abdomen and lower leg of infants without skin lesions.

Results: The prevalence of AD was 15.9% and 12.7% at four-month-old and eighteen-month-old infants respectively. The prevalence of eighteen-month-old infants was significantly decreased compared with that of four-month-old infants. The infants diagnosed as AD showed significantly higher TEWL values compared with healthy infants both four-month and eighteen-month-old infants at all skin portions.

Conclusion: The prevalence of AD in infants was tend to be decreased from four month old to eighteen month old. It was also suggested that a relationship between development/exacerbation of AD and skin barrier dysfunction.

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Validity for atopic dermatitis diagnostic criteria in an adult hospital population

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Background: In contrast to the advancement of knowledge on the pathogenesis of atopic dermatitis (AD), establishing international diagnostic criteria standards remains a challenge. The criteria of Hanifin and Rajka, though extensively mentioned in research settings, rely on features with significant variation due to age, ethnicity or other factors and may be unsuitable for epidemiological studies when clear-cut case/control definitions are needed. The validity of the UK Working Party criteria has been reported in population and hospital-based studies, mostly in children. Data concerning adults are however very limited.

Objective: We aimed to test the validity of the UK Working Party criteria in adult patients referred to a university hospital in Bulgaria. We also tested the relevance of six clinical signs commonly associated with AD without being criteria, namely, fine hair, periorbital and periauricular dermatitis, infraauricular fissures, cheilitis angularis and Hertoghe sign.

Methods: 118 patients aged 18–60 years were recruited for the study. All patients were examined independently by a senior dermatologist and a resident, both blinded to the study aim. The results were evaluated by a third independent and blinded observer using contingency tables.

Results: We found higher sensitivity (94,03%), specificity (92,16%), positive (94,03%) and negative (92,16%) predictive value for the UK Working Party diagnostic criteria than for the ones of Hanifin and Rajka (88,06%;82,35%; 86,76;55,26 respectively). Family history for atopy and erythroderma were significant factors for false positive results. Of the additionally examined features, the highest sensitivity and specificity was found for "infraauricular fissures" (59,70% and 84,31%, respectively).

Conclusion: Our results confirm the validity of the UK Working Party criteria in an adult hospital population in Bulgaria. Although the cases may represent a more severe disease end spectrum, there are only few systematic publications on AD features in adulthood and our study provides valuable data on the performance of the diagnostic criteria and relevance of the individual clinical signs in this age group.

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Is atopic dermatitis really atopic?

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Background: There is little data on the correlation between atopic dermatitis and other atopic diseases (asthma, rhinitis) and atopy markers (positive skin prick test and specific IgE to Dermatophagoides pteronissinus /Dpt./). This is why the comorbidity of atopic dermatitis and allergic rhinitis and bronchial asthma and the presence of atopy markers should be further investigated as well as the correlation between the positive atopy markers and a positive patch test with Dpt.

Methods: The correlation between patch-test and skin prick test with Dpt. and specific IgE was evaluated in 32 patients (8 men), with atopic dermatitis, 27 had rhinitis as a concomitant disease (17 with allergic rhinitis) and 16 had bronchial asthma. All of them were tested for specific IgE and skin prick test and patch test with Dpt. were performed.

Results: 17 patients had a positive patch test with Dpt., 12 had positive specific IgE antibodies to Dpt. and 14 - positive skin prick test with Dpt.

Conclusion: Cases with a positive patch test prevail when compared to positive skin prick test and specific IgE. Positive patch test does not always correlate with atopy, evaluated as positive skin prick tests and specific IgE to Dpt. The patch test could be used as a routine and reliable method in practice for evaluating the role of Dpt. in the pathogenesis of atopic dermatitis.

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Atopic dermatitis in children: immunophenotype of inflammatory cells in skin lesions

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Background: Immune cells play a main role in development of atopic dermatitis (AD); however, so far only limited data are documented on the distribution on these cells in the skin during cutaneous inflammation.

Objectives: To gain better insight into the number and phenotype of immune cells in lesional skin of AD patients.

Methods: The lesional skin biopsies of patients with different forms of atopic dermatitis (an exudative, an erytemato-squamouse, a lichenoid) has been investigated. It is carried out 38 skin's biopsies in the children aged 1,5–15 years. Biopsies were made by needles (diameter - 1,6MM). Avidin-biotin immunoperoxidase staining of paraffin-embedded skin sections, with quantitative counting of cells labeled by anti-CD4, anti-CD8, anti-CD16, and anti-HLA-DR (Human Leukocyte Antigen) anti-IgE monoclonal antibodies was used.

Results: The results of immunomorphologycal analysis showed a great infiltration of T-lymphocytes and a high intraepidermal expression of IgE. The amount of main immune cells were (per 100 cells): CD4 = 47 ± 5,8; CD8 = 17,3 ± 3,2; CD4:CD8 = 2,8 ± 0,5; CD16 = 5,7 ± 0,7; HLA-DR = 1,1 ± 0,1.

Conclusion: Immunophenotyping was found to be a useful diagnostic method in AD patients. These data help to elucidate the pathogenesis of AD.

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Undiagnosed alllergic dermatitis - a prospective study

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Introduction: Undiagnosed cases of dermatitis were referred to our clinic after treated by number of dermatologists in our city. These cases were treated with different types of symptomatic drugs for last 7 to 10 yrs, but did not yield any significant result and these were labeled as difficult dermatitis.

Materials and Methods: Ten such patients who were referred to our clinic were subjected to detailed medical history & clinical examinations. We decided to perform skin prick test (modified SPT) as to determine possible

role of airborne allergens. These patients showed positive skin prick test reactions to many allergens such as Asp. flavus, Asp.Tarmare, Alt Alternaria, D. Farinae, D.Pter., House dust, Hay dust, Parthenium & Paltophorm among airborne allergens, while reactions were observed significantly positive to certain food allergens also (Milk, Almond, Yeast, Hazel nut).Since there patients showed strong reactivity to certain allergens (IgE mediated hypersensitivity), an attempt was made to administer allergen immunotherapy course. Allergens which were strongly positive and showed airborne dominance in the vicinity of the patient were selected for allergen immunotherapy mixture. The allergen immunotherapy was initiated on this subject as per guidelines laid down by WHO.

Results and Discussion: The improvement was observed & symptomatic relief was recorded within 3 to 4 month of initiation of allergen immunotherapy.

Conclusion: Allergen immunotherapy could play a major role in the treatment of airborne contact dermatitis.

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Prognosis of allergic contact dermatitis in patients patch tested in a university hospital in Beijing

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Background: Although it is believed that the prognosis of allergic contact dermatitis (ACD) is good, long-term prognosis of ACD has not been reported.

Objectives: To investigate the 1-year outcome of ACD in a university dermatology setting.

Patients and Methods: In a two-year period, 655 consecutive patients were patch tested in Peking University Third Hospital for suspected ACD. At one year after patch testing, all patients were asked a revisit and prognosis of ACD was evaluated by the rate of clearance (complete free of dermatitis without recurrence for more than 3 month before the final evaluation).

Results: Of 599 patients finished study, 167 cases were ACD. The rates of clearance and relapse were 49.7% and 41.9% respectively. Concurrence of atopic diathesis, ichthyosis, history of drug allergy as well as disease location had no effect on the clearance rate. Significant lower clearance rate (26.8%) was found in patients with longer disease duration (over 6 months) before diagnosis. The clearance rate in patients with non-continuous ACD was also lower than that of continuous ACD (28.3% vs. 59.6%, chi square test). Ignorant re-exposure to contact allergens was the mean reason for relapse.

Conclusion: 1-year outcome of ACD is not as good as expected. Longer disease duration before diagnosis and non-continuous ACD are main risk factors for poor prognosis. Patients with suspected ACD should be patch tested as early as possible. Mandatory ingredient labeling and sufficient patient education are necessary to improve the prognosis of ACD.

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Fishing for allergens hiding as prohaptens: cinnamic aldehyde but not cinnamic alcohol identified as potent sensitizer

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Skin is a major target organ of allergic reactions to small molecular weight compounds such as allergic contact dermatitis or allergic drug allergy. It has been generally accepted that most small molecular weight compounds must be bound to high molecular weight compounds in order to become immunogenic. For this binding of the hapten to a protein the hapten must be a highly reactive chemical such as intermediate metabolites of cytochrome P450 (CYP) dependent metabolism of prohaptens. Recently we demonstrated the usefulness of an organotypic CYP cocktail to identify prohaptens by combining such a CYP cocktail with a dendritic cell-based in vitro model to identify sensitizing small molecular weight compounds (Bergström et al., JID 127 (2007) 1145–1153; Mukhtar et al., JID (2007) 992–993). Cinnamic alcohol is oxidised to cinnamic aldehyde by a CYP-dependent metabolism. Therefore we were interested whether cinnamic aldehyde is the nominative antigen in allergic reactions to cinnamic alcohol. We generated immature DCs from human peripheral blood monocytes by depletion of CD2, CD7, CD19, CD56, CD16, CD235a positive leukocytes and consecutive incubation with GM-CSF and IL-4. At least 6 million cells were stimulated with the following test compounds for 30 hours respectively: 2,4,6-trinitrobenzene sulfonic acid - TNBS (200 mg/ml), sodium dodecyl sulfate - SDS (5 mg/ml), dimethyl sulfoxide - DMSO (0.1%), CAld (0,1 mM) and CAlc (0.1 mM). After RNA isolation of the treated cells, reverse transcriptase-PCR analysis was performed using a primer/probe set specific for the detection of IL-8 mRNA expression. A sensitizing effect of CAld could be shown for all 6 donors as indicated by a median IL-8 upregulation of 34.3 fold normalized relative to medium-treated controls. In contrast, the median enhancement of IL-8 mRNA expression was not significantly greater after incubation with CAlc (2.13) than after stimulation with SDS (2.22), DMSO (1.2). In conclusion, cinnamic aldehyde possesses a stronger allergenic activity than cinnamic alcohol which is CYP-dependently metabolised to cinnamic aldehyde. These results further supports the concept that the combination of prohapten modified organotypic CYP cocktails with a dendritic cell-based in vitro model of contact allergens may streamline and increase the predictive efficiency of current safety test methods in allergic contact dermatitis.

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The profile of patch test reactions to common contact allergens is related to sex

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Background: It is a worldwide observation that patients exposed to diagnostic patch tests are predominantly women. This is commonly explained by distinct sex-related patterns of allergen exposure that cause different patterns of sensitization in men and women and by a higher health awareness of women. Nevertheless, additional sex-related differences in the responsiveness to contact allergens should be taken into consideration.

Methods: Sex-related reaction profiles of 16 common patch test allergens that are used in standard patch test series were retrospectively analysed based on data of 47.000 patients filed by the Information Network of Departments of Dermatology in Germany within a period of 10 years. All patch tests had been done by use of identical methods in patients with suspected contact allergy but no history of atopic dermatitis. The proportions of weak positive reactions and of questionable and irritant reactions were evaluated by calculating the positivity ratio and the reaction index separately for men and women for each allergen.

Results: Out of the 16 allergens evaluated, 8 had a slightly higher positivity ratio and a slightly higher reaction index in women than in men. 4 allergens had a lower positivity ratio and another 4 had a lower reaction index in women than in men, but no allergen had a lower positivity ratio plus a lower reaction index in women than in men. In particular for allergens with similar rates of positive reactions in men and women female sex was significantly related with a higher positivity ratio and reaction index ($p<0.01$).

Conclusion: There is a small but statistically significant disparity in the reactivity of men and women to common patch test allergens. Women in general have a higher rate of weak positive reactions but less questionable or irritant reactions to an allergen than men. This marginal difference is probably not relevant for patch testing but adds a new element to the pathogenetic puzzle of contact allergy.

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Reduced allergy rates to contact allergens/haptens with both oral and cutaneous exposure in atopic eczema subjects. Atopy and the "hapten hypothesis"

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Background: Whilst allergy to food proteins is almost exclusively seen in atopics, it has been our impression that this does not hold true for haptens/contact allergens which are present in our diet. Diallyl disulfide is the major haptenic allergen in garlic and allergic contact dermatitis to this has been observed in people who handle garlic whilst preparing food. Parabens and lanolin are both contact allergens commonly used in many cosmetic and medicament creams. However, unlike lanolin, the preservative parabens is used in some processed foods. We retrospectively reviewed data from our patch test clinic, comparing the frequency of allergy rates (patch test positive, PT+) to these haptens between atopic eczema/dermatitis AD and non-AD dermatitis patients.

Results: Between 1980 and 2006 in total population of 36658 patients with eczema/dermatitis were patch tested, of whom 10326 (28.2%) had AD; in contrast 13/83 (12.5 %) patients PT+ to diallyl disulfide/garlic had AD (AD/total population vs AD/diallyl disulfide PT+ $p = 0.011$). 54/239 parabens PT+ had AD (22.6%), whilst 181/608 lanolin PT+ had AD (29.8%) ($p< 0.05$)

Discussion: Frequency of contact allergy to haptens with both oral and skin exposure is reduced in AD patients compared to non-AD patients in direct contrast to food protein allergy. This decrease was not observed in lanolin, which is used only on the skin. Possible reasons for these results could be 1) confounding factors e.g. AD patients handle garlic less than non-AD patients, or 2) AD patients are very efficient at tolerising haptens, and this is in some way secondary to their atopic status, or 3) oral tolerance of haptens may in some way antagonise tolerance of proteins and contribute to the development of atopy (the hapten hypothesis). We note that the increase in atopy over the last 50 years also coincides with a period where processed food and milk, with chemical/hapten content, is used in western diets.

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Incidence and prevalence of para-phenylenediamine allergy in an adult Thai population: a public health problem

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Background: Most studies on the prevalence of allergy to the permanent hair dye chemical para-phenylenediamine (PPD) are reported from populations of eczema patients attending patch-test clinics, and are assumed to be much higher than in the normal population. No data exists on incidence of sensitisation to PPD resulting from the use of commercial hair dye preparations over a defined time period.

Methods: 2545 healthy adult volunteers (Bangkok) were screened for PPD allergy through standard patch testing. Volunteers not allergic to PPD were then recruited into two groups; one group applying a commercial hair dye brand as instructed on a monthly basis for 6 months ($n = 548$); controls ($n = 516$) were instructed not to dye their hair for 6 months. Sensitisation to PPD resulting from the use of hair dye over this period was then detected by repeat patch testing.

Results: The prevalence of PPD allergy in a normal adult population was 2.7% ($m = 2.4\%$, $f = 3.2\%$). Projected to the adult Thai population, at least 1,000,000 Thai individuals could be allergic to PPD. The incidence of sensitisation through monthly application of standard commercial hair dye preparations over a 6 month period was 1.3%, substantially higher than in controls (0.4%), although numbers were small and not statistically significant.

Interpretation: There is a higher prevalence of hair dye allergy amongst the normal population than previously thought. The incidence of new cases of PPD allergy would indicate that current regulations and practice of hair dye exposure lead to PPD sensitisation and allergy, which is a public health problem.

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An unusual form of autoimmune progesterone dermatitis (APD): the role of diagnostic challange test

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Hypersensitivity to sex hormones has long been recognized as cyclic and recurrent rushes. Progesterone sensitivity is termed as autoimmune progesterone dermatitis (APD) which is shown with a positive skin test . Our patient was a 33-year-old female who presented with cyclic skin eruptions of two years' duration, right after she delivered her baby. At the beginning a fixed single maculopapular and itchy skin lesion of 1-2 cm was occuring two days prior to her monthly cycles and resolving by the beginning of mens. However, new lesions occured in other fingers, lately. She had no prior exogenous hormone use. Her menstruel cycles were irregular since she was 15 years old. Progesterone levels were found to be high before her pregnancy, but wasn't treated. The possibility of APD was suspected because of the cyclic nature of her lesions.

Serum levels of sex hormones were normal, as well as her gonadotrophins, and prolactin. Skin biopsy showed a non-spesific dermatitis with a moderate perivasculer infiltration of lenfocytic cells in the upper dermis. The direct immunofluorescence study was negative. Challenge test with aqueous progesterone was performed. Prick and patch tests were negative, whereas intradermal test with 50 mg/ml of progesterone was positive confirming the diagnosis of APD. Despite many localizations reported, there wasn't any case appearing only in fingers, as it was seen in this case.Exposure to exogeneous progesterone as oral contraceptives (OC) has been suggested as a stimulus for the production of autoantibodies reacting with endogeneous progesterone in APD. However, in some cases OC use isn't necessary as was observed in our patient. There is a possibility of autoimmune damage to progesterone containing ovarian tissue which could be resulted with premature ovarian failure. In our patient there was no evidence of ovarian disfunction as she had a child.

In this case, APD was diagnosed with patient's typical history of recurrent lesions during the luteal phase of her menstrual cycle and this was confirmed by intradermal test with progesterone. As the endogenous progesterone is produced during ovulatory cyclus, the goal of therapy is the suppression of ovulation by a combination OC. If this is ineffective, danazol, gonadotropin releasing hormone analogs, tamoxifen, and oophorectomy may be tried. Our patient didn't accept any treatment as she wanted another baby.

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Analysis of discoidin domain receptor-1 expression by tissue-infiltrating eosinophils in allergic skin diseases

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Background: Discoidin domain receptor 1 (DDR1) is a receptor tyrosine kinase that is activated on binding to its ligand-collagen a component of the extracellular matrix (ECM) in the organs and is known to be constitutively expressed in the normal tissues such as the lungs, kidneys, colon, and brain. It has been reported that the tissue-infiltrating eosinophils expressed DDR1 and interaction between endogenous DDR1 on eosinophils and collagen in the ECM might affect eosinophil survival in the tissue microenvironment through NFK-B activation in Churg-Strauss syndrome (CSS) characterized by necrotizing granulomatous angitis with massive eosinophil infiltration, asthma, and hypereosinophilia.

Patients and Methods: DDR1 expression on peripheral blood eosinophils from asthma patients, and healthy volunteers has been reported to be significantly lower than that from CSS patient. However little is known about the DDR1 expression and its significance in tissue infiltrating eosinophils in allergic diseases. Therefore, we analysed DDR1 expression by tissue-infiltrating eosinophils in allergic skin diseases such as atopic dermatitis, drug eruption and Kimura's disease with eosinophil infiltration.

Results and Conclusion: DDR1 was significantly expressed by eosinophils in the lesional skin of CSS, but not atopic dermatitis, drug eruption or Kimura's disease. Our current results indicate that DDR1 expression by eosinophils in the local inflammatory sites is specific for CSS and could be the disease marker in allergic skin diseases with eosinophil infiltration.Mechanisms of eosinophil survival and activation might be different between CSS and allergic skin disease such as atopic dermatitis.

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Gene-gene interaction between IL-13 and IL-13 receptor α1 is associated with total IgE in Korean children with atopic dermatitis

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Background: Interlukin(IL)-13, which is essential for IgE synthesis, mediates its effects by binding with a receptor composed of IL-4 receptor(R) α and IL-13Rα1. We investigated the effects of IL-13 and IL-13Rα1 polymorphisms in Korean children with atopic dermatitis, and whether these have been associated with IgE production.

Methods: We enrolled 567 children with atopic dermatitis and 172 nonatopic healthy children. IL-13 and IL-13Rα1 genotypes were identified using PCR-RFLP method.

Results: There was an association between the protection of atopic dermatitis and homozygosity for risk allele of IL-13Rα1 A+1398G(adjusted odds ratio of 0.69[95% CI, 0.47–1.00]), but the genotype of IL-13 G+2044A was not associated with the development of atopic dermatitis. The hetero-or homozygous for +2044A in IL-13 and +1398G in IL-13Rα1 tended to higher total serum IgE levels versus those homozygous for +2044G in IL-13 and + 1398A in IL-13Rα1, but not to a significant degree. The gene-gene interaction between risk alleles of IL-13 G+2044A and IL-13Rα1 A+1398G polymorphism was associated with higher total IgE in children with atopic dermatitis($P = 0.038$).

Conclusion: These findings indicate that the IL-13Rα1 A+1398G may be associated with the development of atopic dermatitis and IL-13 and IL-13Rα1 polymorphisms may interact to enhance IgE production.

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Age-specific high serum IgE levels are associated with worse symptomatology in children with atopic dermatitis

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Background: Atopic dermatitis (AD) is a distressing disease associated with excoriations, pruritus, sleep disturbance and elevation of serum total IgE levels.

Objective: We evaluated if serum IgE levels correlate with the symptomatology and plasma chemokine levels in children with AD.
Methods: AD patients younger than 18 years old were recruited from the paediatric dermatology clinic of a university teaching hospital and AD severity evaluated with the SCORing Atopic Dermatitis (SCORAD) index. Concentrations of serum total IgE, eosinophil count and plasma AD-associated chemokines (cutaneous T cell attracting cytokine, CTACK; thymus and activation regulated chemokine, TARC) were measured.
Results: One hundred and seventeen Chinese children with AD (64 boys and 53 girls), with mean (SD) age of 10.7 (4.4) years, were recruited. Their mean (SD) overall SCORAD was 51.1 (22.8). Æ (defined as total serum IgE level divided by age-specific upper limit) correlated well with the extent and intensity of AD except for oozing/crusting which was significant only in the males. There was significant correlation for Æ with pruritus or sleep loss only in the females. 82 (70%) of patients reported history of allergic rhinitis and 35 (30%) reported history of asthma or hyperactive airway disease. In a subgroup of patients who had never had allergic rhinitis, asthma or urticaria (n = 26), log-transformed Æ correlated well with the objective SCORAD (Spearman's rho = 0.554, p = 0.003).Levels of IgE, CTACK, and TARC and eosinophil count differed significantly among patients with mild, moderate and severe disease. Æ correlated well with TARC (r = 0.50, p < 0.001) and eosinophil count (r = 0.41, p < 0.001) but not CTACK (r = 0.11, p = 0.270). The prediction of moderate-to-severe eczema by Æ gave an area under the receiver-operating characteristic curve of 0.76 (95% CI 0.65–0.86, p = 0.004). An optimum positive predictive value of 94.2% was achieved with a cut-off point of Æ of 2.95, sensitivity of 75.0% and specificity of 66.7%.
Conclusion: Æ correlates well with the objective clinical and serum TARC level, and may serve as an overall marker for disease severity. Furthermore, Æ of 2.95 predicts moderate-to-severe disease. Unlike the chemokines, Serum IgE measurement is inexpensive and readily available in most clinical service. IgE is not only a laboratory marker for atopy but also a parameter of disease severity.

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Soy intake and reduced prevalence of skin allergic deseases symptoms

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Introduction: Soy is one of the few foods that contain important levels of linoleic acid, an omega-3 polyunsaturated fatty acid (O3FA). It is known that O3FA reduces the production of arachidonic acid derivates which are able to reduce chronic inflammatory response.
Objective: To evaluate the association between dietary soy intake and the presence of skin allergic symptoms in adults.
Materials and Methods: We conducted a cross-sectional study of 765 students from Centro Educativo Latinoamericano University in Rosario, randomly chosen, 638 females (83.4%) and 127 males (16.6%), aged from 17 to 65 years old (x = 21.42 ± 4.18). They answered questionnaires about urticaria/angioedema, contact dermatitis, atopic dermatitis and about the frequency of soyfood intake.
Results: Differences were found when the food intake was classified into two groups: A (never or less than once a month) and B (once or more than once a month, week or day). For soy-based mayonnaises and margarines intake, 48.1% of the subjects were found in group B and the prevalence of skin diseases within this group was significantly lower than in group A; for example atopic dermatitis: B = 22.1% vs A = 28.7% (OR = 0.71; IC:0.51–0.99; p<0.05), contact dermatitis: B=14.3% vs A = 22.8% (OR = 0.57; IC:0.38–0.85; p<0.05) and food angioedema: B = 12.0% vs A = 40.0% (OR = 0.20; IC:0.03–1.01; p<0.05). For soy hamburgers and soy breadcrumbs intake, 70.0% of the subjects were found in group B and the prevalence of the different skin diseases within this group was significantly lower than in group A; for example eczema symptoms: B = 10.2% vs A = 15.7% (OR = 0.61; IC:0.38–0.99; p<0.05), medical diagnostic eczema: B = 15.0% vs A = 21.3% (OR = 0.65; IC:0.42–0.99; p<0.05), contact dermatitis: B = 16.6% vs A = 23.4% (OR = 0.64; IC:0.43–0.95; p<0.05) and food angioedema: B = 13.8% vs A = 66.6% (OR = 0.08; IC:0.01–0.74; p<0.05). Group B for soy oil ingestion was only 19.5% and in this group the prevalence of angioedema symptoms (1.3%) was lower than in the rest of groups (5.9%) (OR = 0.22; IC:0.04–0.96; p<0.05). The soyfood ingestion was significantly higher in male and men presented significantly less prevalence of the symptoms studied. The statistical significance remained the same after adjustment for sex and ingestion of histamine-releasing food. No differences were found between soy intake and urticaria symptoms.
Conclusion: The results obtained allow us to estimate that the intake of a soy rich food, at least once a month, could protect against some skin disorders.

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Is house dust mite sensitivity a factor in chronic urticaria?

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Introduction: chronic urticaria is the most common cutaneous disorder seen in outpatient allergy clinics. There are a few reports associating house dust mite sensitivity with chronic urticaria. This study investigates the possible association between house dust mite sensitivity and chronic urticaria.
Methods: In this study four groups of patients were enrolled. Group I: Chronic urticaria (35 subjects). Group II: allergic rhinitis(135 subjects). Group III: asthmatic patients (14 subjects).Group 4: allergic rhinitis +asthma(12 subjects). Group II, III and IV considered as positive controls. All of the patients underwent skin prick testing with antigens of the house dust mite, Dermatophagoides pteronyssinus (DP) and Dermatophagoides farinae (DF), with positive and negative controls.
Results: In Group I, 13/35 (37.1%): In group II 64/135(47.4%); In group III 5/14(35.7%) and in group IV 4/12(33.3%) patients had skin sensitivity to house dust mites .There were no statistical differences between prevalence of positive skin test to mite in four groups.
Conclusion: we suggest a possible association of house dust mite sensitivity with chronic urticaria.

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Sensitisation and colonisation status of Malassezia in patients with atopic dermatitis

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Background: Malassezia represent a contributing factor in AE. Recently, the genus Malassezia was classified into eleven species based on their morphological, biochemical, and genetic characteristics. It is still unknown how, where and why patients suffering from AE become sensitized to allergens of this lopophilic yeast, it becomes evident that sensitization to Malassezia particularly M. Sympodialis occurs almost exclusively in AE patients. The success of antimycotic treatment in some patients with AD indicates that Malassezia is involved in the pathogenesis of AD.
Methods: We collected skin scales from 52 patients with atopic eczema. Specific IgE was measured against Malassezia using commercial kit (m70 and m 223). Presence of the various Malassezia species were analysed by culture, microscopically examination and a real-time PCR based method for testing qualities and quantities of patient skin scales so as to detect directly and confirm the identity of Malassezia species.

Results: In 33 of 52 patients Malassezia PCR was positive. The presence of Malassezia did not correlate with the level of specific IgE found against this yeast. Also we did not detect a correlation between severity of AD and levels of specific IgE. However response to systemic antifungals was more prone in patients with high levels of specific IgE against Malassezia. The presence of M. sympodialis, M. globosa and M. restricta was highly variable.
Conclusion: We identified three different species of Malassezia colonizing the skin and leading to IgE-mediated sensitization in atopic eczema. Further analysis of quantitative fungal load will probably help to identify AE patients that benefit from antifungal therapy.

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Frequency of selection MRSA-strains in composition of a microbiocenosis of a skin the patient's atopic dermatitis

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Background: to Define a share of allocation MRSA-strains and structure of a microbiocenosis of a skin of patients an atopic dermatitis. To define properties staphylococcus with skin of patients.
Methods: 63 patients an atopic dermatitis are examined at the age from 3 till 35 years. Degree of a gravity of the patients defined with use of the international standard - index SCORAD (Scoring of Atopic Dermatitis). Research of microbiocenosis of skin is carried out bacteriological method. 155 strains staphylococcus with skin of patients are investigated. Are checked up on stability to antibiotics by method of disks on cups Petri. Activity catalases in lysates staphylococcus by a colorimetric method on Sinha A. K. The statistical analysis of results is executed by means of statistical programs Microsoft Excel 2000 and «Statistica 6» StatSoft.
Results: As a part of a skin microbiocenosis in 84, 2% was *Staphylococcus aureus*, in 15, 8% was coagulase negative *staphylococcus*. High frequency of occurrence MRSA-strains (75%) in structure microbiocenosis skin of patients is revealed. High frequency has been found out among MRSA, repeatedly from steady pressure to antibiotics.
Conclusion: As a part of a microbiocenosis with skin of patients sick an atopic dermatitis dominate MRSA-strains. Level of activity for pressure MRSA correlates catalases with their ability to survive in an organism. More poisonous MRSA also are more often allocated from patients with heavy degree (SCORAD 87,6 ± 1,4).

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Phenotypic features of coccal microflora of skin in the norm and with atopic dermatitis

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Background: Phenotypic features of coccal microflora of skin and disease severity are correlated in children with atopic dermatitis (AD).
Methods: Forty eight AD patients (age 0, 1–16 years) were recruited. The SCORing Atopic Dermatitis (SCOARD) we used for clinical score for assessing AD symptomatology. The skin analyzed for Staphylococcus aureus and coagulase-negative staphylococci detecting. Ig-protease, collagenase, lysozyme and DNAase activities of staphylococcal isolates were studied. Methicillin-resistant Staphylococcus aureus isolates were determined by disk diffusion method.
Results: S.aureus colonization increased in skin with AD with increasing SCORAD - 5,2 ± 1,2 log10 CFU/sm2 (SCORAD = 35,5 ± 5,5) and 5,7 ± 0,8 log10 CFU/sm2 (SCORAD = 91,9 ± 5,8). 41% staphylococcal isolates from skin with moderate AD (SCORAD = 35,5 ± 5,5) and 100% isolates from skin with severe AD (SCORAD = 91,9 ± 5,8) had DNAase activity. Also Ig-protease, collagenase activities of staphylococcal isolates from skin with atopic dermatitis were higher then from skin in the norm.
Conclusion: Virulence factors of staphylococcal isolates correlated with SCOARD index and disease severity. And staphylococcal isolates from skin with AD were essentially aggressive in comparison with isolates from healthy skin

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The role of fecal microflora in children with allergic eczema/dermatitis syndrome

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Rationale: The role of intestinal microflora in children with allergic eczema dermatitis syndrome (AEDS) is still disputable, although a beneficial influence on AEDS course of certain intestinal bacteria, administered as probiotics has been described in intervention studies. The purpose was to investigate the relationship between gut microbiota and sensitization and cytokines production in children with AEDS.
Materials and Methods: A case-control study with 57 children (4–7 years) having AEDS and 55 age-matched healthy control subjects was conducted. For differential diagnosis we used Skin Prick Test (SPT) with common allergens, total and specific IgE. IL-5, IL-10, γ-IFN levels were assessed by ELISA. All patients had investigation of gut microflora.
Results: In children with AEDS were found significantly low counts of *Bifidobacterium* comparing to healthy control subjects (10^5–10^6 vs 10^9–10^{10} CFU/g) as well as Lactobacillus (10^5 vs 10^7–10^8 CFU/g) ($p<0.05$). At the same time atopic children presented high level of *Escherichia coli*, *Staphylococcus aureus*, *Clostridium dificile*, *Candida albicans* in investigated microflora profile. Furthermore AEDS children demonstrated positive SPT to common allergen (especially food allergen); increased level of total and food specific IgE (mostly - egg, milk, peanut, hazelnut). In this group were shown increased production of IL-5 to 192,3 ± 6,1 pkg/ml (N = 74,3 ± 3,3 pkg/ml); IL-10 to 153 ± 9 pkg/ml (N = 5,8 ± 0,25 pkg/ml) and decreased production of γ-IFN to 241 ± 6 pkg/ml (N = 331 ± 35 pkg/ml) ($p <0,05$).
Conclusion: This investigation demonstrated the relationship between perturbation in intestinal microbiota and IgE sensitization in children with AEDS. There is an augmentation of Th-2 type lymphocyte function reflected by increased IL-5 and IL-10 production in AEDS children. Disorder of the intestinal microflora might play a role in the onset of atopic eczema.

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Linear dermatitis reactions to paederus beetles (coleoptera: staphylinidae) in Kazeron, Iran

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Background: Dermal irritation resulting from contact with arthropods or infested products is well documented and often occupational in nature with entomologist, food handlers and others. Between them, linear dermatitis is a

self-healing blistering disorder of the skin caused by a small insect belonging to genus Paederus, family Staphylinidae, order Coleoptera. It appears on the skin by hemolymph of Paederus beetles. This study was undertaken to identify the dominant species of paederus beetles and to report the expose parts of the body in the study area.

Methods: Peaderus beetles were collected by hand and UV black rays every month for entomological studies. For medical studies questionnaires were completed by physicians in health centers in the studied areas.

Results: By this investigation, it was revealed that *Paederus ilsae* Bernhauer and *paederus iliensis* Coiffait are two species that were identified for the first time in the studied area. This research has revealed the 27.02%, 23.42%, 18.01%, 17.11% and 14.41% cases of linear dermatitis occurred in face, neck, eyes, hands and legs respectively in kazeron area. Furthermore, 42.6% of cases also had secondary infections later.

Conclusion: Public awareness of how to avoid contact with the adult beetles is considered to be the best way to reduce the cases of linear dermatitis.

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A new cause of facial eczema: the ferret

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Observation: a 36 year old patient has had for one year a face eczema, mainly on eyelids and lips. She had no history of allergy and no medication. She was a beautician and suspected a professional sensitization but the eczema persisted during her holidays. She had been exposed to numerous perfumed products in her institute for six years. For many years, she had had horses and pets: dogs, cats, and for two years a ferret.

Patch-tests of the standard, cosmetic product, methacrylate and fragrance series, with the products brought by the patient (creams, perfumes, household and professional products) were negative at 48 and 96 hours. Prick-tests with aeroallergens, molds, latex and 28 foodstuffs were negative. Specific IgE for ferret were undetectable with normal total IgE. The ferret was given to one of her friend for two months and the eczema dramatically improved. When she had a short new contact with the ferret the eczema relapsed.

Conclusion: a few cases of asthma or urticaria caused by ferrets have been reported but there is no former case of eczema. The ferret belongs to the fissiped family (mink, marten, ermine and polecat). Theorically a cross reactivity between them could occur but no case has been yet reported. The culprit allergens are not well known (albumin?).

Ferrets, like felides, lick themselves and the salivary proteins could be the allergens (protein contact dermatitis?).

We emphasize that sensitization to ferret, that is a new pet, has to be evoked in case of eczema or asthma, even when specific allergological tests are negative.

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Allergic contact dermatitis due to Propolis

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Propolis is a product made by bees from various vegetal sources, mainly from the resin of the poplar; it is well known as an inducer of occupational dermatitis in apicultors. Its use as a natural cosmetic is widespread. given its antiinflammatory and antiseptic properties.

We present the case of a 24 years old woman without an atopic background who presented an eczematous, itchy rash in the areas where she had used a cosmetic that had Propolis among its ingredients. Patch test were performed with True Test series, the cosmetic series and Propolis. The tests were positive for Propolis (+++ at 48 and 96h), and were negative for the other contact allergens, including Balsam of Peru.

The sensitizing capacity of Propolis is mainly attributed to the methylbutenil cafetate and to phenylethil cafetate mainly. It has cross-reactivity with the Balsam of Peru with which it shares at least 13 components. We therefore present an allergic contact dermatitis to Propolis in a "natural" cosmetic in a patient who is not an apicultor and who has no relationship with this hobby.

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Adverse reactions to dark chocolate - a case report

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Background: Atopic dermatitis (AD) is a chronic inflammatory skin disorder characterized by pruritus, inflammation, lichenifications, and typically distributed eczematous lesions. Environmental factors as well as allergens from various sources may trigger skin inflammation. The exact cause of AD is unknown, but immunological and psychological factors should be taken into account. We report a case of a 28-year old female suffering from atopic dermatitis with a history of recurrent episodes of pruritus, urticaria and facial angioedema starting 6-12 hours after eating dark chocolate pralines. With the elimination of milk chocolate and dark chocolate from the diet no more episodes occurred. White chocolate is well tolerated and consumed frequently.

Methods: Skin prick tests to commercial extracts of common aeroallergens and food allergens were performed. Furthermore prick to prick tests using white chocolate, milk chocolate, dark chocolate (65% cocoa powder and 85% cocoa powder), and cocoa powder were carried out. Specific IgE, IgG1 and IgG4 levels to a large panel of purified food allergens from milk, egg, peanut, hazelnut, soy, peach, apple, celery, and chocolate extracts were determined by ELISA experiments.

Results: Skin prick tests with commercial extracts were negative. IgE and IgG1 levels to various food allergens and chocolate extracts were not increased compared to non atopic individuals, whereas IgG4 levels to all tested allergens and chocolate extracts were significantly higher than in control sera. Prick to prick tests with chocolate brands revealed erythema with increasing intensities according to higher contents of cocoa powder. White chocolate (without cocoa powder) did not elicit any skin reactions.

Conclusion: This case report illustrates the complexity of diagnosing adverse reactions to foods. Ingestion of dark chocolate elicits angioedema and AD, while white chocolate is tolerated. Even in the absence of allergen-specific IgE, an allergen-specific Th2 reactivity (high IgG4 levels) can be observed.

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Case report-contact dermatitis to perfume

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Background: Dermatitis is an inflammation of the skin. Contact dermatitis is a localized rash or irritation of the skin caused by contact with a foreign substance. Substances that cause contact dermatitis in many people include "poisonous" plants such as poison ivy, certain foods, some metals, cleaning solutions, detergents, cosmetics, perfumes, industrial chemicals, and latex rubber. Allergic contact dermatitis is a type IVa1 T-cell-mediated hypersensitivity.

Case Report: A 26-year-old female with clinical signs of persistent allergic rhinitis (sneezing, itching and rhinorrhea). She presented a severe eruption with intense itching after using her new perfume. Clinical aspect of skin

lesions were: erythema, papules and vesicles on the neck and chest with augmentation of symptoms after solar exposure. The manifestations have begun after 24 hours of exposure to the new perfume.

Results: Skin prick tests revealed an atopic patient with sensitization to Alternaria. Skin patch tests with european standards to cosmetic fragrances (fragrance mix, balsam of Peru), cosmetic preservatives (paraben mix, quaternium) and other cosmetic ingredients (colophony, thiomerosol, ethylenediamine, formaldehyde) and her own perfume revealed late sensitization (72 hours) to her perfume and fragrance mix. We recomanded to avoid contact with known allergens and treatment with H1 anihistamine and topical steroids.

Conclusion: The patient with contact dermatitis may be very uncomfortable and have poor quality of life. The best treatment is to identify and avoid the substances that may have caused the allergic reaction.

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Influence of Psidium guajava tea for atopic dermatitis

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I previously reported the antihistaminic effect and anti-leukotriene effect of Psidium guajava(PG) which was a native plant of North America. Thus I made PG tea and perviewed an effect to atopic dermatitis.

Methods: Seventy-five patients with atopic dermatitis who were treating in Sanno Hospital,were enrolled in this study. Ages of them 7 to 34 years old. PG tea made 0.5g with hot water of 200ml and drank it three times per day. After one week observation without any treatment, I used PG tea for fours weeks and skin condition. And I measured eosinophil cationic protein (ECP) values and histamine levels of blood.

Results: Forty-two (56%) improved itching of skin and Thirty (40%) improved skin condition. ECP valued in serum were decreased from 82.4 ± 42.4ng/ml to 21.8 ± 23.4ng/ml. The Histamine levels in serum decreased from 211.0 ± 50.4ng/ml to 98.6 ± 52.4ng/ml.

Conclusion: I investigated the effects of PG tea for atopic dermatitis. This study showed it was able to improve the clinical features of this disease significantly. Furthermore,the ECP values and histamine levels in serum which are indexes of allergic inflammation were decreased. I concluded from these results that Pg tea is an Effects method for the treatment of atopic dermatitis.

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The role of contact sensitization in contact dermatitis patients among the seaweed farmers in Bantaeng, South Sulawesi, Indonesia

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Background: Many materials such as cosmetic products in daily use, cookware and jewellery can cause contact dermatitis. Contact dermatitis which are suffered by the seewed farmers are probably cause by many materials that are used in daily activity.

Aim: To evaluate level of CD4 and CD3 that correlated with patch test results on seaweed farmers.

Methods and Results: Cross sectional study was conducted in 41 contact dermatitis on seaweed farmers with CD4 and CD3 levels, also was conducted patch test with TROLAB patch test series. The most patients are between 19 – 28 years old (39%) consist of 10 males (24.4%) and 31 females (75.6%). The increasing of CD4 levels are found on 27 patients (65.9%). The high levels of CD4 and CD3 seem not to be related significantly with the result of patch test ($p > 0.05$).

Conclusion: The occured contact dermatitis might be caused by other material in seaweed processing or other component from the sea. Larger amount of sample is needed to get significant results and other further investigations might be contributing in the seaweed industries.

Keywords: Contact dermatitis, patch test, CD4, CD3, TROLAB

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Using of probiotics in treatment of children with atopic dermatitis

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Atopic dermatitis remains one of the serious problems of pediatrics as prevalence of disease among children is quite high (10–20 % according data of different countries) and have increasing tendency. The aim of the study was to assess the effects of probiotics on clinical course of atopic dermatitis in children. We studied 38 patients aged 1–6 year, who admitted at M. Guramishvili Pediatric clinic (2006–2007). All patients fulfilled the criteria (at least 3 major and 3 minor) for diagnosis of atopic dermatitis (Hannifin and Rajka). The children were divided into 2 groups. The first group (20 children) for 8 weeks received probiotic (Lactobacterine containing Lactobacillus acidophilus and BifidumBacterine containing Bifidumbacterin,) and emollient ointment the second group (18 children) only emollient ointment. The SCORAD index was used to evaluate severity and extent of AD at the end of the study of disease. Objective signs - spread of lesions, intensity (erythema, edema, oozing, excoriation, lichenification, and xerosis) and subjective signs (pruritus and sleeping disorders) were assessed. The mean SCORAD index before treatment was 45, 9.

After the 4 week as well as 8 week treatment was founded improvement of clinical (SCORAD index decreases) in both groups. At the same time the reduction in the SCORAD index was significant in the probiotic group. After the 8 week of study 65% of children had a better SCORAD index then baseline, comparing to the second group improvement was in 43%. So, we can conclude that supplementation with probiotics is beneficial in improvement severity of AD in children.

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Influence of peeling procedure in allergic contact dermatiitis

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Background: The prevalence of allergic contact dermatitis (ACD) in patients previously undergone peeling has been rarely studied.

Objectives: We compared the frequency of positive patch test (PT) reactions in a patient group with a history of peeling, to that of a control group with no history of peeling.

Patients and Methods: The Korean standard series and cosmetic series, was performed on a total of 262 patients. Sixty-two patients had previously undergone peeling, and 200 patients did not.

Results: The frequency of positive PT reactions on Korean standard series was significantly higher in the peeling group compared to that of the control group ($p < 0.05$, Chi-square test). However, the most commonly identified allergens were mostly cosmetic-unrelated allergens. The frequency of positive PT reactions on cosmetic series in the peeling group was higher than that of the control group, but lacked statistical significance. There was no relationship between the frequency of peels and the frequency (%) of positive PT reactions or the total number of positive PT results.

Conclusion: It appears peeling may not generally affect the development of contact sensitization. Further work is required focusing on the large scale prospective studies by performing a PT before and after peeling.

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The therapeutic efficacy & safety profile of allergenic extracts(EA) in in treating patients with labial lichen planus

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Introduction: Lichen planus an uncommon skin complaint, thought to be due to an abnormal immune reaction provoked by a viral infection (such as hepatitis C),or a drug. Inflammatory cells seem to mistake the skin cells as foreign and attack them. In 80–85% of cases it clears from skin surfaces within 18 months but when affecting the mouth may persist longer.

Methods: The classical lichen planus is characterized by shiny, flat-topped, firm papules varying from pin point size e.g.guttate to larger than a centimeter. The lesion had shinny flat-topped, firm papules varying from pinpoint to much larger sized lesions. In the study concerned 2 individuals were included. Both had been medicated with different empirical therapies. There was a remitting/ relapsing course patients were treated with a combination regimen of anti histaminic & gradual lowered dilutions of dust mite therapeutic extracts over a period of 4 months. The remission of the lesions were evident in the form of a reduction in the intensity of itchiness, ooze & redness. The final outcome was evident in the form of scar formation.

Results: In the ongoing trials the therapeutic response was dramatic in female than in the male patients.

Conclusion: Lichen planus a chronic intractable condition with multiple causes some times with a diagnostic dilemma. A detailed drugs/dietary history supplemented by immunological assessment provides diagnostic clue. *1Pre- treatment *2Skin prick test profile *3Post treatment presentation.

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Tacrolimus ointment is effective and well tolerated for the treatment of atopic dermatitis in asian countries

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Background: Tacrolimus ointment is used for the treatment of atopic dermatitis (AD) in adult and pediatric patients worldwide. However, no studies have been reported on the efficacy and safety in a large population of Asian patients with AD. The aim of this overview is to survey the assessments of the efficacy and safety conducted in the patients across 8 Asian areas, China, Indonesia, Korea, Malaysia, Philippines, Singapore, Taiwan and Thailand.

Methods: We analyzed the combined data of patients with AD from studies conducted in the Asian areas. Adult and pediatric patients with moderate to severe AD were enrolled. Adult patients applied 0.1% or 0.03% tacrolimus ointment and pediatric patients applied 0.03% twice daily for 3 to 4 weeks. The primary efficacy end point was Physician's Global Evaluation of Clinical Response (PGE). Other evaluations included Eczema Area Severity Index (EASI), Percent Body Surface Area affected (%BSA), Patient's Assessment of Itch and Patient's Assessment of Overall Response, and each category of Dermatology Life Quality Index (DLQI) and children's DLQI (CDLQI).

Results: More than 800 patients were included in these Asian studies. Success based on PGE was observed in 83.6% of all patients. The improvement in Patient's Assessment of Overall Response was similar to PGE. Percent BSA affected, EASI and Patient's Assessment of Itch improved at weeks 1, 2, and at the end of treatment. The decrease of Patient's Assessment of Itch was greatest compared with decrease of %BSA affected and EASI. All DLQI subscales (Symptoms and feelings, Daily activities, Leisure, Work and school, Personal relationships, and Treatment) improved, and also the subscales of CDLQI (Symptoms and feelings, Leisure, School or holidays, Personal relationships, Sleep and Treatment) improved at the end of treatment. In particular, Symptoms and feelings showed a marked decrease in both age groups. In subscales of CDLQI, Sleep also had marked improvement. Improvement in the PGE was associated with the decrease in %BSA, EASI and Patient's Assessment of Itch, and also with DLQI/CDLQI. Adverse events frequently reported were skin burning and pruritus at the application site. The incidence of skin burning was lower in pediatric patients than that in adults. No serious adverse events were reported from the studies.

Conclusion: Tacrolimus ointment is effective and well tolerated in the treatment of patients with AD in Asian countries.

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Downmodulatory effect of epinastine on Th2 chemokine production by epidermal Langerhans cells

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Background: Epinastine belongs to the second generation of antihistamines and also possesses anti-inflammatory or anti-allergic properties in cutaneous as well as systemic allergy, including mast cell stabilization, suppression of co-stimulatory molecule expression, inhibition of eosinophils chemotaxis and granulocytes accumulation, and suppression of cytokine and chemokine production. In Japan, systemic preparation of epinastine has been approved for not only atopic dermatitis but also pruritic psoriasis vulgaris. Skin is a well-orchestrated immune organ where epidermal Langerhans cells (LCs) function as antigen-presenting cells and keratinocytes serve as producers of various cytokines and chemokines. We have previously reported that epinastine suppresses the production of Th1 chemokines and RANTES/CCL5 by keratinocytes. Since keratinocytes and LCs are the main epidermal sources of Th1 and Th2 chemokines, respectively, the effect of epinastine on the Th2 chemokine production by LCs is an issue to be clarified.

Methods: We prepared epidermal cell suspensions from BALB/c mice and enriched them for LCs. The LC-enriched epidermal cells were cultured with epinastine for 48 h, and the expression levels of Th2 chemokines, TARC/CCL17 and MDC/CCL22, were assessed by real-time PCR analysis.

Results: The expression of CCL17 was depressed by epnastine as low as 10-6 M, and CCL22 expression was also inhibited by epinastine 10-7 M or more in a dose-dependent manner.

Conclusion: Our study suggests that epinastine exerts not only an anti-histaminic action but also a downmodulatory effect on Th2 cell migration toward the epidermis by inhibiting LC production of Th2 chemokines. Taken together with the previous finding that epinastine downregulates Th1 chemokine production by keratinocytes, we conclude that this antihistamic

drug has an inhibitory potential for migration of both Th1 and Th2 cells in the skin.

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Study of efficacy of epinastine hydrochloride in patients with pruritic dermatosis and chronic urticaria and its effects on QOL of the patients

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Epinastine hydrochloride (Alesion®, Nippon Boehringer Ingelheim Co., Ltd.) 20 mg once daily was administered to patients aged 15 years and older with pruritic dermatosis or chronic urticaria for 4 weeks, and its efficacy and effects on the patients' QOL were assessed. This study enrolled 57 patients in total, 35 patients with eczema/dermatitis, 6 patients with cutaneous pruritus, 7 patients with urticaria, and 9 patients with other pruritic dermatosis, and their mean age was 56. The endpoints were the severity of pruritus, the severity of skin eruption, visual analog scale (VAS) for pruritus, and QOL of the patients (symptoms, emotional status, daily living, and leisure, work/school, and treatment). Concerning general improvement, remarkable and moderate improvement was achieved in 43% and 33% of all patients, respectively. The severity of pruritus indicated on VAS was significantly decreased, and QOL of the patients was also improved. As a possible adverse drug reaction, an increase in AST was observed in 1 patient. These results show that oral epinastine hydrochloride once daily appears to be safe and beneficial for symptoms and QOL of patients with pruritic dermatosis or chronic urticaria.

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A randomised, double-blind, placebo-controlled study of the efficacy and tolerability of a Chinese herbal medicine concoction for atopic dermatitis

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Background: There has been considerable interest in traditional Chinese herbal medicine (TCHM) as a treatment for atopic dermatitis (AD). A twice-daily concoction of an ancestral formula containing five herbs has been found to be beneficial in an open study. The five herbs include Flos lonicerae (Jinyinhua), Herba menthae (Bohe), Cortex moutan (Danpi), Rhizoma atractylodis (Cangzhu) and Cortex phellodendri (Huangbai). We demonstrated that there was no corticosteroid (CS) or CS related compound in this formulation.

Objective: To assess the efficacy and tolerability of the concoction in children with AD.

Methods: Following a 2-week run-in period, children with longstanding moderate-to-severe AD were randomised to receive a 12-week treatment with twice daily dosing of three capsules of either TCHM or placebo. The SCORing Atopic Dermatitis (SCORAD) score; Children Dermatology Life Quality Index (CDLQI), allergic rhinitis score, requirement for topical corticosteroid and oral antihistamine were assessed before, and at week 4, 8, 12 and 16 after treatment. Adverse events, tolerability, haematological and biochemical parameters were monitored during the study.

Results: Eighty-five children with AD were recruited. Over 12 weeks, the mean SCORAD fell from 58.3 to 49.7 in the TCHM group ($n = 42$; $p = 0.003$), and 56.9 to 46.9 in the placebo group ($n = 43$; $p = 0.001$). However, there was no significant difference in the scores at the corresponding time points between the two groups. The CDLQI in TCHM treated patients were significantly improved compared with patients receiving placebo at end of the 3-month treatment and 4 weeks after stopping therapy ($p = 0.008$ and 0.059, respectively). The total amount of topical corticosteroid used was also significantly reduced by one-third in the TCHM group ($p=0.024$). No serious adverse effects were observed between the groups. Analysis of biochemical data also revealed no significant change in the IgE levels, haematological (complete blood counts, eosinophil counts) and biochemical (electrolytes, renal and liver functions) parameters monitored. No patient complained that the capsule was unpalatable.

Conclusion: The TCHM concoction is efficacious in improving quality of life and reducing topical corticosteroid usage in children with moderate-to-severe AD. The formulation was palatable, well tolerated and can probably be used as an adjunct treatment for children with refractory AD.

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Histamine (H1R) blocker inhibits histamine-induced collagen synthesis in dermal fibroblasts

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Background: Mast cell-derived histamine is known to act on dermal fibroblasts and contribute to formation of an intractable chronic allergic dermatitis. Although this fibrotic event may also occur in other organs such as nasal mucosa, no direct evidence has been reported as to whether responsiveness to histamine by fibroblasts derived from different organs is of the same intensity. Furthermore, while histamine (H1R) blocker has been proved to be effective for alleviation of the symptoms of allergic diseases, its ability to affect histamine-induced tissue remodeling has not yet been clarified.

Objective: Our aim was to study the effect of H1R-blockers on histamine-induced tissue remodeling.

Methods: A macro array assay was used for a comprehensive analysis of histamine-induced gene expression by normal human fibroblasts. Fibroblasts derived from skin or nasal mucosa were cultured in the presence of various concentrations of histamine, and the synthesis of type 1 collagen was measured by means of semi-quantitative RT-PCR and ELISA assay. To determine the effect of the H1R blocker, diphenhydramine hydrochloride and emedastine difumarate were investigated in this assay.

Results: Histamine induced various kinds of fibrogenic molecules from fibroblasts. Increased type 1 collagen expression was observed in fibroblasts treated with high-dose (-4 to -6 logM) and low-dose (-12logM) histamine. This histamine-induced type 1 collagen synthesis was effectively diminished by emedastine difumarate.

Conclusion: We found that the expression of these fibroblast-derived genes are regulated differently by different concentrations of histamine, and that the robustness of the inhibitory action of H1R blockers is different for skin-derived and nasal mucosa-derived fibroblasts. We believe that our findings may contribute to a better understanding of the mechanisms of histamine-induced tissue remodeling, and provide information useful for the management of refractory allergic dermatitis.

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Attenuating effect of fexofenadine hydrochloride on the development of cutaneous inflammatory responses through the inhibition of substance P production in mice

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Background and Purpose: Fexofenadine hydrochloride (FEX), a second-generation antihistamine, is used for the treatment of both allergic diseases and inflammatory skin reactions and successful results are reported. The primarily therapeutic mode of action of FEX is generally believed to be owing to its

suppressive effect on effector cell activation, which responsible for the development of allergic inflammation. Recently, neuropeptide such substance P (SP) attracts attention as the essential factors that trigger, exacerbate or modulate allergic skin reactions. However, the influence of FEX on neuropeptide-induced inflammatory skin reaction is not fully understood.
Methods: NC/Nga mice (8-weeks of age, female) were received repeated local application of 2,4,6-Trinitrochlorbenzene (TNCB) to provoke chronic cutaneous inflammation. FEX at a single dose of 1.0 mg/kg was orally administered into mice once a day for 2 weeks from either 1 day before or 1 week after the application of TNCB. The number of scratching for 5 min was counted 30 min after the final FEX administration. After counting the number of scratching, the SP content in skin tissues and serum IgE levels were examined by ELISA.
Results: Repeated local application of TNCB induced itching skin lesions, together with an increase in the levels of SP and IgE. Treatment of mice with FEX caused significant decrease in the levels of SP, but not IgE. Scratching behavior was also decreased significantly by the treatment of mice with FEX: the number of scratching in non-treated mice was 80 ± 13 and that in FEX-treated mice was 48 ± 9.
Conclusion: These results may suggest that FEX inhibits the scratching behavior observed in chronic cutaneous inflammation through suppression of SP production in the skin lesions.

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The efficacy and safety of levocetirizine 5mg vs. cetirizine 10mg in patients with dermatitis or eczema with pruritus: A multi-center, double-blind, double-dummy, radomized, active-controlled study

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Background: Levocetirizine is a lately developed selective H1 antagonist. The purpose of this study is to assess the efficacy and safety of levocetirizine 5 mg comparing to cetirizine 10 mg in patients suffering from dermatitis or eczema with pruritus (moderate-severe).
Method: This study was conducted in randomized, 6 centers, parallel, double-blind, and double-dummy. After screening for 3 to 7 days, subjects were randomly assigned to either levocetirizine or cetirizine treatment group and each were administered for 2 weeks with 1% topical hydrocortisone. Patients visited at screening (visit 1), randomization (visit 2), 7 days after randomization (visit 3) and 14 days (visit 4). The pruritus severity score was assessed by patients using the 4-point score scale (0; none, 1; mild, 2; moderate, 3; severe) and responders were defined as patients who reported a post treatment score rating of "None" or "Mild" at the visit 4 or at the study completion visit.
Results: 506 patients were screened and 466 patients of these were randomized. 423 patients completed the study and 340 patients were adequate for efficacy evaluation. The proportion of responders was 77.98% (131/168) in the levocetirizine treated group and 77.91% (134/172) in the cetirizine treated group. Three patients (1.44%) in the levocetirizine treated group and six patients (2.80%) in the cetirizine treated group had drug related adverse events. Unexpected adverse drug reaction was not reported in both groups.
Conclusion: Levocetirizine was non-inferior to cetirizine for the improvement of pruritus in patients with dermatitis or eczema with pruritus. Levocetirizine is safe and efficient treatment in patients with dermatitis or eczema with pruritus.

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Immunotherapy of atopic dermatitis patients with recurrent pyoderma

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Background: The aim of study was to investigate the sensitization to house dust mite (HDM) and S. aureus in atopic dermatitis patients with recurrent pyoderma (AD RP patients) and investigate the efficacy and safety of specific immunotherapy with staphylococcal and HDM allergovaccines in these patients.

Materials and Methods: The group of patients consisted of 156 AD RP patients. Intradermal tests with Staphylococcal allergen and prick tests with Der. pteronissinus and Der. farinae allergens were carried out in all AD RP patients. Skin testing was carried out in all AD RP patients in remission period. Specific IgE levels to Staphylococcal enterotoxin B as well as to Der. pteronissinus and Der. farinae in sera of AD RP patients were measured with UniCap 100 system, Phadia. 28 AD RP patients with positive skin tests and positive specific IgE to HDM and staphylococcal enterotoxin B received specific immunotherapy (SIT) with staphylococcal and HDM allergovaccines. Subcutaneous injections of allergen and vaccine were made daily, in increasing doses. The duration of the main course of specific immunotherapy was 10–14 days and then one maintenance dosage every 2 weeks; further observation period lasted 12 months.

Results: We observed sensitization to HDM in 83,9% AD RP patients, to S. aureus in 66,6% patients, both to HDM and S. aureus in 63,5% patients. There was positive effect of SIT with HDM and staphylococcal vaccine. After 1,6,12 months of the further observation period index SCORAD decreased in 26,1%, 58,3%, 44,8% accordingly. There were no exacerbations of RP in 42,8% of patients. The topical corticosteroids and antibiotics decreased significantly. Index of respiratory symptoms decreased significantly too. While treatment mild local reactions, such as redness, edema, and flare were observed in 47,6% patients during Exacerbations of AD – in 9,5% and RP – in 4,8% patients were observed. Inspite of adverse events none of the patients were excluded from the study. The levels of specific IgG4 in sera against HDM and S.aureus increased significantly. The levels of specific IgE in sera to HDM and S.aureus did not change.

Conclusion: According to our data HDM and S. aureus are the most common offending allergens in AD RP patients. Thus SIT application in these patients is needed. SIT in AD RP patients with HDM and staphylococcal vaccine was effective and safe.

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Topical application of epigallocatechin-3-gallate improves prolonged atopic dermatitis by suppressing macrophage migration inhibitory factor

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Background: There are some growing evidences that epigallocatechin-3-gallate (EGCG) exerts anti-inflammatory effects on the chronic inflammatory skin conditions such as atopic dermatitis (AD). Macrophage migration inhibitory factor (MIF) has been recently suggested to be one of the most crucial immunoregulatory cytokines of Th1/Th2 imbalance in AD. In this study we assessed the anti-inflammatory effect of EGCG by its topical application to the AD skin lesions in NC/Nga mouse model, and then we determined whether this effect of EGCG is mediated by immunoregulatory cytokines including MIF.

Methods: Induction of AD skin lesions was made by painting *Dermatophagoides pteronissinus* extract (DPE) onto the surface of each ear of NC/Nga mice. One group was then treated with EGCG solution, another group with vehicle, and the other group sham-treated for 4 weeks. To compare the clinical signs of the 3 groups, we measured ear thickness every week. At the last day, all the ears were excised and prepared for the following procedures; 1) hematoxyline and eosin stain, 2) immunohistochemistry for MIF, TNF-α and IFN-γ 3) quantitative reverse transcription polymerase chain reaction for MIF, TNF-α, IFN-γ, IL-2, IL-4 and IL-12. Blood sampling was also performed for enzyme-linked immunosorbent assay for MIF and total IgE in serum.

Results: As compared to vehicle treatment, topical application of EGCG significantly reduced the ear thickness ($p<0.05$). Likewise, the histological grading of chronic inflammation including epidermal hyperplasia and mononuclear cell infiltration into the dermis was also significantly decreased on the skin lesions of EGCG group. On the immunohistochemical staining, in comparison with vehicle treatment, EGCG treatment significantly diminished the expression of TNF-α, IFN-γ as well as MIF on the lesions ($p < 0.05$). Similarly, the mRNA expression of MIF, TNF-α, IFN-γ, IL-2 and IL-12, but not IL-4 of the lesions was significantly down-regulated by EGCG treatment ($p < 0.05$). Serum MIF and total IgE production was also significantly reduced with the EGCG. treatment ($p < 0.05$).

Conclusion: These results demonstrated that topical application of EGCG could reduce the chronic inflammation of AD lesions by suppressing a series of immunoregulatory cytokines including MIF. Taken together, it is suggested that topical application of EGCG can be a potential therapeutic modality in AD.

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Anti-IgE treatment in a patient with hyper-IgE syndrome and hepatitis c virus Infection

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Background: Hyper-IgE syndrome (HIES) is a primary immunodeficiency characterized by recurrent infections, dermatitis and elevated serum IgE. The underlying cause of HIES is unknown and the management of the patient is difficult, the general goals are to control pruritus, eczema, and to prevent infections. We report a patient with typical features of HIES and hepatitis C virus (HCV) infection, in whom anti-IgE monoclonal therapy for allergic manifestations could be an effective therapeutic alternative.

Methods: A 29-year-old woman of non-consanguineous parentage presented with moderate asthma and severe atopic dermatitis since early infancy. She had a history of recurrent respiratory infections, delay of shedding of primary teeth and slight facial dysmorphia. She had persistently elevated serum IgE levels (3000 kU/L). She suffered relapses of multiple discrete and confluent erythematous scaly papules and plaques with follicular prominence over face, trunk and limbs, with lichenification and warm hypersensitive skin, which limited her activities of daily living. In 1999, she was diagnosed of HCV infection. Later on, she developed unusual autoimmune associations:

haemolitic anaemia, alopecia universalis and autoinmune hypothyroidism. The rash had been treated with antihistamine drugs and topical steroids, with poor response. Given the high levels of serum IgE, she was given Omalizumab therapy in high dose (450 mg sc) and every 15 days schedule, monitoring IgE, IgG4 levels, hemogram and biochemical analyses.

Results: The patient improved her current symptoms of asthma since the first dose of Omalizumab and two months after the eczema severity decreased to a mild form. The patient cleared almost all her skin lesions although light lichenification areas persisted in flexures, improved xerosis, lowered itching and suffered no relapses. Serum IgE level persisted elevated and there were no change in the biochemical analyses. She did not present any side effects.

Conclusion: Therapy with anti-IgE monoclonal antibodies was found to be well-tolerated and effective. Although, current knowledge and therapeutic options in HIES are limited and with variable success, anti-IgE monoclonal therapy for allergic manifestations seems to be an effective and safe therapeutical alternative, even more as a steroid sparing agent considering her HCV infection.

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Efficacy of Omalizumab in the treatment of 3 patients with severe refractory atopic dermatitis

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Background: Atopic dermatitis is a chronic cutaneous inflammatory disease mediated by type 1 hypersensitivity reaction. High IgE serum levels are commonly found and correlated with the severity of AD. Treatment of moderate-severe cases included phototherapy, cyclosporine, azathioprine, mycophenolate mofetil and intravenous immunoglobulin with variable results. Omalizumab, humanized monoclonal antibodies bind to free serum IgE and decrease the levels of its high affinity receptor. It has been successfully used in allergy asthma, rhinitis,allergic reactions to certain foods and drugs,latex and insect bites. The experience in the treatment of AD is limited and controversial. We report the preliminary results on 3 AD patients who received anti-IgE treatment.

Methods: 3 adult patients(aged 29–44) with severe refractory AD and high IgE levels>700kU/L(5.720–12.310kU/L), were treated with omalizumab (Xolair, 450mg/kg subcutaneously every 2 weeks), 2 patients during 6 months and one patient is now on the third month of treatment. All patients had allergic rhinitis, one of them has current symptoms of asthma and other one has history of asthma. They were previously treated with topical and systemic steroids, topical calcineurin inhibitors and cyclosporine, with partial control of the symptoms. All patients were informed this was an offlabel use and the side effects profile.

Results: All the patients experienced any improvement during the treatment. After two months the eczema severity decreased in all of them, from severe to moderate-mild forms. Two patients stopped all medications either antihistaminic drugs and only one of them continues taking cyclosporine. One patient obtained partial response, with clearing of lesions, lower itching and was able to restart sport; other patient suffered autolimited relapses and did not require other therapies, and finally reach complete clearance at 6-months; the last one improves her skin lesions and asthma symptoms at 2-months. This later patient diminished the dose of cyclosporine (200 to 50 mg/day). All the biochemical analyses remained normal. None of them suffered adverse reactions.

Conclusion: Omalizumab did not systematically diminished serum IgE levels, but achieve significantly clinical improvement even on monotherapy at 6 months. On the other hand, Omalizumab showed a safe profile and might be a promising treatment in those patients in whom systemic therapy has not been successful.

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Treatment of infected severe atopic dermatitis and common variable immunodeficiency with endovenous high dose immunoglobulines (HdIVIg): a case report

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Atopic dermatitis (AD) is a chronic inflammatory skin disease whose pathophysiology is the product of complex interaction between susceptibility genes, host environments, infectious agents, defects in skin barrier and immunologic responses. Basic treatment of AD consists in topical emollients, topical and systemic CS and immunosuppressive drugs, antihistamines, phototherapy. Adjunctive HdIVIg for resistant AD may offer a useful approach, as suggested by small number of uncontrolled trials and case reports, via a number of immunomodulatory effects. Most patients with AD are colonized with S aureus and experience exacerbation after infection with this organism. In these cases treatment with antibiotics can result in reduction of skin disease. We signal the case of a 46-y-old man affected by pollens-mites-allergic rhinitis from adolescence, suffering from severe AD for 7 years with typical signs of lesional chronicity with lichenified plaques and fissuration on face and neck, upper arms and back, palm and dorsa of hands. To our attention, typical phenomena of microbial colonization were evident.

The subject confirmed high sensitization to mites, grass; total IgE sum of 979 KU/L. Negative for celiac disease and food allergy, normal common haemato-biochemical parameters. Cultural microbiology of lesions demonstrated S aureus, sensible to common antibiotics.

First line treatments resulted not efficacious. Short courses of oral antibiotics and CS associated to topical antibiotics obtained a decrease of flare for just few weeks; relapse occurred frequently justifying a not respondent disease to common treatments. Further analysis demonstrated a total gammaglobulins reduction related to a selective decrease of IgG class (325 mg/dl), normal lymphocyte fenotipe and proliferative response to common antigens, a mild deficit of granulocytes chemotaxis to IL8. Diagnosis of common variable immunodeficiency was formulated and HdIVIg instaured. After the first administration of 200 mg/kg the severe lesions of AD significantly improved with evident resolution of infections. First line treatment alone resulted efficient in controlling the disease in the following 7 months.

Subjects affected by severe not respondent to conventional therapy AD, especially with evident infections, may reasonably be investigate for possible immunodeficiency. In these cases HdIVIg treatment may be a useful support for both immunomodulatory effect and neutralization of cutaneous pathogens.

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The influence of laser therapy on the functional activity of nuetrophiles

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While carrying out NBT-tests, we have found that in 67 patients with atopic dermatitis in the original state the number of spontaneously activated neutrophiles was 17.8 + 1.3% which is considerably higher than in healthy people (6.4 + 0.9% $p<0.001$). While studying the neutrophiles activation index which reflects their reactivity to extrinsic stimulation, we have found that, in the studied group, the general drop in the index was over 2.5 times larger than compared to the norm of 3.4 + 0.7 and 8.2 + 0.6 ($p<0.001$). Along with this, in patients with atopic dermatitis we have also registered a considerable decrease in phagocytic activity of nuetrophiles, which amounted to 59.8 + 3.3, compared to the norm of 72.0 + 3.4% ($p<0.05$).

All the examined patients were divided into 2 groups, comparable by all criteria. Group I – 34 patients who were treated with laser and traditional therapy, and a control group of 33 patients, treated with the traditional drug

therapy. The positive dynamic of the studied parameters were registered with the application of different treatment methods; however, it was at its highest under the influence of laser therapy. Thus, in the main group the content of neutrophile leucocytes recovered to the normal quantity and dynamic of the above mentioned parameters was on average 48% higher than in the control group. Hence, this newly developed combined method of treating atopic dermatitis has been proven to have a desensitizing and anti-allergic effect, which is indicated by the recovery of the functional activity of neutrophiles.

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Quality of life and economic burden in Korean patients with atopic dermatitis and their parents

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Background: Because of chronic course and highly disfigured skin lesions, atopic dermatitis (AD) has a substantial impact on quality of life (QOL). Moreover, previous studies assessing the financial cost of AD have shown that it represents a notable economic burden. However, little has been known about QOL and economic burden in Korean patients with AD.

Methods: A questionnaire specifically designed by our clinical experience was used to determine how AD affects the lives of the patients and/or their parents. 66 and 39 parents completed the self-administrated questionnaires, including the questions on multidimensional aspects of QOL. They were also surveyed to find how much money they have spent in the treatment of AD during the last 6 months. All of the QOL scores are linearly transformed to 0–100 scale, with 100 indicating the worst QOL, and 0 the most favorable. In addition, for each patient the disease severity was evaluated by using SCORAD index.

Results: The QOL data revealed that AD had significantly negative effects on QOL through all aspects of life including daily living activities (37.4), psychological status (29.7) and social functioning (23.1). The average of the total QOL score was 30.1. The QOL scores significantly increase in proportion to the increasing severity of AD ($p<0.0001$). With regard to daily living activities, a major stress-causing factor was restriction on cosmetics use (44.5) and choice of foods (40.6) or clothes (36.7) as well as concern about exposure of their skin (41.2). For psychological status, 89% of patients reported that they feel deeply nervous about the likelihood of their offspring having AD in future (54.5). Regarding social functioning, it was told that AD patients had difficulties in making relationship with someone (26.4) and were less interested in the opposite sex (24.7). AD parents also had high level of stress associated with modifying lifestyles (33.8) and keeping their children from scratching (28.2). The estimated cost related to AD has the positive correlation with the severity of AD, widely ranging from 60,000 won to 1,990,000 won per patient a month. Of importance, the average direct medical cost was 77,000 won, corresponding to only 29% of the total, while the indirect cost represents 71% of the total with average cost of 189,000 won.

Conclusion: These results suggest that AD has a significant impact on the various components of QOL as well as economic burden in Korean patients and their parents.

ALLERGY & SPORTS

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Nerve growth factor (NGF) and allergic disease in top athletes

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Background: Extensive evidence has been accumulated on the role of Nerve Growth Factor (NGF) in allergic diseases. Although physical exercise has been associated, in animal models, with an increased NGF expression, no data are available about its effects on NGF in humans.

Aim: To study NGF serum levels in top athletes, a population sample in which allergic diseases and neuroimmune disorders – such as amyotrophic lateral sclerosis (ALS)–are reported with a significant increased prevalence.

Methods: 103 male soccer players (mean age 26.1) from five first division teams were studied for allergic diseases trough an original specific questionnaire (AQUAc2007), history, physical examination, skin prick tests and/or Phadiatop. Sera were taken in all athletes, at different time periods, and stored at −70°C for NGF determination trough a double-antibody ELISA. Data from a pilot sample of 36 athletes and 15 non-allergic, sedentary, matched controls are presented.

Results: The prevalence of allergy in soccer players was 47.8%. In spite of a high intra- and inter-individual variability of values, possibly due to the different training loads during the soccer season, mean NGF serum levels were significantly higher in soccer players than in controls (384.19 ng/ml +/− 298.41 ng/ml vs 128.0 ng/ml +/− 10.0 ng/ml; $p<0.05$). The NGF serum levels in athletes were increased independently from the presence of allergy.

Conclusion: Our study confirms the high prevalence of allergic diseases in athletes. The increased NGF serum levels in allergic soccer players are consistent with the role of this neurotrophin in allergic inflammation. This is the first report of increased serum levels of NGF related to intense physical activity in humans.

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Special features of allergy testing in elite sportsmen

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Sport is inevitable part of modern society. However up today the precise and convincing data on prevalence and features of current allergic diseases in elite Russian sportsmen are absent. The purpose of the present research is to study prevalence, features of allergen spectrum and structure of allergic diseases in elite sportsmen. The skin prick-tests with different groups of allergens, the estimation of total IgE end IgA, IgM, IgG in blood serum, the challenge bronchial tests with physical exercise are used in modern clinical, laboratory and allergic research methods including gathering allergic, the pharmacological and food anamnesis. Screening of 319 elite sportsmen was carried out (fencing, sport and art gymnastics, dzudo, softball, water polo, volleyball, figure skating, hockey on a grass, rowing on a canoe, freestyle, biathlon, a youth soccer team, football). The analysis of carried out research has shown the following. 23,5 % (75) sportsmen have clinical attributes of allergic disease: allergic dermatitis - 16%, allergic urticaria - 17,3%, allergic rhinitis - 30,7%, pollen-allergic rhinitis - 17,3%, bronchial asthma - 5,3%, drug allergy - 0,94%, food allergy - 0,63%. Increase of total IgE level in blood serum has been revealed in sportsmen with allergic diseases (23%) from 130 to 1000 ME/ml. Immune status analysis demonstrated the decrease of IgG and IgA level and increase of IgM level in elite sportsmen without any connection to atopy. In 94,5% of sportsmen allergy was diagnosed for the first time. Obtained data testifies widespread of allergic diseases among elite sportsmen and allergic diseases hypo-diagnostics. The hypo-diagnostics of allergic diseases results in late prescription of adequate therapy and restricts sport achievements. Changes in immune status indexes require further investigation.

MECHANISMS OF ASTHMA

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Antiproliferative and anti remodelling effect of beclomethasone dipropionate, formoterol and salbutamol alone or in combination in primary human bronchial fibroblasts

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Background: Bronchial asthma is characterized by lower airway inflammation and remodelling. Anti-inflammatory treatment with inhaled corticosteroids provides the mainstay of asthma therapy together with bronchodilation induced by short and long-acting inhaled β2-agonists. Lower airway fibroblasts may play a critical role in airway inflammation and remodelling suggesting they might represent an important target for the major antiasthmatic drugs.

The aim of our study was to investigate the effects of beclomethasone dipropionate, salbutamol and formoterol either alone or in combination on in vitro cultures of human bronchial fibroblasts.

Methods: Fibroblasts were cultured in the presence of proinflammatory and proliferative stimuli, beclomethasone dipropionate, salbutamol and formoterol. The effects of drugs on cell proliferation were ascertained by 3H-thymidine incorporation. CD90 and CD44 expression were detected by flow cytometry and fibronectin secretion using an ELISA technique.

Results: This study showed that beclomethasone dipropionate alone has significant anti-proliferative effects on lung fibroblasts treated with bFGF while LABAs or SABAs by themselves did not show any significant effect in the different cultures. The combination of BDP with formoterol or salbutamol strengthens these effects. CD44 and CD90 expression and fibronectin production were modulated by proinflammatory and proliferative stimuli; the addition of the drugs brought them back near to the basal level.

Conclusion: From this in vitro study we can conclude that beclomethasone dipropionate, when combined with salbutamol or formoterol, exhibits enhanced anti remodelling activity in bronchial fibroblasts, providing new insights on the additive effects of inhaled corticosteroids and β2-agonists for asthma therapy.

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Galectin-9 inhibits CD44-hyaluronan interaction and suppresses a mirine model of allergic asthma

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Background: Galectin-9 (Gal-9) belongs to the galectin family that exhibits affinity for β galactosides. Gal-9 has a variety of biological activities, however its role in allergic inflammation is unknown. We evaluated the effect of a stable form of the human protein on allergic airway inflammation in a mite allergen-induced asthma model.

Methods: Human stable gal-9 was given by intravenous injection to mice during antigen challenge. The effect of gal-9 on airway inflammation and airway hyper-responsiveness (AHR) was then evaluated.

Results: Gal-9 reduced AHR as well as T helper type 2 (Th2)- associated airway inflammation. Furthermore, administration of gal-9 as well as anti-CD44 monoclonal antibody inhibited the infiltration of peripheral blood Th2 cells into the airway. Interestingly, gal-9 directly bound the CD44 adhesion molecule and inhibited interactions with hyaluronan (HA). Consistent with the concept that CD44-HA interactions mediate the migration of T cells into the lung, gal-9 blocked CD44-dependent adhesion of BW5147 mouse T cells to HA.

Conclusion: We conclude that gal-9 inhibits allergic inflammation of the airway and AHR by modulating CD44 dependent leukocyte recognition of the extracellular matrix.

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Suppressive effects of a water-soluble fraction from Artemisia capillaris on the ovalbumin-induced allergic asthma model

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Background: Asthma is a chronic inflammatory disease of the airways characterized by reversible airway obstruction and hyperreactivity and remodeling of the airways. Even though a number of medications or treatments for the disease are available, the demand for a rather mild and preventive medication using the natural products such as Chinese traditional medicine is increasing, especially, in a number of Asian countries. *Artemisia carpillaris* is a perennial herb easily found around the temperate Asian regions including the entire Korean peninsula. It is called "Injin" in Korea and "Yin Chin" or "Yin Chen Hao" in China, which is traditionally used for the treatment of liver diseases and also applied to some allergic symptoms such as hives or rash. In order to identify an anti-allergic principle of the plant, we have isolated a water-soluble glycoside fraction of smaller than 1kDa and determined its biological activities on an allergic asthma model animal.

Methods: We have investigated suppressive effects of the fraction on an ovalbumin-induced allergic asthma in BALB/c mice and studied the cellular and molecular mechanisms of its anti-allergic activity.

Results: A water-soluble fraction of *Artemisia carpillaris* significantly reduced the pulmonary eosinophilia and the Th2 cytokine expressions such as IL4 and IL5 in the lungs as well as serum IgE levels. The surface expression of the CD11c and class II MHC on the lung dendritic cells was also reduced by the treatment of the fraction, indicating that the fraction also modulates the dendritic cell development in lung tissues.

Conclusion: Considering that the pulmonary dendritic cells are crucial in the differentiation of Th2 cells and the production of IL4 and IL5 in lung tissues and that the cytokines are important in the IgE antibody production and eosinophil infiltration, the fraction appears to include an anti-allergic principle modulating the Th2 differentiation and the resulting allergic asthma development.

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Protective effect of Glu27 allele of β2-adrenergic receptor gene in thai asthmatic patients

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Background: Genetic polymorphisms involving a variation at the 16th (*Arg→ Gly*) and 27th (*Gln→ Glu*) amino acid positions of the adrenergic receptor-β2 (ADRB2) possibly associated with various asthma related phenotypes including the adverse effect on lung function after regular use of albuterol in asthmatic patients who are homozygous *Arg16* and a protective effect of homozygous *Glu27* on asthma severity.

Objective: To determine association between ADRB2 polymorphisms and asthma phenotypes in Thai patients.

Methods and Materials: One hundred and thirty asthmatic patients were genotyped for *ArgGly16* and *GlnGlu27* polymorphisms. Patients' demographic, disease severity, pulmonary function test results and medication used were collected. Haplotype with unknown phase was inferred using EM algorithm method. ANOVA was applied to compare means among groups.

Results: Prevalence of *Arg16* and *Gln27* alleles were 56.9% and 91.2%, respectively. Linkage disequilibrium coefficient between the two SNPs was 0.36. Three haplotypes were inferred, which were *Arg-Gln*, *Gly-Gln*, and *Gly-Glu* with frequencies of 148 (56.9%), 89 (34.2%), and 23 (8.6%). Mean percentage of predicted FEV1 for these corresponding haplotypes were 73.5 (SD = 16.3), 72.4 (SD = 17.4), and 80.7 (13.1), respectively. Although the means were not statistical different (p = 0.258), the predicted FEV1 of *Gly-Glu* was 6%–9% higher than the rest haplotypes. Number of hospitalization and emergency visit were also lower in *GlnGlu27* than *GlnGln27* genotypes (i.e., 0% versus 11.9%, p = 0.089 for hospitalization; 4.5 % versus 18.8 %, p = 0.084 for visiting emergency). Finally, inhaled corticosteroid/long-acting

β2-agonist (ICS/LABA) usage was significantly lower in patients with *GlnGlu27* than patients with *GlnGln27* (i.e., 50% versus 76.6%; p = 0.042).
Conclusion: Presence of *Glu27* allele in Thai asthmatic patients is associated with better parameters of asthma severity, including higher percentage of predicted FEV1, less hospitalization and emergency department visit during the past year and significant lower amount of ICS/LABA usage.

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Clinical implication of vascular endothelial growth factor in children with asthma and eosinophilic bronchitis

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Background: Eosinophilic bronchitis (EB) is a condition characterized by a corticosteroids responsive cough and sputum eosinophilia, without bronchial hyperresponsiveness. Vascular endothelial growth factor (VEGF) is an important mediator of airway inflammation and remodeling in asthma. We aimed to explore whether VEGF is expressed at elevated levels in airways with asthma or EB and associated with pulmonary function and bronchial hyperresponsiveness in children.
Methods: One hundred seventeen asthmatic children, 77 children with EB and 84 healthy controls (mean age, 8.9 years) were enrolled in the study. Sputum supernatants were collected, and VEGF and eosinophil cationic protein (ECP) levels were measured. We performed pulmonary function tests and methacholine challenge tests, while measuring total eosinophil count, serum total IgE and ECP in all subjects.
Results: Asthmatic children had significantly higher levels of VEGF in induced sputum (1.82 ± 0.55 log pg/mL) compared to children with EB (1.50 ± 0.53 log pg/mL; p = .00165) or healthy controls (1.48 ± 0.48 log pg/mL; p = .00034). A positive significant correlation was found between FEV1/FVC and sputum VEGF (r = 0.257; p = .006), whereas no significant correlations were found between sputum VEGF and bronchial hyperresponsiveness, sputum eosinophil count or sputum ECP. Sputum eosinophil count showed higher levels in children with asthma or EB than in controls (p < 0.001). Sputum ECP also demonstrated higher levels in children with asthma or EB than in controls (p = .044, p = .001, respectively).
Conclusion: Our findings suggest that sputum VEGF might be a valuable marker to distinguish asthma from EB in children with chronic cough. It is also suggested that VEGF would affect the airway inflammation contributing to airway remodeling which may reflect in FEV1/FVC.

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Proportion of eosinophilic and non-eosinophilic inflammation phenotypes in patients with mild to severe bronchial asthma, its influence on main clinical and functional outcomes and efficacy of inhaled steroid treatment

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Background: Bronchial asthma is probably not a single disease, but a complex of separate syndromes. One of the methods to standardise asthma phenotypes is identification of inflammatory pattern. Inflammation subtypes may influence treatment efficacy with ICS.

Objective of our study was to determine the percentage of patients with eosinophilic and non-eosinophilic asthma phenotype, sputum inflammatory cell spectrum in each patient group, its influence on main asthma control parameters and ICS treatment efficacy.
Methods: Lung function, PD20 of Mch, induced sputum cell count, asthma symptom score and β2-agonist use was investigated in non-smoking, steroid naive and ICS treated patients with mild to severe persistent bronchial asthma. The cut-off point between patient groups was defined as sputum eosinophilia >3%. Steroid naive patients were allocated to the treatment with FP 250 μg twice daily for 12 weeks and treatment efficacy on previously mentioned parameters was evaluated.
Results: 21 steroid naive and 77 patients previously treated with ICS were included in our study. In steroid naive patient group 72% of patients were classified as asthmatics with eosinophilic phenotype and 28% with non-eosinophilic phenotype. In ICS treatment group 39% were classified as eosinophilic and 61% as non-eosinophilic asthmatics. Patient demographic data and ICS dose was similar. In patients with non-eosinophilic asthma we found significantly higher relative and absolute sputum neutrophil count, asthma symptom score and β2-agonist use (p < 0.05). There was no significant difference between FEV1 83.1 ± 18.1% vs. 87.6 ± 25.6% and 90.1 ± 20.7% vs. 96,3 ± 12.5% in steroid naive and ICS treated patients respectively; p>0.05) and lgPD20 Mch (− 1.343 ± 0.765 vs. − 1.894 ± 0.733 mg and − 0.729 ± 0.702 vs. − 1.462 ± 1.023 mg in steroid naive and ICS treated patients respectively; p>0.05).

11 patients with eosinophilic and 10 with non-eosinophilic asthma were treated with FP. We observed statistically significant better improvement of FEV1 and morning PEF in patients with initial airway eosinophilia compared to non-eosinophilic asthma (p < 0.05). Change of lgPD20 of Mch, asthma symptom score and β2-agonist use was similar in both patient groups.
Conclusion: Non-eosinophilic asthma phenotype is associated with increased sputum neutrophilia, more severe symptoms, higher rescue medication use and less effect of ICS treatment on lung function.

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Influence of anti-IgE antibody omalizumab on airway remodeling and the expression of interleukins in asthma

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Background: To study the relation between interleukin-4 (IL-4), IL-5, IL-13, transforming growth factor-beta (2) (TGF-beta (2)) and airway remodeling and to investigate the effects of omalizumab on airway inflammation and airway remodeling of asthma.
Methods: Thirty five female BALB/c mice were randomly divided into a remodeling group and a treatment group (omalizumab group), with 10 BALB/c mice in each group. The mice were sensitized by ovalbumin (OVA), and only the omalizumab group was treated with omalizumab. The number of total cells and eosinophils in bronchoalveolar lavage fluid (BALF) were counted. Light and electronic microscope were used to detect the pathologic histology and morphologic change. In situ hybridization and reverse transcription-polymerase chain reaction (RT-PCR) were used to measure IL-4, L-5, IL-13, and TGF-beta (2) mRNAs in the lung.
Results: The numbers of total cells and eosinophils in BALF of the remodeling group were (5.7 ± 1.3) x 10(5)/ml and 2.43 ± 0.18, while those of the treatment group were (4.1 ± 1.4) x 10(5)/ml and 1.67 ± 0.23, respectively, the difference being significant (P < 0.05). Histological and electronic microscopic examination showed extensive airway inflammation, notably accumulation of significant numbers of eosinophils and lymphocytes in the remodeling group. Other features including prominent proliferation of airway epithelial cells protruded like fingers, increased thickness of smooth muscle, hyperplasia of connective tissue, goblet cell hyperplasia and a marked increase in airway mucus secretion with mucus plugging and extensive collagen deposition around the airways were also noted in the remodeling group. In the treatment group, the inflammation was significantly decreased, with decreased production of mucus, decreased collagen and granule of mucus around airway, less proliferation of airway epithelium, smooth muscle hypertrophy and airway spasm. In situ hybridization showed that the expression of IL-13 mRNA and TGF-beta (2) mRNA in the lung oρf the

remodeling group were 22 ± 9 and 18 ± 3 respectively, while those of the treatment group were 16 ± 5 and 9 ± 4.
Conclusions: Omalizumab could effectively inhibit airway remodeling and decrease in the expression of IL-13 mRNA and TGF-beta (2) mRNA as well as IL-4 mRNA and IL-5 mRNA in the lung in asthma.

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IgE and IgG4 autoreactivity in bronchial asthma

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Currently, it pays much attention to the phenomenon of autoreactivity in the immunopathogenesis of allergic diseases. The chronic allergic inflammation in the bronchial tree leads to the tissue injury, which results in the modification of its structure and releasing intracellular autoantigens. It was discovered IgG - autoantibodies to DNA and bronchial endothelium cells antigens in patients with bronchial asthma (BA).
Goal: to determine the content of IgE and IgG4-autoantibodies (Ab) to some tissue antigens in patients with atopic BA different degrees of severity.
Materials and Methods: We identified IgE-autoAb and IgG4-autoAb with ELISA to the following tissue antigens: epithelial keratin, III and VI collagen types, myelin basic protein (MBP), elastin and myosin (Sigma, USA) using specific IgE and IgG4 reference sets "Dr. Fooke" (Germany). It was studied serum samples of 64 adult asthmatic patients and 25 healthy individuals the same age.
Results: The level of IgE-autoAb determined from control subjects was taken us for the average homeostatic norms and it was 1.5 ± 0.07 IU/ml. Compared to them, in patients with easy and middle BA was noted reliable elevation of IgE-autoAb to keratin (13.7 ± 4.3 IU/ml), collagen III and VI types (4.4 ± 2.1 IU/ml, 6.8 ± 3.2 IU/ml), elastin (4.9 ± 2.4 IU/ml) and myosin (18.21 ± 11.5 IU/ml). In patients with severe asthma were raised autoAb only to elastin (2.2 ± 0.7 IU/ml), collagen type III (1.7 ± 0.2 IU/ml) and myosin (7.5 ± 3.1 IU/ml). At the same time, increased total IgE correlated with IgE- autoAb to elastin and myosin (r = 0.68 and r = 0.45, respectively). IgG4 auto-Ab were also determined in the 35 serum samples of severe and middle asthmatic subjects and in the serum of 25 healthy people. Its normal average value was 2380 ± 89.5 ng/ml. The level of IgG4-autoAB to all tissue antigens in asthmatic patients was a 1.5 times higher than in healthy controls. We also revealed significant negative correlation between IgG4 and IgE-autoAB to elastin (r = −0,7), III collagen type (r = −0,56), MBP (r = −0,9). So the data indicate that IgE and IgG4-autoreactivity contributes to the development of chronic inflammation in patients with atopic BA and needs to be further investigated.

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Changes of internal diameter of large airway during and after exacerbation of bronchial asthma

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Background: Exacerbation of bronchial asthma was characterized by increased airway responsiveness induced by lower airway inflammation and resultant airway obstruction. Small airways are known to be the main area of obstruction which causes limitation of airflow, but inflammatory change during exacerbation occurs in entire airway including large airway. We evaluated the changes of internal diameter of large airway during and after exacerbation.
Methods: High resolution computed tomography (HRCT) and pulmonary function test were done as soon as possible after admission in patients with exacerbation of asthma and follow up studies were done after improvement of symptoms and discharge. With HRCT scanned images during exacerbation, internal diameters of large airway were measured consecutively from right main to right subsegmental bronchus and measurement was repeated after discharge at the same level and angle of scanned bronchus.
Results: Ten patients (male 5, mean age 43 [range 19–64]) completed studies. Pulmonary function was markedly improved after treatment of exacerbation; mean FVC from 2.93 ± 0.82 L to 3.72 ± 0.64 L (p = 0.001) and mean FEV1 from 1.55 ± 0.55 L to 2.61 ± 0.56 L (p = 0.001). Mean internal diameter on HRCT during exacerbation was 3.38 ± 0.81 mm and after treatment 3.41 ± 0.80 mm (p = 0.80). There was one patient with significant increase of internal diameter and it decreased in 2 patients. Improvement of FEV1 was not associated with changes of internal diameter on HRCT (r = 0.39, p = 0.26).
Conclusion: Before and after treatment of asthma exacerbation, there was marked improvement of pulmonary function but changes of internal diameter measured by HRCT were not consistent.

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Effect of soluble TNF-α receptor on airway inflammation in mice model of bronchial asthma

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TNF-α is an important pro-inflammatory cytokine that is also a well known inducer of the inflammatory response and a regulator of immunity. A strong argument exists for TNF-α being a critical cytokine in the pathogenesis of chronic inflammatory disorders of the airways. Once released in the airways, TNF-α acts by inducing a general inflammatory response mainly through enhanced release of pro-inflammatory/chemotactic mediators and upregulation of adhesion molecules such as E-selectin, VCAM-1, and ICAM-1, thus facilitating the migration of neutrophils and eosinophils. To date there is very limited information on the use of TNF-α blocking agents in asthma. The present study investigated the effect of soluble TNF-α receptor on the airway inflammation in mice model of bronchial asthma. Mice were treated with intraperitoneal soluble TNF-α receptor during the OVA challenge. Mice exposed to OVA developed sustained eosinophilic airway inflammation and sustained AHR to methacholine compared with control mice. Intraperitoneal administration of soluble TNF-α receptor inhibited the development of AHR and eosinophilic inflammation. Moreover, soluble TNF-α receptor treatment reduced IL-4, IL-5, IL-13 and IL-10 level in bronchoalveolar lavage fluid. These results suggest that soluble TNF-α receptor can modulate the airway inflammation and AHR via inhibition of inflammatory cytokine production.

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Detection of multi-locus genetic interaction in aspirin-intolerant asthma with multifactor-dimensionality reduction analysis

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Background and Objective: Aspirin-intolerant asthma (AIA) is a common phenotype of aspirin hypersensitivity and affects about 10~20% of asthmatic patients. Recently, the single gene polymorphism associated with the AIA susceptibility has been investigated, but identification of multi-locus single nucleotide polymorphism (SNP) set in association with the susceptibility has not been investigated.
Subjects and Methods: In this study, we selected 23 SNPs in 13 candidate genes for 94 asthmatics with aspirin hypersensitivity (AIA) and 152 asthmatics without aspirin hypersensitivity (aspirin-tolerant asthma, ATA)

and genotyped each SNP by a primer extension method. Multi-locus genetic interactions were examined with multifactor-dimensionality reduction (MDR) to test all multi-locus SNP combinations for the efficient prediction of AIA.
Results: Through a MDR analysis, we identified four-locus gene-gene interaction models that predict AIA disease risk among asthmatic patients with 64.2 % balanced accuracy.
Conclusion: These results suggest that significant epistatic effect of four-locus genetic interaction may exist in the susceptibility for AIA in asthmatic patients which may be a useful in vitro method to diagnose the AIA with acceptable sensitivity.

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IL-5 production in response to Candida Albicans secretory aspartic protease 2 is the marker of isolated late-phase bronchial responses upon inhalation challenge for nonatopic asthma

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To delineate the mechanisms of nonatopic asthma, peripheral blood mononuclear cells (PBMC) obtained from atopic asthmatics, nonatopic asthmatics, and healthy controls were incubated with various allergen molecules.

IL-2, IL-4, IL-5, IL-13, and IFN-γ productions were measured by specific ELISAs. T cell proliferation was assessed by 3H-thymidine uptake. IgE and IgG antibody were assayed by RAST and ELISA, respectively. Intradermal and bronchial inhalation challenges of the antigens were performed according to the standard procedures. Histamine releasing tests (HRT) were performed using peripheral blood leucocytes.

Proliferative response to crude *Candida albicans* (CA) extract was not statistically different among the three groups, indicating a common sensitization against CA antigen. Significant amount of lL-5 was produced by PBMC obtained from several nonatopic asthmatics upon incubation with crude CA extract and a purified antigen, secretory aspartic proteinase 2 (SAP2). IL-5 production was undetectable for the PBMC obtained from healthy control subjects in response to SAP2. Upon intradermal and bronchial challenge of SAP2, late but not immediate skin and bronchial responses were observed for the IL-5-producing asthmatics, respectively. Neither IAR nor LAR was detectable for the IL-5-nonproducing asthmatics, indicating the specificity of the responses. LAR was not induced for the IL-5-nonproducing, IL-13-producing asthmatics. IgE-dependent mechanism was ruled out by negative RAST, HRT, or immediate skin reaction. Anti-SAP2 IgG antibody (precipitin) was not detectable in the serum of either the asthmatics or the control subjects.

Nonatopic asthma may be caused by an IgE-independent, T cell-dependent immune-recognition, and in vitro cytokine synthesis become a reliable diagnostic test for "T cell allergens".

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Correlation between serum immune markers and bronchial hyperreactivity

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Background: Asthma is characterised by an underlying allergic inflammation resulting bronchial hyperreactivity. The aim of the examination was to study the correlation between serum biological markers (IL-13, RANTES, total IgE) and bronchial hyperreactivity. Symptom-free adults with childhood bronchial asthma and their symptom-free children were studied (n:76).
Methods: Bronchial hyperreactivity was proved with metacholine airway provocation in 51 symptom-free patients (30 adults, 21 children), but it was not in 25 patients (21adults, 4 children). Serum IL-13, RANTES and IgE detection was perfomed by ELISA.
Results: Significant higher IL-13 (X±SD:13,53 ± 20,24 pg/ml vs. 5,64 ± 4,24 pg/ml, $p<0.01$), RANTES (X±SD:1098 ± 543,1 pg/ml vs. 901,8 ± 322,8 pg/ml, $p<0.01$) and IgE (X ± SD:360,7 ± 352,9 KU/l vs. 130,0 ± 117,8 KU/l $p<0.0001$) concentrations were proved in symptom-free patients with bronchial hyperreactivity compared to non-hyperreactives. In symptom-free patients with bronchial hyperreactivity significant positive linear correlation was proved between the serum levels of IL-13 and RANTES, IL-13 and IgE.
Conclusion: The serum IL-13 level correlated with the IgE concentration in non-hyperreactive patients as well. In bronchial hyperreactivity IL-13 shows a correlation with the chemokine RANTES and total IgE. A persisting inflammation can be detected in bronchial hyperrectivity even in symptom-free patients.

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The local and serum level of IL-6 and its soluble receptor (srIL-6) at teenagers with bronchial asthma

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The analysis of dynamics of system and local content SR-IL-6 at teenagers with various clinical forms of allergic diseases has revealed higher levels of systemic content SR-IL-6 in comparison with its local maintenance in nasal lavage. Despite of different levels of IL-6 and SR-IL-6 the correlation of these markers was found in studied biological liquids estimation the results of the research. So, bronchial asthma (BA) SR-IL-6 level was 23,09 + 0,27 ng/ml in serum of blood and 2,24 ± 0,2 ng/ml locally.

However, IL-6 research revealed the opposite direction in the present group of patients: higher levels in nasal lavage in comparison with serum (1,97 + 0,4 pg/ml and 3,97 + 0,61 pg/ml accordingly). Combined SR-IL-6 level in serum of blood BA and the atopic dermatitis (AD) patients tended to increase in comparison with group of patients BA and made 27,24 + 0,32 ng/ml, and the SR-IL-6 level in nasal a secret was below, than at BA (1,02 + 0,04 ng/ml).

IL-6 research at BA patients in combination with AD revealed authentic (ð<0,05) increase of serum level of present cytokine (3,31±0,22 pg/ml) and decrease local one (0,97 ± 0,06 pg/ml). Content SR-IL-6 practically did not differ comparing BA and atopic rhinitis (AR) patients and BA patients making 23,64 ± 0,38 ng/ml in serum of blood and 2,03 ± 0,14 ng/ml in nasal secret. However, quantity of IL-6 in the present group was the highest both in serum of blood, and in nasal secret (9,68 + 1,11 pg/ml and 4,73 + 0,56 pg/ml accordingly) (ð<0,05). Combining clinical forms of AR and AD levels IL-6 and SR-IL-6 cytokines were the least of the studied clinical variants of atopic march, making 21,19 ± 0,11 pg/ml in serum and 0,49 ± 0,01 pg/ml locally for IL-6 and 0,18 ± 0,001 ng/ml and 0,17 ± 0,02 ng/ml for SR-IL6 accordingly.

Thus, the greatest increase of IL-6 level in serum of blood and in nasal secret occured while combination BA? and A?R with children and teenagers having all forms of atopic march. The level of soluble receptor of IL-6 with children also increased while combining BA? and A?R, whereas with teenagers - while having clinical evidences of BA? and AD more essentially.

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Zinc iron ratio in allergic asthma and allergic rhinitis

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Background: Zinc was shown to have protective effect on the respiratory system exposed to oxidative stress, soit may play a role in allergic airway diseases. Iron can have a role in oxidative stress in allergic respiratory

diseases. Iron, as well as copper, can transfer electrons and produce reactive oxygen species which can damage epithelium in allergic diseases. Both trace elements also have significant influence on immune system.

Methods: Using atomic absorption spectrophotometry, we have measured concentration of zinc and iron in serum and in supernatant of induced sputum in 21 patients with allergic asthma, in 13 patients with allergic rhinitis and in 10 control subjects.

Results: Postivie corelation was found between serum zinc and iron concentrations of patients with allergic asthma. There was no correlation between zinc and iron concentration in sputum.

Conclusion: Zinc and iron trace elements with antagonistic action on respiratory epithelium are positively correlated in serum of alergic asthmatics.

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Immunohistochemical co-localization of transient receptor potential vanilloid-1 in the trachea using a guinea-pig asthma model

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Aim: We investigated the distribution of transient receptor potential vanilloid-1 (TRPV1) in the airway of guinea-pigs.

In this study, we compared the changes in TRPV1 in the trachea using a guinea-pig asthma model.

Methods: We created an asthma model of 5 guinea-pigs by the administration of ovalbumin (OA) and 5 controls (Sham) according to a double blind study. Each trachea was removed from them and sectioned (30μm) in a cryostat. The slide-mounted sections were incubated in 10% normal donkey serum for 1 hr followed by 0.3% H2O2 for 30 minutes. In addition, they were blocked by avidin/biotin blocking kit. And the subsequently sections were incubated in polyclonal anti-TRPV1 antibody (1:30,000) for 40 hr. Next, they were incubated with biotinylated donkey anti-rabbit immunoglobulin G (1:400) for 90 minutes.

Then they were incubated in streptavidin biotin-peroxidase complex for 1 hr followed by fluorescein tyramide (1:75) for 7 min.

Finally, we observed the TRPV1 activity of each section by confocal microscopy and then compared the results.

Results: The TRPV1 immunoreactive axons were localized to the fine axons within the epithelium and around the smooth muscle area. The TRPV1 axons were found to be stronger and more frequent in the OA group than in the Sham group.

Conclusion: These findings suggest that the TRPV1 immunoreactive axons of the trachea increase in number under allergic inflammatory conditions.

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Water soluble chitosan attenuates mite allergen-induced macrophage activation in allergic asthmatic patients

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Chitin and chitosan have versatile anti-tumor, anti-fungal, and anti-microbial biological properties. Oral intake and intranasal administration of chitin attenuated allergen-induced airway inflammation in sensitized mice, which may be due to its Th1 adjuvant properties. However, the detailed mechanism of action is not clear. In this report, we demonstrated that water soluble chitosan had specific immunomodulatory effects on dust mite allergen Dermatophagoides farinae (Der f)-stimulated, monocyte-derived macrophages (MDM) in the shifting of Th2 cytokine polarization, decreasing inflammatory cytokine production of IL-6 and TNF-α, down-regulation of CD44, CD14, TLR14, and PAR2 receptor expressions, and inhibiting T-cell proliferation in the presence of allergen-stimulated MDM. Under scanning electron microscope (SEM) examination, chitosan reduced cellular change of pseudopodia formation in Der f-stimulated MDM from allergic asthma patients. The effect of chitosan on allergen-stimulated MDM may occur through inhibited PKC£a phosphorylation and NF-kB pathway activation. In a murine model of asthma, we found that intranasal application of chitosan attenuates Der f-induced lung inflammation by reducing infiltration of inflammatory cells, epithlieal damage, and goblet cell hyperplasia. The production of Arg1, iNOs and thymic stromal lymphopoietin (TSLP) in the bronchial epithelium of allergen-challenged sensitized mice was markedly decreased in chitosan-treated mice. Therefore, we believe that these results of examination of the anti-allergic effect of chitosan may provide a new therapeutic modality for allergic asthma.

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Basic FGF2 plays a key role in the allergic sensitization of airways and the pathogenesis of asthma

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Background: Asthma is characterized as a chronic inflammatory disorder of the airways associated with airway hyperresponsiveness (AHR), mucus production, and airway remodeling such as smooth muscle hypertrophy and subepithelial fibrosis. Basic FGF (FGF2) is a member of the large FGF family and plays roles in the proliferation of fibroblast, and the migration and proliferation of airway epithelial cells during wound healing. However, the role of FGF2 in the allergic sensitization is totally unknown, although FGF2 levels are thought to be elevated in patients with asthma.

Objective: To evaluate the immunological roles of FGF2 on airway allergen sensitization, and pathogenesis of asthma.

Methods: 6-week-old female BALB/C mice were sensitized intranasally with LPS-depleted ovalbumin (OVA) in the presence or absence of 10μg of rFGF2 at days 0, 1, 2, 7, and then challenged intranasally with OVA at days 14, 15, 21 and 22. AHR was measured by whole both plethysmography after 24 hours from last challenge. Mice were sacrificed at day 24, and then BAL cellularity, lung histology, and several immunologic parameters were assessed to evaluate the effects of rFGF2 co-treatment.

Results: AHR against methacholine challenge was significantly increased in mice sensitized with OVA plus rFGF2 compared with other groups. And number of total cell, macrophage, eosinophils, and lymphocytes in BAL fluids were also makedly increased in rFGF treated mice, accompanied by severe peribronchial and perivascular eosinophilic inflammations from histologic findings. Mucus secretion and subepithelial fibrosis were also increased in these mice, respectively. IL-4, IL-13, and TGF-β1 levels in BAL fluids were markedly enhanced in mice sensitized with OVA plus rFGF2.

Conclusion: These finding suggested that FGF2 plays important roles in the pathogenesis of allergic asthma, especially promoting allergic sensitization via production of inflammatory cytokines, such as IL-4, IL-13, and TGF-β1.

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The role of VEGF and its receptors on airway allergic sensitization and pathogenesis of asthma

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Background: VEGF (vascular endothelial growth factor) is known to be a key regulator of angiogenesis that is a known feature of chronic inflammatory

diseases such as asthma and chronic bronchitis. It has been reported that the levels of VEGF in tissues and biologic samples are increased in patients with asthma. VEGF has been assumed to contribute to asthmatic tissue edema through its effect on vascular permeability. The recent study using lung specific VEGF over-expressing transgenic mice showed that VEGF induced TH2 lung inflammation. However, it still remains whether the VEGF affects during allergen sensitization or adaptive immune responses by allergen specific T cells.

Objective: To assess the roles of VEGF and its receptor (Flt-1/KDR) in allergic sensitization of airways.

Methods: C57BL/6 wild type mice were sensitized intranasally with LPS depleted with ovalbumin (OVA) in the presence of 10 ¥šg of LPS on days 0, 1, 2 and 7 and then challenged intranasally with OVA on days 14, 15, 21 and 22. Before sensitization, the mice were intraperitoneally injected with SU5416 as the inhibitor of VEGFR1 and VEGFR2 at the only sensitized period. On the proper times (days 4, 23), we sacrificed mice and analyzed phenotypes including BAL cellularity, lung histology and evaluation of immunologic parameters such as the production of cytokines in BAL fluid.

Results: On day 4, the number of total cells, macrophage, lymphocytes and neutrophils increased in BAL fluid from the mice sensitized with OVA in the presence of LPS compared to only OVA treated mice. Interestingly, SU5416 blocked the recruitment of inflammatory cells in sensitization period. At this time, the production of TNF-α and IL-12 also decreased in BAL fluid from SU5416 treated mice. After OVA challenge (day 23) the number of total cells was decreased in BAL from mice treated SU5416 at sensitized period. And histological finding showed that infiltration of inflammatory cells was also significantly decreased in SU5416 treated mice.

Conclusion: VEGF induced during allergic sensitization and its signaling pathway play a key role in allergen specific T cell priming.

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Cytokine responses during exacerbation compared with stable phase in asthmatic children

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Background: Bronchial asthma is a chronic inflammatory disorder of the airways. T lymphocytes are crucial for the initiation and maintenance of the allergic inflammatory response, particularly T helper 2 (Th2) cells. Balancing in Th1 and Th2 response is targeted in treatment. Recent studies show that interleukin-10 (IL-10) have an important role in the regulation of Th2 and allergic responses and decreased in asthmatic patients.

Objective: To examine cytokine responses, including interferon-gamma (IFN- γ), IL-4, IL-10 in asthmatic children during acute exacerbation.

Methods: Fourteen asthmatic children were included in this study. Fresh whole blood obtained from patients at two phases: exacerbation and stable phase, were separated to peripheral blood mononuclear cells and were stimulated with phytohemagglutinin (PHA) and mite allergen (Der p1) for 72 hours. ELISA assays were applied to measure cytokine concentration (IFN-γ, IL-4, IL-10) of supernatant.

Results: IL-10 level (PHA-stimulated) was significantly decreased in acute asthma exacerbation (464 ± 628.25 pg/mL) compared with stable phase (859.5 ± 796 pg/mL) (p = 0.03). IL-4, IFN-γ levels and cytokine ratios for all stimulations were not significantly different among episodes. For PBMC with PHA-stimulated, IL-10 levels were slightly decreased in moderate persistent asthma compared with mild persistent asthma (737 ± 691.25 vs. 1,375 ± 870.75, p = 0.055). There were no correlations between severity of exacerbation, asthma score, size of skin prick test and cytokine levels.

Conclusion: The decrease of IL-10 production in asthmatic children during acute exacerbation may have a role in asthma exacerbation. The decrease of IL-10 production in more severe asthma (moderate persistent) supports the idea of defective immune regulation of IL-10. Further studies in IL-10 producing cells and treatment to enhance IL-10 responses might be useful in prevention and treatment of asthma.

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Alternative transcripts of cysteinyl leukotriene 1 receptor ($CysLTR_1$) in patients with bronchial asthma

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Introduction: Cysteinyl leukotrienes are lipid mediators that have been implicated in pathogenesis of several inflammatory processes, including asthma. They cause bronchoconstriction, mucous hypersecretion, increased microvascular permability, bronchial hyperresponsiveness, and eosinophil infiltration. The biological action of cysLTs is mediated via $CysLT_1$ and $CysLT_2$ receptors. Human *$CysLTR_1$* gene consists of five exons variably spliced. Transcript I, composed of 1, 4 and 5 exons is a major transcript present in human leukocytes, smooth muscles. Transcript II, composed of 1 and 5 exons is less abundant but find in blood leukocytes, smooth muscles, hart, brain. The role of alternative transcripts in asthma pathogenesis is still not known.

Aim: The goal of our study was to investigate differences in expression of $CysLTR_1$ alternative transcripts I and II in patient with bronchial asthma and healthy control group.

Methods: PBMCs were obtained from peripheral blood of 20 patients with severe and non-severe asthma and 15 healthy volunteers. Total RNA was obtained from mononuclear cell using TotalRNA and cDNA was synthesized using Reverse Transcriptase. We designed specific primers spanning exon-exon junction in the transcript I (exons 1–5) and transcript II (exon 1–4) and common primer in the $CysLTR_1$ coding region according to published $CysLTR_1$transcripts sequences. Real-time PCR reactions were performed on RotorGene using Evagreen dye.

Results: We found that $CysLTR_1$ transcripts I and II are present in patients with asthma and in control group. The $CysLTR_1$ transcripts II/I ratio was 26,02 % in patient with severe asthma, 25,99 % with moderate asthma, 56,44 % with mild asthma and 44,20 % in control group. In addition, we found significant difference in $CysLTR_1$ transcripts II/I ratio in severe asthmatic subgroup in comparison to controls [p = 0,004], severe asthmatic in comparison to mild [p = 0,018] and in $CysLTR_1$transcripts II/I ratio in moderate asthmatic in comparison to control group [p = 0,0004] and in mild asthmatic in comparison to control group [p = 0,007].

Conclusion: Differences in $CysLTR_1$ alternative transcripts expression might possibly contribute to airway inflammation in patient with bronchial asthma.

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Prostaglandin production in different asthma patients

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Background: Special regulatory role of prostaglandin E2 has been postulated in aspirin-induced asthma. The aim if this study was to investigate the effects of aspirin on systemic production of prostaglandin E2 and cysteinyl leukotrienes in patients with asthma.

Methods: We determined urinary concentrations of two main prostaglandin E2 metabolites: 13,14-dihydro-15keto-PGE2 using commercial enzyme immunoassay and 9,15-dioxo-11alpha-hydroxy-2,3,4,5-tetranor-prostane-1,20-dioic acid using gas chromatography/mass spectrometry; and leukotriene E4 using immunoassay. Determinations were performed at baseline and following oral aspirin and celecoxib challenges, in two well-defined asthma phenotypes: aspirin-sensitive and aspirin-tolerant patients.

Results: Aspirin precipitated bronchial reactions in all aspirin-sensitive, but in none of the aspirin-tolerant patients. Celecoxib 400 mg was well tolerated by all patients except for one with aspirin-induced asthma. At baseline mean prostaglandin E2 metabolites values did not differ between the groups. Following different aspirin provocation doses, the two main prostaglandin E2 metabolites were decreased in the aspirin-tolerant group, but their mean level remained unchanged in the aspirin-sensitive group. The dose of aspirin had no effect on the magnitude of the response on the prostaglandin E2 metabolites and its duration. In both groups urinary prostaglandin E2 metabolites decreased following celecoxib challenge. No correlation was found between prostaglandin E2 metabolites and leukotriene E4.

Conclusion: Aspirin-precipitated asthmatic attacks are not associated with changes in the systemic prostaglandin E2 production. In contrast, prostaglandin E2 systemic production becomes depressed by aspirin in non-sensitive patients. This different response might indicate COX-1 dependent prostaglandin E2 control of inflammatory cells in AIA. Thus, PGE2 is released during the clinical reactions to aspirin through an alternate COX-2 pathway. Clinical implications of this finding are in line with current observations of good tolerance of the selective COX-2 inhibitors in sensitive patients.

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Mast-cell activation in aspirin-induced asthma

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Background: There is increasing evidence of the importance of cysteinyl leukotrienes (LT) as mediators of aspirin-induced bronchoconstriction in aspirin-sensitive asthma but the cellular origin of the LT is not yet clear.

Methods: Urinary concentrations of leukotriene E4 (LTE4), 11-dehydro-thromboxane B2, 9alpha,11beta-prostaglandin F2, and Ntau-methylhistamine were measured during the 24 h following cumulative intravenous administration of increasing doses of lysine aspirin to asthmatic patients. In addition, the urinary concentrations of these metabolites were measured on 10 consecutive days in a patient who suffered an asthma attack after percutaneous administration of nonsteroidal anti-inflammatory drugs.

Results: In aspirin-induced asthma patients (AIA, n = 18), the basal concentration of urinary LTE4, but not the other metabolites, was significantly higher than that in aspirin-tolerant asthma patients (ATA, n = 16). After intravenous aspirin provocation, the AIA group showed a 12.6-fold (geometric mean) increase in excretion of LTE4 during the first 6 h, and 9alpha,11beta-prostaglandin F2 also increased in the AIA group during the first 0–6 h and the 6–9 h collection period. Ntau-methylhistamine excretion was also increased, but to a lesser degree. Administration of aspirin caused significant suppression of 11-dehydrothromboxane B2 excretion in both the AIA and ATA groups. When the percentage of maximum increase of each metabolite from the baseline concentrations was compared between the AIA group and the ATA group, a significantly higher increase in excretion of LTE4, 9alpha,11beta-prostaglandin F2, and Ntau-methylhistamine was observed in the AIA group than the ATA group. An increased excretion of LTE4 and 9alpha, 11beta-prostaglandin F2 has been detected in a patient who suffered an asthma attack after percutaneous administration of non-steroidal anti-inflammatory drugs.

Conclusion: Considering that human lung mast cells are capable of producing LTC4, prostaglandin D2, and histamine, the results of our study support the concept that mast cells, at least, may participate in the development of aspirin-induced asthma.

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Markers of inflammation in asthmatic patients

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Background: Bronchial asthma is characterized by airway inflammation, hiperresponsiveness to variety of stimuli and airway obstruction, which can be reversible. Different cells are involved in the pathogenesis of asthma, like: T-lymphocytes, mast cells, eosinophils, macrophages etc. A number of different inflammatory cells infiltrate the airways. The trigger can cause the release of inflammatory mediators from the cells, which could be the markers of inflammation.

Methods: We have investigated the role and the importance of inflammatory cells and mediators like eosinophils, ECP, IL-4 and IL-5. We studied 77 subjects divided in three groups as follows: 1. asthma patients; 2. patients with obstructive bronchitis and 3. control group.

Results: The number of eosinophils was significantly increased in the group of asthma patients versus second and third group. The eosinophils have their very important role in allergic inflammation. We found that the presence of ECP demonstrate an ongoing inflammation, with or without clinical symptoms in asthma patients. There was significant difference between the values of ECP of asthma patients versus second group with bronchitis and healthy controls. The values of IL-4 were not significantly increased between the groups. We found the explanation in the fact that IL-4 is a marker for early allergic inflammation. Our results have shown that IL-5 is involved in the pathophysiology of asthma. The values of IL-5 were significantly increased versus second group and controls. We also found the decrease of the values of inflammatory markers after the treatment with corticosteroids.

Conclusion: We concluded that eosinophils, ECP and IL-5 could be useful markers for selecting allergic patients and they could be used together with other examinations. The markers of inflammation could have their important role in the measurement of the allergic inflammation and they could be the monitors of treatment effects.

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Mucosal immunotherapy with CpG oligodeoxynucleotides increases local IL-10 concentration in a murine model of chronic asthma

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We examined CpG motif effects on a previously developed murine model of asthma in which chronic airway inflammation was induced by repeated allergen [Chenopodium album (Ch.a)] inhalation. Using this model, we examined the responses to mucosal administration of CpG DNA (oligonucleotides) and specific antigen immunotherapy. CpG-based immunotherapy significantly reversed both acute and chronic markers of inflammation compared with specific Ag immunotherapy. IL-10 levels were also measured both in splenocyte and lung culture supernatants. The results showed that antigen recall responses of lung culture from mucosal treated mice demonstrated an antigen-specific enhanced release of IL-10, but the concentration of this regulatory cytokine had no significant changes in splenocyte culture medium. These results suggest that mucosal immunotherapy with CpG DNA may induce local production of cytokines without systemic effects on Th responses. We suggest that mucosal co-administration of CPG /Ag may provide the basis for a more efficient form of immunotherapy in allergic asthma. Indeed local induction of cytokines by this procedure may diminish potential toxicity of systemic antigen administration.

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Assessment of airway remodeling by HRCT in asthmatics: correlation with age, smoking, disease duration and severity

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Background: To date, airway remodeling is usually assessed using histological examination of airways. However, now it is possible to assess and quantify the extent of airway remodeling in vivo using high-resolution CT (HRCT). The aim of this study was to prospectively evaluate airway wall thickness as indicator of remodeling by using thin section HRCT in asthmatics, and to correlate these findings with pulmonary function tests (PFTs) results and other clinical indices in asthmatics.

Methods: 41 patients with asthma and 20 healthy controls participated in the study. Remodeling as measured by whole airway wall thickness was assessed with HRCT. Thickness-to diameter ratio (TDR) and the percentage wall area (PWA) were determined. Spirometric tests were also performed.

Results: TDR and PWA were significantly higher in asthmatics than in controls. Both TDR and PWA were strongly correlated with disease severity and duration. Also, TDR and PWA were inversely correlated with the percentage of predicted forced vital capacity (FVC %), forced expiratory volume in 1st second (FEV1%) and FEV1/FVC and post- bronchodilator reversibility in asthmatics.

Conclusion: These findings indicate that HRCT is useful non- invasive method for assessment of airway wall thickness. Airway wall thickening occurs more in patients with moderate and severe asthma and its degree is related to the duration and severity of asthma and degree of airflow obstruction.

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Clinical and immunologic findings of methylene diphenyl diisocyanate -induced occupational asthma in a single car upholstery

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Background: Although methylene diphenyl diisocyanate (MDI) has been widely used in many industries, there have been few studies of MDI- induced occupational asthma.

Objectives: We investigated to present the clinical and immunologic findings in methylene diphenyl diisocyanate-exposed workers in a single industry of car upholstery.

Methods: Fifty-eight exposed workers in a single industry were enrolled. Work related respiratory symptoms were screened using a respiratory questionnaire. Serum specific IgE and IgG antibodies to MDI-human serum albumin conjugate were measured by ELISA. Atopy status was evaluated using allergy skin prick test. For confirmation of MDI-induced occupational asthma, methacholine bronchial challenge test and MDI-specific inhalation test with changes of sputum eosinophil counts were performed in symptomatic workers.

Results: Thirteen (22.4%) subjects had complained of respiratory work-related symptoms. The prevalence of MDI-occupational asthma was noted in 5 (8.6%) workers and occupational eosinophilic bronchitis was noted in 2 (3.45%) workers. The prevalence of specific IgG (20.7%) was higher than that of specific IgE (8.6%). The prevalence of MDI-occupational asthma/ eosinophilic bronchitis was strongly associated with the presences of work-related respiratory symptoms and MDI-specific IgG antibodies ($P < 0.01$, $P < 0.05$, respectively).

Conclusion: MDI may be a causative agent of occupational asthma among MDI-exposed workers. The prevalence of MDI-occupational asthma was 8.6%, and MDI-eosinophilic bronchitis was confirmed in 2 workers. The presence of work-related lower respiratory symptoms and serum specific IgG to MDI-human serum albumin conjugate may be useful to predict MDI-occupational asthma/eosinophilic bronchitis in MDI exposed workers.

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Risk factors for spider mite (Tetranychus urticae) allergy among table grape farm workers in South Africa

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Background: Various studies among workers in vineyards and orchards have reported an increased prevalence of respiratory symptoms including asthma. The aim of this study was to identify risk factors associated with spider mite allergy among table grape farm workers.

Methods: A cross-sectional study of 207 workers in table grape farms was conducted. Skin prick tests (SPT) used extracts of 8 common aeroallergens (ALK), grape mould (Botrytis cinerea) and Tetranychus urticae (TU). Specific IgE to TU was quantified using ImmunoCAP (Phadia). Erythrocyte cholinesterase (AChE) levels were determined using the Test-mate ChE Cholinesterase Test System (Model 460).

Results: The average duration of employment of workers was 10 years and 12% were pesticide crop sprayers. Work-related wheeze (26%), ocular-nasal (24%), urticaria/skin symptoms (14%) were more prevalent in the orchards. The prevalence of sensitization (SPT) was the highest to TU (22%) followed by house dust mite (16%), with 25% being atopic. The prevalence of allergy to TU (skin reactivity and work-related symptoms) was 9.5%, with respiratory (6%) more common than skin allergy (3%). Work-related ocular-nasal (OR = 4.9) and skin (OR = 3.7) symptoms were more commonly reported by

pesticide crop sprayers. Workers with TU-allergic rhino-conjunctivitis and probable asthma were more likely to be atopic, spray pesticides and have low (<30 U/g Hgb) AChE levels.

Conclusion: This study demonstrates that spider mite, T. urticae, is an important outdoor allergen among table grape farm workers. The increased risk of spider mite allergy appears to be related to high pesticide exposure among crop sprayers.

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A novel murine model of toluene diisocyanate-induced asthma

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Background: Toluene diisocyanate (TDI), a highly reactive industrial chemical, is one of the leading causes of occupation-related asthma in industrialized countries. The pathogenesis of TDI-induced asthma remains not fully understood, in part due to lack of appropriate animal models. In our study, we established a novel TDI asthma mouse model by epicutaneous sensitization and intranasal challenge.

Methods: Thirty two 6–8 week BALB/c mice were randomly divided into four groups (n = 8): TDI, acetone olive oil (AOO), ovalbumin (OVA) and saline (sal). On days 0, 7 and 14, the mice were epicutaneously sensitized with 1% TDI in 100 μl acetone: olive oil (3:2, AOO), 100 μl AOO, 100 μl 0.1% OVA in sal and 100 μl sal respectively, every allergen or solvent was placed on a patch of sterile gauze (1×1 cm) which was taped to the skin of shaved mouse back and kept for 3 days per sensitization. On days 21, 23 and 25, mice were challenged intranasally with 25 μl identical allergen or solvent respectively once a day. All mice were sacrificed on day 27. 24 hours before first intranasal challenge and after last intranasal challenge, airway responsiveness to methacholine (Mch) was measured in an unrestrained whole body plethysmogragh and was expressed as enhanced pause (Penh). Mice were exposed to nebulized phosphate-buffered saline as the baseline and then increasing dose of Mch, namely 5%, 10%, 20% and 40% followed by measurement of Penh values for 5 minutes. TDI- and OVA-specific IgG1, IgG2a and IgE in sera were measured by ELISA and expressed as optical density (OD) mean value's.

Results: 1. Histology: TDI-exposed mice exhibited neutrophil-dominant pulmonary inflammation in the peri-bronchial and peri-vascular regions by lung tissue haematoxylin and eosin (HE) staining, and increased mucus secretion by PAS staining, while OVA-treated mice exhibited an eosinophil-dominant inflammation and mucus production. 2. After challenge in TDI-treated mice airway responsiveness was significantly augmented, but not in OVA- and solvent-treated mice. 3. Significant increased production of sera TDI-specific IgG1, IgG2a and IgE in TDI-treated mice was observed as well as the increased OVA-specific IgG1, IgG2a and IgE in OVA mice.

Conclusion: Our study showed that allergic asthmatic responses to a chemical sensitizer such as TDI may occur after dermal sensitization and airway challenge via IgE-mediated mechanism.

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A case of occupational allergy in a platic industry

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We report a case of a patient male of 33 years old with recurrent symptoms of rhinitis, conjunctivitis, and dermatitis related with his job.

He hasn't had family nor personal antecedents of atopy. His symptoms were recurrent pruriginous eczema located to body and legs, accompanied of rhinitis and conjunctivitis, without symptoms of bronchial reactivity. These symptoms appeared while he was in the place of work and improved at the weekend and holidays. He was working for eighteen months in a pipe's factory as support agent. They used polyvinyl for made the pipes and was in contact with resins, cobalt, styrene and glass fibre in this factory.

We preformed a skin prick test with a battery of common aeroallergens and a standard patch test (True test®) with negative result. The basal espirometry performed had a normal pattern and the bronchodilatation test was negative. The blood analysis showed a normal biochemical levels and normal haematological values, with antinuclears and antitissue antibodies negatives. The specific IgE determination against anhydride phthalic was 25,7 KU/l and against formaldehyde was <0.35kU/l.

Our diagnosis' suspicion was sensitization to anhydride phthalic as the cause of the symptoms that he presented.

The patient's evolution after the job's change was very favourable. He has remained asymptomatic until nowadays and the specific IgE levels to anhydride phthalic has gone lowering, being of 0.42 KU/l eighteen moths later. The use of anhydride phthalic as hardener of epoxy resins forms polymers that are useful to make materials as glass fibre. Sometimes there are too many substances in the occupational environment then it can difficult the causal diagnosis.

The acid anhydrides are substances with a low molecular weight that can cause rhinitis, asthma and contact dermatitis associated to an IgE mediated mechanism. The airborne exposition to anhydride phthalic can happen more frequently in the plastic industry.

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Predictors of work-related symptoms, allergic sensitisation and occupational asthma among supermarket bakery workers in South Africa

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	TDI-specific-IgG1	TDI-specific-IgG2a	TDI-specific-IgE	OVA-specific-IgG1	OVA-specific-IgG2a	OVA-specific-IgE
TDI	3.01 ± 0.09*	1.96 ± 0.80*	0.93 ± 0.34*	0.20 ± 0.05	0.15 ± 0.11	0.10 ± 0.05
AOO	0.24 ± 0.16	0.15 ± 0.05	0.11 ± 0.01	0.11 ± 0.04	0.11 ± 0.02	0.08 ± 0.01
OVA	0.31 ± 0.11	0.20 ± 0.12	0.15 ± 0.08	2.97 ± 0.11*	0.48 ± 0.36#	0.20 ± 0.12#
sal	0.23 ± 0.07	0.21 ± 0.08	0.11 ± 0.02	0.17 ± 0.15	0.13 ± 0.04	0.09 ± 0.03

Compared to solvent control, P < 001; compared to solvent control, P < 0.05.

Background: A recent study reported a high risk of developing work-related asthma among supermarket bakers. This study aimed to determine the predictors for work-related symptoms, allergic sensitisation, non-specific bronchial hyper-responsiveness (NSBH) and baker's asthma in small bakeries of a supermarket chain store in South Africa.
Methods: A cross-sectional study of 517 (current and previously employed) bakers was conducted in 31 Cape Town bakeries using a modified European Community Respiratory Health Survey (ECRHS) questionnaire, skin prick tests to common aeroallergens and cereal flours, as well as serum specific IgE to wheat flour, rye flour and fungal alpha amylase by ImmunoCAP-system (Phadia, Sweden). NSBH was assessed using the Medic Aid Pro Nebulizer Dosimeter method. Exposure-response modeling was conducted to identify significant determinants.
Results: The mean age of bakers was 32 years and 47% were current smokers. The prevalence of atopy (positive SPT to ≥1 common aeroallergen) was 42%. Common work-related symptoms were ocular-nasal (31%) and chest tightness/ wheezing (17%). One third of bakers were sensitised to bakery allergens with 25% sensitised to cereal flours such as wheat and rye. There were 22% of the workers who demonstrated evidence of bronchial responsiveness with 2/3 of these having airway obstruction. Doubling the employment duration was associated with an increased odds for specific IgE reactivity to wheat (OR: 1.28, CI: 1.01 – C 1.62), rye (OR: 1.37, CI: 1.07 – C 1.75), and allergic ocular-nasal symptoms due to wheat (OR: 1.32, CI: 1.0 – C 1.83). Workers in the bakery sensitized to wheat flour were almost four times more likely (OR: 3.75, CI: 1.79 – C 7.84) to be bakers and six times more likely to be supervisors/ managers (OR: 6.03, CI: 2.37 – C 15.38), the latter group having generally lower dust exposures. Stratification of the jobs per employment duration maintained the inverted U-shaped exposure response curve observed.
Conclusion: Job title and, to a lesser extent, employment duration are important predictors of sensitisation among supermarket bakery workers. The non-linear exposure response relationship observed in this study needs further exploration.

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Rhinitis, conjunctivitis and asthma due to deer antler

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Background: Occupational allergy in cutlers is a very strange phenomena even though they handle a lot of potencially allergenic substances.
Case Report: A 40-year-old man, cutler since he was 14, active smoker of 20 cigarettes/day. He was referred to our deparment because he presented during the last 4 months daily dysnea, wheeze, cough and thoracic oppression accompanied by rhinitis and conjunctivitis of diurnal predominance and clearly related to his work place. He had been treated with formoterol 9 mcg/ budesonida 320 mcg every 12 hours since 45 days before his visit. He got worse throughout the week and specially on Fridays, when he swept the floor of the factory, forcing him this fact to go to the emergency deparment in 3 occasions. He improved during the weekend and referred to work with metals, wood of olive tree, oak ("with resin") and an artificial wood, although other workers next to him handled deer antler (since 4 years before) and bull horn. In his previous work in a cutlery (16 years before), he used to work with deer antler and wood; during the last three months of working there he presented daily cough, dysnea and wheeze which were related to wood dust. Once he changed the company to the present one, he remained asymptomatic during 15 years until now.
Results: Skin prick test (SPT) with a standard battery of inhaled and epithelial allergens and a battery of woods and mites: all of them were negative. SPT with an extract of deer antler gave positive results (4×4 mm) with 10 negative controls (5 were atopic patients). SPT with bull horn: negative. Respiratory functional tests after remaining 48 hours without treatment: Forced spirometry: mild obstructive pattern (FVC: 82%, FEV1: 75%) with positive bronchodilator test. Total IgE: 75 UI/ml. Specific IgE was measured by EAST method (Enzyme AllergoSorbent Test) obtaining the following results: deer antler, 0.7 kU/l (class 1); fur and dander from cow, 1.6 kU/L (class 2); epithelium from dog, cat and horse, all of them less than 0.35 kU/L (class 0). The serum specific IgE levels to serum albumins from bovine, cat and chicken (alpha livetin) was also measured, and all of them were below 0.35 kU/L (class 0).

The molecular mass of the IgE binding proteins was determined by SDS-PAGE inmunoblotting: two bands of 63 and 52 kDa were detected in deer antler extract.
Conclusion: We report the first case in the literature of allergic rhinitis, conjunctivitis and asthma caused by deer antler.

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Acute allergic reaction and chronic pulmonary effects of occupational inhalation exposure to talc dust

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Objectives: The main purpose of this study was to assess and characterize the pulmonary reactions associated with occupational exposure to talc dust.
Methods: Ninety-seven talc workers and 110 unexposed employees as the reference group were randomly selected from a local rubber industry. Standardized respiratory questionnaires were administered to the subjects, they underwent chest X-ray and were examined by a specialist for any possible respiratory abnormality to be diagnosed. Furthermore, Pulmonary Function Tests (PFTs) were measured just before and after the work shift. Moreover, to assess the extent to which workers had been exposed to talc dust, using standard methods, inhalable and respirable dust concentrations were measured in different dusty worksites.
Results: The average (mean ± SD) age (years), weight (kg), height (cm) and duration of exposure to talc dust (years) for the exposed group were 35.8 ± 6.75, 73.1 ± 9.2, 172.3 ± 5.9 and 11.79 ± 5.3, respectively. The corresponding values for the non-exposed group were 36.1 ± 6.87, 73.36 ± 8.1, 173.2 ± 5.7 and 0 ± 0, respectively. Atmospheric concentrations of inhalable and respirable talc dust were estimated to be 41.8 ± 23.52 and 19.8 ± 8.04 mg/m3, (mean ± SD), respectively. Talc exposed subjects had a significantly higher prevalence of respiratory symptoms. Similarly, PFTs revealed that exposure to this lubricating agent was associated with significant decreases in the mean percentage predicted of vital capacity (VC), forced vital capacity (FVC) and forced expiratory volume in the first second (FEV1). Likewise, there was a general tendency for VC, FVC and FEV1 to decrease as estimated cumulative exposure (years worked) increased. Moreover, there was an acute reduction in some parameters of pulmonary function such as VC, FVC and FEV1, over the work shift. Chest radiographs of exposed workers showed that pneumoconiosis profusions were between p 0/0 and p 2/1 according to the ILO 1980 chest X-ray classification.
Conclusion: These results support the notion that occupational inhalation exposure to talc is associated with chronic respiratory disorders and induces bronchitis and interstitial lung disease. Additionally, they support the hypothesis that inhalation exposure to talc dust induces acute aalergic reversible adverse effect on PFTs.

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Change of lung function and airway hyperresponsiveness in occupational asthma

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Background: Improvement of respiratory symptoms and airway hyperresponsiveness (AHR) of occupational asthma (OA) may be prolonged after cessation of

exposure to causative agents. Significant improvement or disappearance of symptoms or AHR at follow up period is frequently used for the parameter of OA outcome measurement. However, few studies have longitudinally evaluated the change of lung function and AHR by serial measurement. To evaluate the significance of the lung function or AHR as outcome parameter, we serially measured it and compared its change with the clinical condition of OA.

Methods: Pulmonary function test, methacholine bronchial provocation testing, asthma symptom and medication scores were evaluated at regular interval in 35 patients with OA confirmed by specific challenge tests.

Results: AHR continuously resolved during the follow-up period (mean: 7.5 years, range: 1–16 years) in 5 (14.3%) of 35 patients. AHR transiently disappeared, however, subsequently re-developed in an additional 5 patients. The level of AHR improved (increase in methacholine PC20 by 3.2-fold or more) in 9 (25.7%) patients, but fluctuated in remaining 16 (45.7%) patients. The level of FEV1 was fluctuated along with the severity of asthma symptom, but not in proportion to the level of AHR. In 20 of 24 patients with reduced lung function at initial examination, lung function did not recover during the follow up period. Significant improvement of symptoms and maintenance of nearly asymptomatic state were achieved in 5 patients. Three of them had disappearance of AHR, but 2 patients showed fluctuated levels of AHR. Lung function and severity of asthma at diagnosis, and the interval between the development of OA symptoms and avoidance was associated with functional impairment.

Conclusion: In spite of medical treatment and avoidance, many patients with OA had persistent and fluctuated AHR and lung function impairment even in the absence of respiratory symptom. Intermittent or single assessment of lung function or AHR as outcome measure might lead to incorrect clinical evaluation, and more comprehensive approach will be required.

RHINITIS AND RHINOSINUSITIS

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ARIA suggested drugs for allergic rhinitis and Quality of Life. A GA2LEN review

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Background: Allergic diseases constitute a global health problem, as they have an increasing economic and social impact and, especially, they can deeply interfere with patients' daily life, being a cause of physical and emotional discomfort. This is why health related quality of life (HRQoL) has become increasingly important in healthcare research.

Aim: To examine the scientific literature of the last three years dealing with the impact of allergic rhinitis (AR) treatments suggested by ARIA guidelines on patients' HRQoL, and to identify the unexplored or not fully investigated areas concerning this issue.

Methods: Studies were sought from MEDLINE (1 January 2004 to 31 December 2006) using as key words the ARIA suggested drugs [AND] allergic rhinitis [AND] and quality of life. Only randomized, DBC trials published in peer-reviewed journals, using validated questionnaires were selected.

Results: Our research has lead to a selection of 34 DBPC trials, 4 in paediatric and 30 in adult population. HRQoL has been assessed in a total of 10957 patients. The RQLQ questionnaire was used in 27 studies. The Rhinasthma was applied in one study involving patients with both rhinitis and asthma. SF-36 was used together with the specific questionnaire in 3 trials. Only a paper on sublingual immunotherapy (SLIT) evaluated health status by means of SF-36 and subjective satisfaction by means of Satisfaction Profile (SAT-P).

The duration of the studies varied from 2 days to 3 years. 19 studies were about SAR, 3 about PAR, 1 about seasonal and perennial rhinitis. 6 studies investigated rhinitis according to ARIA classification: 3 of them were about persistent allergic rhinitis, and 3 considered persistent allergic rhinitis and its comorbidity with asthma. 3 studies investigated rhinitis without providing any classification of the disease. Out of 34 trials, only 8 evaluated the possible presence of comorbid asthma.

Discussion: On the basis of our research, it is possible to identify some unexplored areas in HRQoL of patients affected by allergic rhinitis: the new ARIA classification (intermittent and persistent rhinitis) should be carried out for all the treatments in order to fully appreciate the effect of drugs used in AR on aldult and children patients' HRQoL; the effect of comorbidity on HRQoL should be extensively investigated; HRQoL study should include some other aspects of patient's perspective life (i.e. satisfaction, adherence).

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Disturbances in nocturnal sleep, excessive daytime sleepiness and sleep-specific quality of life impairment in patients with allergic rhinitis

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Background: Nocturnal sleep disturbances, excessive daytime sleepiness and impairment in quality of life (QOL) in allergic rhinitis are often overlooked both by patients and physicians alike.

Methods: Consecutive skin allergy test positive patients with allergic rhinitis and matched controls were subjected to computed tomography of paranasal sinuses (CT-PNS) Patients were categorized as allergic rhinitis (group 1), allergic rhinitis with sinusitis (group 2) and controls (group 3). They were evaluated for nocturnal sleep, excessive daytime sleepiness and sleep specific QOL disturbances using the Pittsburgh Sleep Quality Index (PSQI), Epsworth Sleepiness Score (ESS) and Nocturnal Rhinoconjunctivitis Quality of Life Questionnaires (NRQLQ) respectively. They were also categorized as "sneezer-runners" or "blockers".

Results: Of the 207 patients (141 males, 66 females), 75(36%) were in group 1, while 132(64%), in group 2 and 23 in group 3. Patients in group 2 had significantly higher mean scores for PSQI, ESS and NRQLQ questionnaires as compared to group 1.

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Mean score	allergic rhinitis with sinusitis (group 2) N = 132 (64%)	allergic rhinitis only (group 1) N = 75 (36%)	P value
PSQI	9.8 + 4.1	8.6 + 4.4	0.02
ESS	11.4 + 5.8	7.4 + 3.2	0.001
NRQLQ	47 + 8.9	41.7 + 10.3	0.06

Group 1 subjects had higher mean scores for PSQI, ESS and NRQLQ which were significant as compared to group 3.

Mean score	allergic rhinitis only (group 1) N = 75 (36%)	controls (group 3) n = 23	P value
PSQI	8.6 + 4.4	1.2 + 0.5	0.001
ESS	7.4 + 3.2	2.3 + 0.8	0.012
NRQLQ	46.7 + 10.3	4.7 + 0.5	0.016

158 subjects (84%) had persistent disease whose mean PSQI, ESS and NRQLQ scores were significantly higher as compared to 33 patients (16%) having intermittent allergic rhinitis.

Mean score	persistent allergic rhinitis n = 158 (84%)	intermittent allergic rhinitis n = 33 (16%)	P value
PSQI	9.7 + 4.4	4.0 + 4.5	0.016
ESS	13.3 + 7.0	7.0 + 7.1	0.028
NRQLQ	41.0 + 15.1	20.1 + 12.1	0.001

133/158 (84.2%) subjects with persistent allergic rhinitis and sinusitis had significantly higher means for PSQI, ESS and NRQLQ scores respectively as compared to 25/158 (15.8%) patients having persistent allergic rhinitis without sinusitis.

Mean score	persistent allergic rhinitis with sinusitis n = 133/158 (84.2%)	persistent allergic rhinitis without sinusitis n = 25/158 (15.8%)	P value
PSQI	9.8 + 4.5	5.2 + 5.0	0.001
ESS	13.1 + 7.5	8.61 + 7.3	0.01
NRQLQ	41.6 + 15.8	24.5 + 20.7	0.016

Patients of allergic rhinitis with urticaria/eczema reported higher mean scores on all three questionnaires as compared to those without skin allergy (P = 0.01). 55/207 (26.6%) patients were classified as "sneezer-runners" and 152/207 (73.4%) patients were classified as "blockers". "Blockers" had significantly higher means of 9.5 + 4 (P = 0.001), 11 + 5.6 (P = 0.005) and 46.6 + 10.3 (P = 0.015), as compared to 7.4 + 3.4, 8.8 + 4.5 and 27.5 + 8.1 among "sneezer-runners" for PSQI , ESS and NRQLQ scores respectively.

Conclusion: Nocturnal sleep disturbances and excessive daytime sleepiness are significantly associated and result in significant impairment of sleep specific QOL in allergic rhinitis. These are higher in patients with concomitant sinusitis, persistent disease, "blockers" and allergic skin disorders.

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Seasonal allergic rhinitis symptoms correlate with the quality of life in patients with ragweed allergy in the environmental exposure chamber model

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Seasonal allergic rhinitis (SAR) is a hypersensitive immune response to allergens that causes nasal and ocular symptoms and leads to significant decline in the sufferers' Quality Of Life (QOL). In the Environmental Exposure Chamber (EEC) model of allergen exposure, the relationship between SAR scores and their QOL has not been investigated. To this end, a specialized Rhinoconjunctivitis QOL (EEC-RQOL) Questionnaire has been developed to evaluate the QOL problems experienced by patients while they are in the chamber. The questionnaire contains 16 questions in 4 domains: Non-nose/Eye symptoms (NNE), practical problems (PP), emotional problems (EP), and global assessment (GA).

Aims: To examine whether correlations exist between QOL and instantaneous scoring of SAR symptoms in study subjects at the EEC. Secondly, to determine the effect of sequential EEC visits on QOL.

Methods: A group of 208 Ragweed-sensitive study subjects were exposed to Ragweed pollen (3500 ± grains/m3) in the EEC for 3h in which they recorded instantaneous TSS every half-hour on 4 consecutive days. The relationship between EEC-RQOL scores and TSS over the 4 visits was examined using Pearson's correlation and linear regression.

Results: During each of the four visits, all individual domains as well as the total EEC-RQOL scores significantly correlated with the symptom scores (see Table 1). As subjects participated in subsequent EEC visits, the line of linear regression from 1st to 4th visit shows a clear rightward shift and a gradual increase in slope.

Conclusion: The QOL measured in the EEC using EEC-RQOL Questionnaire significantly correlated with the SAR symptoms of subjects on all visit days. The change in linear regression over the four visits indicates an increasing SAR severity, and that is accompanied by a subsequent decline in QOL. Moreover, subjects appear to experience a greater deterioration in QOL over visits as its rate of decline is faster than the worsening of SAR symptoms. Our findings suggest the negative effect on QOL associated with EEC confinement and thus confirm the importance of developing an EEC-specific RQOL questionnaire. Furthermore, we have demonstrated its cross-sectional construct validity. Table 1: Correlation of QOL Scores to TSS in EEC from Visits 2–5.

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Quality-assessment of disease-specific quality of life questionnaires for rhinitis and rhinosinusitis: a systematic review

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Background: In recent years multiple HRQL questionnaires have been developed specifically for rhinitis and rhinosinusitis. Several reviews described HRQL questionnaires concerning rhinitis and/or (rhino) sinusitis. However, little attention has been paid to the quality of the psychometric properties of these questionnaires.

Objective: The aim of this systematic review is to give a quality-assessed review of the existing disease-specific health related quality of life questionnaires concerning rhinitis and rhinosinusitis for adults.

Methods: We reviewed the literature up to May 2007 in Pubmed, EMBASE and Medline, to identify studies of interest. Additionally the database of the AAAAI Quality of Life Resources and the Patient-reported Outcome and QOL Instruments Database were searched. The quality is assessed by defining the characteristics of a quality of life questionnaire with assessment criteria.

Results: The results of the construction, description, feasibility, and the psychometric performance of the instruments are provided. We finally provide a clinician's guide to choose a questionnaire based on the measurement goals, the discriminant validity, responsiveness and the points obtained in the quality assessment. Of the top scoring instruments regarding the overall quality assessment, only 4 health related quality of life

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RQOL Questionnaire Domains	Visit 2	Visit 3	Visit 4	Visit 5
PP	0.565, p < 0.001	0.361, p < 0.001	0.426, p < 0.001	0.457, p < 0.001
NNE	0.244, p < 0.001	0.279, p < 0.001	0.258, p < 0.001	0.282, p < 0.001
EP	0.272, p < 0.001	0.260, p < 0.001	0.252, p < 0.001	0.244, p < 0.001
GA	0.292, p < 0.001	0.262, p < 0.001	0.168, p < 0.02	0.216, p < 0.002
Total Score	0.368, p < 0.001	0.337, p < 0.001	0.329, p < 0.001	0.344, p < 0.001

questionnaires for rhinitis and rhinosinusitis met our criteria for the discriminant validity and responsiveness.
Conclusion: The current review with its quality assessment of the existing disease-specific HRQL questionnaires concerning rhinitis and rhinosinusitis for adults assists in making a deliberate choice for the most appropriate questionnaire, in a specific context.

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Eustachian tube dysfunction in children with allergic rhinitis

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The cause of a disorder of ventilation and drainage of the middle ear is mainly a disorder opening mechanism of the Eustachian tube.The purpose of the study is to evaluate the persistent symptoms of serous otitis media in children with adenoid hypertrophy and allergic rhinitis.
Materials and Methods: The group study is made up of 128 patients with adenoid hyperplasia and serous otitis media, examined and treated in the Pediatric Hospital of Oradea. From all of this, 72 patients were diagnosed with allergic rhinitis.

The objective endocavitary E.N.T. examination was systematically performed on all patients. Evaluation criteria: degree of hearing loss, serous rhinorrhea, nasal obstruction and sneezing.

We perform adenoidectomy under general anesthesia in all the patients. In the second phase, 3 and 6 month later, patients were re-evaluated clinically and audiological.
Results: In the case of patients group just with adenoid hypertrophy, serous otitis media was remitted in 89% of cases at 3 month re-evaluation. In the case of patients with allergic rhinitis and adenoid hypertrophy, serous otitis media was remitted just at 39% of cases, with fluctuant evolution. At 6 month re-evaluation, after treatment with desloratadine, mucolitic agents, the serous otitis media was remitted completely. On second evaluation, in the case of the first group, the study showed the reduction of nasal obstruction in 78% of patients as compare to the second group, where recovery was present in only 38% of patients, certified by hearing tests and clinical aspects of tympanic membrane at otoscopy.
Conclusion: As a result of adenoidectomy, improvement in permeability of the Eustachian tube, diminish the effusion into the middle ear, but they persist in the case of patients with associated allergic rhinitis. Antihistaminic drugs, desloratadine, have an important role to prevent recurrent attacks of serous otitis media in children.

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Allergic rhinitis and asthma survey: clinical and psychological perspectives

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Background: The existence of a close link between asthma and rhinitis, which brings consequences both in therapy and disease management, has been demonstrated by many studies.
Objectives: The trial we performed had the following aims: to assess physicians' knowledge on rhinitis/asthma comorbidity, to evaluate patients' management behaviour and their experience about symptoms and expectations, to investigate the clinical and psychological meaning of GPs' and patients' knowledge.
Methods: 101 general practitioners and 504 asthmatic patients were involved in the study. They were asked to fill in two different multiple-choice questionnaires about the association between asthma and rhinitis and its impact.
Results: 34.7% of general practitioners are aware of the asthma-rhinitis link, and 43.6% of them assume the comorbidity on the basis of their clinical experience. 21.8% of physicians make the diagnosis autonomously. 27.8% of asthmatic patients experience three or less rhinitic symptoms, 41% from 4 to 6 symptoms, and 31.2 more than 6 symptoms. The symptoms have a deep impact on daily life, as they are cause of sleep problems (87.3%), lack of concentration (78.9%), difficulties in spare time (71.8%) and sport (71.7%). Rhinitis symptoms are responsible for the worsening of asthma, with an increase of dyspnoea (86.3%), cough (73.9%) wheezing (59%). 93% of patients state they are interested in a combination therapy approach.
Conclusion: Asthma and rhinitis are associated in the development of clinical burden and influence on quality of life. The survey results assume different perspectives if are evaluated from the clinical and psychological point of view.

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Clinical efficiacy of antileukotriene threapy in allergic rhinitis: changes in subjective and objective parameters, and quality of life measures

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Background: Leukotrienes and histamine are thought to be quantitatively the most prominent mediators in the final pathways of allergic rhinitis (AR). Intranasal corticosteroids and oral antihistamines are the cornerstones of therapy. Antileukotriene therapy combined with antihistamine showed a synergistic effect in treating AR, however, there is little data on objective parameters and quality of life measures.
Methods: This multi-centered, prospective, randomized and plabebo controlled study randomized 275 patients with mild or moderate intermitant AR into three groups as: fexofenadine (120 mg/day), fexofenadine plus montelukast (10 mg/day) or fexofenadine plus placebo. The nasal endoscopic examinations, objective upper airway measurements (anterior rhinomanometry), were evaluated before and at end of 21 days treatment period. Daily symptom scored by a standard visual analoge scale (VAS) and by a daily diary of quality of life measures.
Results: Nasal endoscopic examination showed an additional effect on turbinate congestion with combination therapy. Objective nasal airway assessment revealed that total nasal resistance decreased from 0,42 Pa/cm3/s to 0,32 Pa/cm3/s with monotherapy, and from 0,43 Pa/cm3/s to 0,27 Pa/cm3/s with combination therapy ($p < 0,05$) in average. Mean symtom scores of nasal congestion, nasal itching and sneezing pointed out a significant decline in the first three days, however nasal congestion scores were better with the antileukotriene add-on therapy. The positive effect on quality of life measures mainly in sleep, daily life activities and performance were increased, but combination therapy revealed significantly better results at the end of 21 days. It is significantly more effective than placebo. No side effects were encountered.
Conclusion: These data provide a basis for optimism in the control of AR with antileukotriene-antihistamine combination therapy. The control on nasal congestion was more pronounced subjectively and objectively comparing to antihistamine alone. The effect might be due to the additional anti-inflammatory activity as provided by reduction of inflammatory infiltrate and cytokine levels. More long-term studies are needed to evaluate the clinical effectiveness of antileukotrienes, especially as add-on therapy. Available data suggest it is reasonable and safe to add these agents to standard therapy if nasal symptomatology remains unresolved.

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The role of montelukast on perennial allergic rhinitis and associated sleep disturbance and daytime somnolence

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Background: One of the main manifestations of perennial allergic rhinitis is congestion. Nasal congestion can predispose to sleep apnea and microarousals. This sleep disturbance can result in daytime somnolence and fatigue. The use of montelukast may be effective at reducing congestion, but objective and subjective studies are lacking that demonstrate if the reduction of congestion will result in improved sleep and reduced somnolence. It was the purpose of this research to determine if montelukast is effective at improving sleep and daytime somnolence in patients with perennial allergic rhinitis.
Methods: The study was a crossover, double-blinded, placebo-controlled study using montelukast 10 mg tablet or matched placebo and was approved by the IRB. Active phase of the study included a 2-week run-in followed by a treatment period of 2 weeks. After a wash out of 2 weeks, subjects were crossed over to the alternate arm of the study for 2 weeks. Subjective instruments to assess sleep and daytime somnolence were utilized. SAS was used for statistical assessment.
Results: Montelukast 10 mg as compared to placebo statistically improved daytime sleepiness ($p = 0.0089$) and daytime fatigue (0.0087). With our small cohort we were unable to significantly demonstrate decreased congestion; however; congestion was reduced by 0.52 on a scale of 0–3, while placebo reduced congestion by 0.16.
Conclusion: Montelukast can reduce daytime somnolence and fatigue and may be a suitable alternative to topical nasal corticosteroids in those unwilling to use or are intolerant to nasal steroids and have daytime impairment from perennial allergic rhinitis.

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Intranasal phototherapy is more effective than fexofenadine hydrochloride in the treatment of seasonal allergic rhinitis

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We recently showed that intranasal phototherapy represents an efficient therapeutic modality in the treatment of allergic rhinitis. The aim of this study was to compare the efficacy of intranasal phototherapy with that of a second generation antihistamine fexofenadine HCl in allergic rhinitis. A randomized open study was conducted in patients with history of moderate to severe ragweed-induced allergic rhinitis. Thirty-one patients were randomly assigned to receive either intranasal irradiation (low doses of UV-B, UV-A and visible light, referred to as mUV/VIS) 3 times a week for 2 weeks ($n = 18$), or 180 mg fexofenadine HCl per day for 2 weeks ($n = 13$). Each patient kept a diary of symptoms for nasal obstruction, nasal itching, rhinorrhea, sneezing and palate itching during the treatment. Total nasal score (TNS), a sum of scores for nasal symptoms was also calculated. In the mUV/VIS group the individual scores significantly decreased compared with baseline for all of the parameters, sneezing ($p = 0.0001$), rhinorrhea ($p = 0.0004$), nasal itching ($p = 0.0005$), nasal obstruction ($p = 0.009$) and palate itching ($p = 0.0001$). In the fexofenadine HCl group none of the scores improved significantly at the end of the treatment except sneezing ($p = 0.03$). TNS was significantly decreased in the mUV/VIS group ($p = 0.00003$), but no significant change was observed in the fexofenadine HCl group. In conclusion, we found that intranasal phototherapy is a more efficient therapeutic tool than fexofenadine HCl in reducing clinical symptoms for allergic rhinitis.

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The burden of illness of allergic rhinitis in Canada

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Background: The objective was to assess the burden of symptoms in Canadian adults with allergic rhinitis (AR).
Methods: A cross-sectional, random-digit-dialing telephone survey of 30,987 Canadian households was conducted in July 2006 to identify adult AR patients. After screening 3671 adults, structured interviews were done with 1001 respondents (patients diagnosed by a physician as having AR or taking medication for AR).
Results: About 45% of Canadian adults report suffering from nasal symptoms due to allergies unrelated to colds. Less than half (45%) have been diagnosed by a physician. Half only have seasonal symptoms, with spring and summer being the worst seasons. Of those with AR, 27% had asthma, 17% chronic or recurrent sinusitis and 5% nasal polyps. More than one-quarter cannot tolerate their symptoms without treatment. Most (83%) have sought medical attention for their symptoms at one time and one-quarter have done so in the past year. The most bothersome symptoms include stuffy nose, runny nose, repeated sneezing and watering eyes. In their worst month, two-thirds of patients reported having a stuffed nose either daily or several days per week. Almost one-quarter reported headaches and sleep loss. One-fifth describe symptoms as poorly controlled or not controlled during the worst month of the year. One-half use only OTC products, 12% use only prescriptions while one-quarter use both.
Conclusion: Despite treatment, many Canadians experience allergic rhinitis symptoms that could be better evaluated and controlled. Asthma, sinusitis and nasal polyposis are common concomitant conditions.

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Radiofrequency tissue ablation treatment in persistent allergic rhinitis: effects on quality of life and objective parameters

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Background: In patients who suffer from persistent allergic rhinitis (PAR), a severe drug-resistant hypertrophy and increase in glandular structures of the inferior turbinates may develop, which leads to constant nasal obstruction and rhinorea. Radiofrequency tissue ablation of the turbinates can reduce nasal obstruction and secretions.
Methods: This prospective, single-sited study randomized 50 patients with mild or moderate PAR who had substantial bilateral hypertrophy of the inferior turbinates to mometasone furoate monohydrate nasal spray (MFMNS), (2 sprays per nostril [total dose 200 μg] once daily), or radiofrequency inferior turbinate ablation (RIFA), (2 or 3 punctures on each site, total dose of 1510 ± 110 joule, plateau temperature of 75°C, energy of 10 watt) treatment groups. Both objective outcomes evaluated by total nasal resistance at anterior rhinomanometry and subjective outcomes assessed with the Quality of Life Questionnaire were analyzed before and at least 12 months after treatment.
Results: The median total nasal resistance in patients treated with MFMNS decreased from 0,49 ± 0,17 $Pa/cm^3/s$ to 0,39 $Pa/cm^3/s$ ($p = 0,42$), and 0,51 ± 0,18 $Pa/cm^3/s$ to 0,29 $Pa/cm^3/s$ in patients with RIFA ($p = 0,003$) 1 year postoperatively. Compared with preoperative scores, the postoperative scores of these patients significantly improved in both 7 separate domain scores and overall Quality of Life Questionnaire scores ($p = 0,004$). Nasal symptomatology was markedly reduced 1 month after radiofrequency application. No adverse reactions including bleeding, infection, adhesions or worsening of allergic symptoms were encountered. The patients experienced a lasting benefit from this procedure.
Conclusion: These results suggest that topical mometasone reduces the volume of inferior turbinate at some point while improving the quality of life in patients with PAR. Radiofrequency inferior turbinoplasty is also

improving quality of life and is more effective for decreasing nasal resistance in patients with PAR who have substantial nasal congestion. This effect might be due to the switch between the inflammatory cells and the fibrotic tissue. Histopathologic and longer term studies are required to enlighten the potential of RIFA in the management of allergic rhinitis.

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The role of allergic rhinitis in suppurative chronic otitis media, prelaminary reports

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Background: Eustachian tube dysfunction has a significant effect on pathogenesis of ear diseases especially chronic otitis media (COM). In the other hand, allergic rhinitis as a prevalent disease is a well-known condition which has some effects on eustachian tube function and its role in serous otitis media has been studied from several aspects, but its effects on chronic suppurative otitis media with & without cholesteatoma has not been studied yet.

Methods: In a prospective case control study, 40 patients with suppurative chronic otitis media who were candidates for surgery and 38 healthy, sex and age-matched persons were evaluated for allergy using the standard questionnaire, prick test and serum IgE.

Results: Fifteen of patients (37.5%) and six of controls (15.8%) have allergic rhinitis. The results showed the higher incidence of allergy in the patients' group ($P < 0.05$).

Conclusion: To the authors' knowledge, this is the first documented survey showing the correlation between allergy and chronic otitis media. More extensive studies on the effect of allergy especially allergic rhinitis on chronic otitis media even postsurgical prognosis strongly recommended.

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Nasal mucosa remodeling and re-activation of epithelial mesenchymal trophic unit in human allergic rhinitis

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Objective: To investigate the histopathologic characteristic of a marked chronic inflammatory reaction and remodeling, and that the role of re-activation of epithelial mesenchymal trophic unit in this marked structure change of the nasal mucosa in allergic rhinitis.

Methods: Our study will be divided into two parts. The first part is to investigate whether remodeling of the airway mucosa are present in nasal mucosa of allergic rhinitis. The second part is to investigate the possible role of epithelial mesenchymal trophic unit in the pathogenesis of inflammation and remodeling in allergic rhinitis. The tissue blocks were embedded in paraffin, and stained with hematoxylin and eosin (HE), alcian blue periodic acid-Schiff (AB-PAS), Masson's Trichrome(MT), and immunohistochemical staining. The infiltrating eosinophils in nasal mucosa were examined, AB-PAS-positive cells in the surface epithelium in nasal mucosa were counted. The percentage area of MT stained extracellular matrix in mucosa and conchae and damage of epithelium were determined by an image analyzer. immunohistochemical staining revealed EGF, EGFR, TGF-β, and α-SMA immunoreactivity in nasal mucosa.

Results: Epithelial damage (shedding), basement membrane thickening, submucosa fibrosis, mucosal goblet cell hyperplasia, submucous gland hyperplasia, and strikingly eosinophil infiltration, lymphocyte infiltration was more prominent in patients with AR compared to the non-AR group ($p < 0.05$). Compared with the control group, immunohistochemical staining revealed EGF, EGFR immunoreactivity in the damaged epithelium were decreased in allergic rhinitis. However, stronger TGF-β and α-SMA immunoreactivity was observed in allergic rhinitis mucosa as compared with normal nasal mucosa.

Conclusion: The histopathologic findings of asthma, epithelial damage, goblet cells hyperplasia and extracellular matrix deposition, namely, inflammation and features of airway remodeling, are also present in allergic rhinitis. It maybe result from activation of the epithelial-mesenchymal unit, leading to myofibroblast activation and propagation of remodeling responses into the submucosa. The concept of epithelial-mesenchymal unit maybe contribute to the further understanding the pathogenesis of allergic rhinitis and may provide potential targets for novel therapy of allergic rhinitis.

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Rhinophototherapy in grass pollen induced allergic rhinitis

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Objectives: Our previous randomised double blind clinical study proved that intranasal phototherapy using a combination of UVB (5%), UVA (25%) and visible light (70%) (mUV/VIS)(Rhinolight®) is an effective modality to treat allergic rhinitis (AR). The aim of this study was to show our clinical experiences on grass pollen sensitized allergic patients.

Methods: We have treated AR patients (N = 243) with intranasal mUV/VIS between 2003–2006. 79 patients had moderate/sever intermittant grass pollen induced AR. The pollen counts were over 40/m3 continuously. Rhinophototherapy was performed in monotherapy or combined with oral antihistamines (once a day) or/with nasal steroids (maximum 400 μg/die). Each intranasal cavity was irradiated 2–3 times a week, for 2 weeks with mUV/VIS. The dose of irradiation was raised step by step from 1.6 to 2.7 J/cm2/nasel cavity. Nasal symptoms of each patient were registered on the treatment days with a scale of 0 to 3 and the total symptoms on a visual analog scale (VAS: 0–10).

Results: 3 patients dropped out (3,8%). Finally 75 patients got full therapy. Monotherapy mUV/VIS: got worse: 6% of the patients, didn't change 19%, improved: 75% by VAS. mUV/VIS+drugs: got worse 5%, didn't change 8%, improved 87%. The nasal symptoms were significantly improved in each group. Rhinophototherapy was tolerated well. Using vitamin A oil 3–4 times a day the development of painful xeromucosa could be avoided. Only 2 patients (2,53%) needed to have extra (more than one) brake days between two treatment occasions.

Conclusion: These results suggest that intranasal phototherapy alone or with drug combined is an effective modality to treat grass pollen induced AR.

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Clinical characteristics of children with allergic rhinitis (AR) vs those with nonallergic rhinitis (NAR) from a university pediatric allergy clinic

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Background: Proper differentiation of chronic rhinitis into AR and NAR is essential since avoidance of offending allergen in AR could rapidly lead to alleviation of symptoms whereas NAR is usually a chronic and non-remitting condition. Unfortunately, allergy skin test is not feasible in general practice in several countries around the world. The aim of this study is to determine specific characteristics among pediatric patients with AR and NAR.

Methods: Three hundred and two children with symptoms of chronic rhinitis presented to a Pediatric Allergy Clinic between January to December 2006 were categorized into 2 groups, i.e, allergic rhinitis (AR) and nonallergic rhinitis (NAR) according to their skin prick test results. Nasal cytology was performed to further subcategorize NAR into nonallergic rhinitis with eosinophilia syndrome (NARES).
Results: There were 222 (73.5 %) patients with AR and 80 (26.4 %) with NAR. Nineteen (23.7%) NAR patients had NARES. The median age of onset in patients with AR and NAR were 4 and 3 years old, respectively. Symptoms of rhinorrhea, postnasal drip and nasal congestion did not differentiate the two groups apart. Interestingly, nasal itching, sneezing and eye symptoms were more common among AR than in NAR group ($p < 0.01$) while snoring and sinusitis were more common in NAR than in AR group ($p < 0.01$). Seasonality, severity of symptoms and the proportion of asthma did not differ between both groups. Prevalence of food allergy were more common in AR than NAR group ($p < 0.05$).
Conclusion: AR was about threefold more prevalent than NAR among children presented to our allergy clinic with chronic rhinitis. Nasal itching and sneezing were more common in AR than NAR group ($p < 0.01$) while snoring and sinusitis were more common in NAR than AR group ($p < 0.01$). Eye symptoms and history of food allergy were more common in AR than NAR group ($p < 0.01$, $p < 0.05$).

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Impact of levocetirizine on symptoms of allergic diseases. Result of questionnaire survey in Hungary in 2005

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Allergic diseases are the increasing global health problem either in all over the world or in Hungary. In 2006 the incidence of allergic rhinitis was 25429 new cases (252‰), and the prevalence 263 925 (2619 ‰), and incidence of allergic asthma was 12 693 (126,0‰) and prevalence 141 561 (1404,,9‰).

The basic treatment of the allergic disease has been H-1 receptor antagonists, and the levocetirizine is a new oral, non-sedating H-1 antihistamine, that has been shown to be effective against allergic symptoms, and offers good tolerability.
Objective: To collect data concerning the treatment of allergic disease in general practice. to measure the effectiveness of levocetirizine in the allergic disease. to report the side effects during the levocetirizine therapy.
Methods: A nationwide survey was organized in 2005 in Hungary with using the same questionnaire, filled two times (at the start and at he end of treatment period) in.The patients participated in survey had allergic disease and was treated by levocetirizine during 6 months.Data of 17800 questionnaires were analysed. 57,8% of patients were female and 42,2 % male ($p < 0,001$). 57,2% of all patients has been aged 20–49 y, 19 % was younger than 19 y.
Results: 14 637 patients (82%) had allergic rhinitis, 3335 (19%) had urticaria, 3660 (20%) had allergic asthma, 1048 (6,8%) had atopic dermatitis, and 365 (2%) had food allergy. 57,1% of patients had symptoms of allergic disease longer than 4 years. Cutan test was performed in diagnosing of allergic disease in 78,5 % of patients, and in 92% was positive.

The allergic disease influenced the way life of patients in 76,3%, but 88% of patients had clinically important symptoms (serious 40,4 %, or intermedient 47,4%). At the end of the survey 6,7% of patient had clinically significant symptoms, so in 93,3 % of patients's symptoms improved. The 58,5% of allergic rhinitis and 51,0% persistent rhinitis patients become symptomless, and the ratio of symptom free cases was 51,3% of allergic asthma, 43,8% of atopic dermatitis, 51,9% of food allergy and 47,0% of urticaria. 52,4% of patients were treated by combination therapy. Side effects occured in 2,1% of patients during the treatment period.
Summary: According the results of 6 months treatment of allergic diseases by levocetirizine it has been established that the levocetirizine was effective, decreased the clinical symptoms of allergic diseases, was well tolerated without important side effects.

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Frequency of adenoid hypertrophy and atopy in childhood

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Background: The adenoid is a part of Waldeyer's ring, the basic function of which is antibody formation, that react later against a great variety of antigens. Pediatric adenoidal obstruction of the nasal airway is associated with significant morbidity and is a frequent indication for surgery, but its relationship to allergy has not been often studied.
Methods: To examine the influence of atopy on the adenoid hypertrophy we studied 35 children 1–11 years old. In the analysed group all children suffered from difficulty in nose breathing, rhinorrhea, recurrent upper respiratory tract infections, hearing disorders, sleep disturbance. For the diagnostic of adenoid hypertrophy we had used the anamnesis, anterior rhinoscopy, endoscopy rhinoscopy. All patients had been consulted by the allergist-immunologist. For the diagnosis of allergic rhintis we had used interview, skin prick-test, presence of sIgE (RAST) in the serum, nasal provocation test with dust and polen allergens.
Results: All children had had adenoid hypertrophy 2 or 3–4 degree, 3 of them had got the relapse of adenoids after the surgery. In the analised group 21 children (60%) had had the positive skin prick-tests and/or positive reactions to specific IgE, mostly to different kinds of pollen or house dust mites; eosiniphils in nasal secrete; positive nasal provocation tests. In this group 9 patients (25%) had had the bronchial asthma, and 6 children (17%) – atopic dermatitis. All patients with adenoids and allergic rhinitis had got the complex therapy – topical nasal steroid and antihistaminic in the ages doses. The children with brochial asthma and atopic dermatitis also received a pharmacological treatment. In a during of 3 weeks we observed the most significant decrease of clinical symptoms and endoscopic adenoid size.
Conclusion: The study shows that allergy and sensitivity to different kinds of allergens is an important risk factor for a greater degree of adenoid hypertrophy in children. Chronic allergic inflammation of the upper airway is the causes of lymphoid hypertrophy with prominence of adenoidal and tonsillar tissue. So, all patients with the difficulty in nase breathing and reccurent upper respiratory tract infections need in the consultation by allergist-immunologist for exception of allegic diseases.

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The quality of life in patients with atopic asthma associated with allergic rhinitis

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Objectives: The aim of the study was to assess the effect of allergic rhinitis on quality of life in patient with atopic asthma.

Methods: We examined 45 atopic asthmatics patients with persistent allergic rhinitis and 36 patients with atopic asthma without allergic rhinitis. Quality of life was measured by the Asthma Quality of Life Questionnaires (AQLQ). Clinical symptom score and use of rescue medications were recorded in diaries for 4 weeks before the assessment of quality of life. Lung function was measured on the same day as the study of quality of life.

Results: Clinical symptom score and use of short–acting β 2–agonists were similar in both groups and there was no difference in FEV1 (63.00 3.75 vs. 65.84 3.58, $p > 0.05$). Asthma symptom scores (2.83 0.14) and activity limitation (3.47 0.22) within AQLQ were significantly lower for patients with asthma and allergic rhinitis compared with those asthma patients without allergic rhinitis (4.01 0.21 and 4.23 0.25, $p < 0.05$).

Conclusion: Persistent allergic rhinitis may have a negative effect both on asthma symptoms and on quality of life in patients with atopic asthma.

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Study of the prevalence of aeroallergens in the patients with allergic rhinitis referred to semnan allergy clinic between December 2005 to December 2006

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Background: Allergic rhinitis is the most common cause of rhinitis affecting approximately 20% of the population. While allergic rhinitis is not a life-threatening condition, complications can occur and the condition can significantly impair quality of life, which leads to a number of indirect costs. In susceptible individuals, exposure to certain foreign proteins leads to allergic sensitization, which is characterized by the production of specific IgE directed against these proteins. This specific IgE can be tracked with Standard Skin Prick Test.

Methods and Materials: During 6 months of study, 334 cases of allergic disorders referred to our allergy clinic, among them 295 were allergic rhinitis. Standard Skin Prick Test with ALUSTAL prick allergens, aero-allergens and food allergens, were performed for all of them and compared with negative and positive standard control. Wheals of 3-mm and flares of 10-mm larger than negative control regarded as positive test. Results were analyzed with SPSS 11.5 and prevalence of allergens was calculated in different groups of patients in regard to age, sex and type of allergic rhinitis (perennial or seasonal).

Results: 150 males with mean age of 25.6 years (SD: 14.31) and 145 females with mean age of 28.32 (SD: 12.93) were studied among them 85 patients (30%) had seasonal Allergic Rhinitis (SAR), 117 patients (41.3%) had Perennial Allergic Rhinitis (PAR), 65 patients (23%) had PAR with seasonal aggravation and 16 patients (5.7%) had only episodic symptoms. 116 patients had less than 5 years, 117 patients had 5–10 years and 102 patients had more than 10 years history of rhinitis. Aeroallergen sensitization were detected in 256 (86.5%) of patients and botanical allergens' sensitization were shown to be present in 225 (76%) of patients. Chenopodiacea including Chenopodium alba and Rough pigweed, trees especially ash, platanus, willow and birch, 12 grasses including artemisia, rye-grass and timothy were among the most prevalent botanical allergens.

Conclusion: Allergic rhinitis is the most prevalent allergies which cause signs and symptoms in genetically susceptible patients confronting environmental allergens. In our study, aeroallergens were studied. As in other studies, aeroallergens especially botanical allergens were shown to be the most common allergens both in Seasonal and perennial allergic rhinitis with 86.5% sensitive to one or more aeroallergens and 76% sensitive to one or more botanical allergens.

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Rhinitis: is it allergic or not?

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Introduction: there are no acceptable criteria for distinguishing between allergic and non-allergic rhinitis. The aim of this study was to compare the clinical and para-clinical features of allergic and non-allergic rhinitis.

Methods: in a retrospective study, we compared the clinical and para-clinical features of 442 allergic and 103 non allergic rhinitis patients (18–50 year-old). The evaluations included respiratory signs and symptoms, lab tests, and respiratory function tests.

Results: patients with allergic rhinitis had more signs and symptoms within their season in terms of cough, sneezing, nasal congestion, rhinorrhea, itchy eyes and nose, and respiratory function test ($p < 0.001$), whereas those with non-allergic rhinitis had more persistent symptoms, and headaches ($p < 0.01$). Food allergy, OR = 1.8, (1.4–2.1), $P < 0.01$, serum Ig E, OR=2.1, (1.9–2.19), $P < 0.001$, and airway hyperresponsiveness, OR=1.7 (1.4–1.9), $P < 0.01$ were more common in patients with allergic rhinitis.

Conclusion: our results indicate that the signs and symptoms have more fluctuation in allergic rhinitis, and more sever within their season. More studies with respect to both adults and children with rhinitis is recommended.

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Allergic and non-allergic rhinitis in adolescent and adults

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Background: The aim of this study was to describe differences between allergic rhinitis (AR) and nonallergic rhinitis (NAR) in a large community-based sample of adolescents and adults.

Methods: A total of 354 subjects, 10–50 years of age, who in a screening questionnaire had reported a history of airway symptoms suggestive of asthma and/or allergy, or who were taking any medication for these conditions were clinically examined. All participants were interviewed about respiratory symptoms and furthermore skin test reactivity, lung function and airway responsiveness were measured using standard techniques.

Results: A total of 64% of the subjects with rhinitis had AR, whereas 46% had NAR. Subjects with NAR were more likely to be females, OR = 2.13 (1.24–3.22), P = 0.004, to have persistent symptoms within the last 6 weeks,

OR = 1.87 (1.23–2.65), P = 0.002, and to have recurring headaches, OR = 1.94, (1.34–3.45), P = 0.021. On the other hand, subjects with NAR were less likely to have airway hyperresponsiveness, OR = 0.34, (0.32–0.46), P < 0.001, food allergy, OR = 0.32, (0.18–0.33), P = 0.007 and to have been treated with antihistamines in the last 6 weeks, OR = 0.23, (0.22–0.35), P < 0.002 compared with subjects with AR. Subjects with AR were symptomatically worse within their season in terms of sneezing (P < 0.002) and itchy eyes (P < 0.002), compared to subjects with NAR, whereas nasal congestion and rhinorrhea were equally frequent in the two groups (P = 0.32 and P = 0.44, respectively).

Conclusion: The proportion of subjects with NAR in an adolescent and adult population with rhinitis is around one-fourth. Women have NAR twice as often as men. In general, subjects with NAR have more persistent but equally severe symptoms compared to subjects with AR. However, subjects with allergic rhinitis have more sneezing and itchy eyes within their particular season of allergy compared to subjects with nonallergic rhinitis.

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Relationship between histamine H1 receptor occupancy (RO) and reduction of symptoms of seasonal allergic rhinitis (SAR) by levocetirizine (L) in subjects exposed to pollen in an environmental exposure unit (EEU)

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Background: the estimation of in vivo RO, which takes into account the affinity of an H1 antihistamine for the histamine H1 receptor and its free plasma concentration, is a far better predictor of human pharmacodynamics evaluated by the inhibition of histamine or allergen-induced wheal & flare than considering in vitro affinity and plasmatic half-life only (Gillard et al. Inflamm Res 2005, Frossard et al. Br J Clin Pharm 2007). The purpose of this study is to assess whether this is valid also for other pharmacodynamic models such as that scoring the symptoms of SAR in ragweed sensitive subjects exposed to ragweed pollen in an EEU.

Methods: 119 subjects (mean age 34 yrs, mean bodyweight 81 kg) were exposed to pollen in the EEU at 9 am of Day 1. At 11 am, they received a single dose of 5mg L and remained in the EEU until 4 pm. On Day 2, subjects were exposed again to pollen in the EEU from 8 am until noon. The primary efficacy variable was the reduction from baseline in the Major Symptoms Complex (MSC) score (calculated as the sum of the following 6 symptoms: runny nose, itchy nose, sniffles, nose blows, sneezes and watery eyes). Baseline value was the MSC score before drug administration (9–11 am of Day 1). The MSC score was evaluated at different times after L administration (see table). Plasma concentrations at different times after administration of a single dose of 10 mg L were obtained from a study in 24 healthy volunteers (mean age 35 yrs, mean bodyweight 67 kg) (Baltes et al. Fund Clin Pharm 2001), normalized to the therapeutic dose of 5 mg (see table). The plasma protein binding data of L come from Bree et al. (Fund Clin Pharm 2002). The RO has been calculated as described by Gillard et al. (Inflamm Res 2005).

Results: The kinetics of change from baseline in MSC score fit much better with those of RO than with those of free plasma concentrations (see table). It is possible however that the free plasma concentrations of L do not represent those at the receptor at very short times and that the RO values calculated are over-estimated at these times. It is also possible that there is a lag time between the RO and the relief of some of the symptoms.

Conclusion: the EEU model confirms previous results that the kinetics of RO by L are a better predictor of the efficacy of the drug than are plasma pharmacokinetics.

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Expression of matrix metalloproteinase 2, 9 and 13, TIMP 1, 2 in the allergic nasal mucosa

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Background: Matrix Metalloproteinases (MMPs) are extracellular matrix proteins that are involved in tissue remodeling and cell infiltration. Tissue

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Time (h) after levocetirizine administration (single 5 mg dose)	Mean plasma concentrations (ng/ml)	Mean free plasma concentrations (nM)	Mean receptor occupancy (%)	MSC score (mean)	Change from baseline* in MSC score (mean)
0.25	124.3	24.2	89	—	—
0.5	230.3	44.9	94	17.11	0.74
1.0	230.9	45.0	94	12.18	−4.18
1.5	205.0	39.9	93	9.86	−6.51
2.0	189.1	36.8	93	8.71	−7.66
2.5	—	—	—	7.53	−8.83
3.0	168.4	32.8	92	7.24	−9.12
3.5	—	—	—	7.14	−9.22
4.0	151.1	29.4	91	7.24	−9.13
4.5	—	—	—	6.96	−9.40
5.0	—	—	—	7.20	−9.16
6.0	110.4	21.5	88	—	—
9.0	75.1	14.6	83	—	—
12.0	54.5	10.6	78	—	—
16.0	35.1	6.8	69	—	—
21.5	—	—	—	8.18	−8.17
24.0	18.4	3.6	55	8.82	−7.52

* MSC at baseline = 16.36

inhibitors of MMPs known as TIMP regulate MMP function. Allergic rhinitis (AR) and asthma share many similarities in their inflammatory response but epithelial shedding, basement membrane thickening and fibrosis are not seen in AR. By contrast, nasal polyps share some features with asthma. As in asthma, we demonstrated an increased expression of MMP-9 and low TIMP in nasal polyps. We hypothesized that the differential expression of MMPs may at least in part contribute to the differences between AR and asthma.

Methods: We performed nasal allergen challenge (NAC) in AR patients with mite allergen disks and control disks and analyzed the number of MMP 2,9,13 and TIMP 1,2 in nasal biopsies at 30 min, 6 hrs and 12 hrs post challenge. By ELISA, we examined the levels of MMP-2, 9 and 13, and TIMP1,2 in the nasal mucosa of AR patients.

Results: At 30 min post NAC, MMP-2 and 13 were increased. At 6 hrs post NAC, MMP-2 and at 12 hours post NAC, MMP-2 and 13 were significantly increased. TIMP-1 was increased at 30 min. At 30 min and 12 hr post NAC, the MMP 2 : TIMP1,2 and MMP-13 : TIMP1,2 ratio was high. The levels of MMP-2 and 13 but not MMP-9 were high in the allergic nasal mucosa.

Conclusion: These results suggest that MMP-2 and 13 may play an important role in the pathomechanisms of allergic rhinitis and that this differential expression of MMP may contribute at least in part contribute to the differences between AR and asthma.

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CRTH2 plays an essential role in the pathophysiology of Cry j 1-induced pollinosis in mice

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Background: Prostaglandin (PG) D2 is the major prostanoid produced during the acute phase of allergic reactions. Two PGD2 receptors have been isolated, DP and CRTH2, but whether they participate in the pathophysiology of allergic diseases remains unclear. We investigated the role of CRTH2 in the initiation of allergic rhinitis in mice.

Methods: First, we developed a novel murine model of pollinosis, a type of seasonal allergic rhinitis. Pathophysiological differences in the pollinosis were compared between wild-type and CRTH2-gene deficient mice. An effect of treatment with ramatroban, a CRTH2/T-prostanoid dual antagonist, was also determined.

Results: Repeated intranasal sensitization with Cry j 1, the major allergen of Cryptomeria japonica pollen, in the absence of adjuvants significantly exacerbated nasal symptoms, Cry j 1-specific IgE and IgG1 production, nasal eosinophilia, and Cry j 1-induced in vitro production of IL-4 and -5 by submandibular lymph node cells. In addition, CRTH2 mRNA in nasal mucosa was significantly elevated in Cry j 1-sensitized mice. Following repeated intranasal sensitization with Cry j 1, CRTH2-gene deficient mice had significantly weaker Cry j 1-specific IgE/IgG1 production, nasal eosinophilia, and IL-4 production by submandibular lymph node cells than wild-type mice. Similar results were found in mice treated with ramatroban.

Conclusion: These results suggest that the PGD2-CRTH2 interaction is elevated following sensitization and plays a proinflammatory role in the pathophysiology of allergic rhinitis especially pollinosis in mice.

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The clinical course of allergic rhinitis and asthma and changes in skin prick test and spirometry in the aftermath of hurricane Katrina

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Background: Allergic rhinitis and asthma are both prevalent diseases seen in everyday practice. In the aftermath of hurricane Katrina several of our patients reported exacerbations of allergic rhinitis, allergic conjunctivitis and either exacerbations or development of asthma. As a result of Katrina many homes were flooded, causing significant increased mold levels. We sought to investigate if these changes in reported symptoms correlated with changes in Skin Prick Test (SPT) reactivity and/or changes in spirometry.

Methods: At the start of the study, we looked at patients who reported increased allergic symptoms and in whom we had previously performed SPT. We then repeated SPT and compared the results to earlier findings. In addition, patients who had a history of asthma or those complaining of respiratory symptoms had spirometry done and their results were compared with those done before Katrina.

Results: Four of five patients were reactive to many antigens to which they were previously anergic. Reactivity to molds was increased, most commonly Alternaria, Aspergilus fumigatus and Cladosporium. In addition changes in spirometry were also noted. In one patient who initially had normal spirometry with negative methalcholine challenge, developed abnormal spirometry. This patient's FEV1 was markedly decreased and improved by more than 15% after bronchodilators.

Conclusion: There has been much concern about the effect Katrina may have had on patient health. Many patients have demonstrated increased symptoms relating to allergic rhinitis and asthma. These cases demonstrate that atopic patients in New Orleans have been sensitized to new allergens. Patients that are experiencing increased symptoms of rhinitis, conjunctivitis or asthma should be re-evaluated with SPT, spirometry and a thorough history to environmental exposure. With the large number of people affected by Katrina, further studies should be performed as more patients return to the New Orleans area.

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Mechanisms of mast cell migration into the allergic nasal epithelium

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Objective: Mast cells are increased in the nasal epithelium of patients with allergic rhinitis (AR). Yet, the precise mechanisms of this increase are unclear. We previously reported that nasal mast cells express CCR3 and exhibit increased chemotaxis to RANTES suggesting a role for RANTES in mast cell migration. To further confirm this, in the present study we examined the levels of RANTES, eotaxin and SCF in the epithelium and lamina propria of patients with AR and the kinetics of RANTES+, tryptase+, and CCR3+ cells in the epithelium and lamina propria after nasal allergen challenge.

Methods: By ELISA, we examined the levels of RANTES, eotaxin and SCF in homogenized nasal scrapings and lamina propria of AR patients. In AR patients, we performed nasal allergen challenge with house dust mite, took biopsies at 30 min, 6 hrs, and 12 hrs and by immunohistochemistry, we examined the number of Tryptase+ RANTES+, eotaxin+ and CCR3+ cells as compared to control.

Results: The levels of RANTES, but not Eotaxin and SCF was greater in the epithelium than in the lamina propria. At 30 minutes after nasal allergen challenge, Tryptase+, RANTES+ and CCR3+ cells were increased in the epithelium. At six hours post challenge, Tryptase+ and RANTES+ cells were increased in the epithelium but at 12 hrs only an increase in Tryptase+ cells was detected.

Conclusion: Migration of the mast cells in the allergic nasal epithelium occurred as early as 30 min with a parallel increase in CCR3+ and RANTES+ cells. These results further confirm that RANTES is one of the critical factors regulating mast cell migration into the allergic nasal epithelium.

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The effect of levocetirizine and dextrocetirizine on the transport of tetraethylammonium in chinese hamster ovary cells transfected with the human organic cation transporter OCT2

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Background: Recently a pharmacokinetic interaction between cetirizine and pilsicainide was reported (Tsuruoka S, et al, CPET 2006 ; 79(4) :389–96) and explained as a consequence of competition between the two drugs for renal excretion mediated by the transporter proteins organic cation transporter 2 (hOCT2) and P-glycoprotein. The effect of levocetirizine and dextrocetirizine, the eutomer and the distomer of cetirizine, respectively, on the transport of the hOCT2 substrate tetraethylammonium (TEA), was investigated *in vitro* in chinese hamster ovary (CHO) cells stably transfected with hOCT2.

Methods: CHOhOCT2 cells were seeded in 12 well plates and grown to confluence. Once confluent, transport experiments were conducted. Transport buffer containing [^{3}H]TEA, and levocetirizine, dextrocetirizine or positive control (cimetidine) were added to the wells. At intervals, the transport buffer was removed, and each well rinsed three times with 1 mL of ice cold buffer to stop transport. Cells were solubilized and aliquots removed for scintillation counting.

Results: Levocetirizine and dextrocetirizine inhibited TEA uptake in CHOhOCT2 cells with IC50 values of 197 and 714 μM, respectively. In comparison, the positive control cimetidine inhibited TEA uptake with an IC_{50} of 26μM.

Discussion: Levocetirizine is a weak inhibitor of hOCT2. The IC_{50} value for inhibition of tetraethylammonium transport by levocetirizine is more than 300 fold the C_{max} (0.66 μM) at steady state following the therapeutic dose of 5 mg. It is unlikely that levocetirizine can cause drug interactions by interference with renal elimination through inhibition of renal OCT2. As dextrocetirizine is a weaker inhibitor of hOCT2 than levocetirizine, the racemate cetirizine is also unlikely to cause drug interactions through inhibition of hOCT2.

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Expression of glucocorticoid receptor-β in glucocorticoid-resistant allergic rhinitis

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Background: Glucocorticoid (GC) has been commonly used as an anti-inflammatory reagent in the treatment of chronic allergic diseases including allergic rhinitis. The effects of GC are mediated by glucocorticoid receptor-α (GR-α). Upon binding GC, activated GR-α can not only enhance transcription of anti-inflammatory genes but also interact with other protein regulating inflammation, such as nuclear factor-κB (NF-κB). However, these clinical benefits are sometimes limited because some patients demonstrate persistent tissue inflammation despite treatment with high doses of GC. It is generally considered that the interference of non-functional GR variants in immune cells may result in GC-resistance. GR-β is a well-known natural spliced variant consisting of 742 amino acids including exon 2-8 and part of 9β as coding region, but cannot bind to GC. It has been reported that there was an increase of GR-β expression in patients with bronchial asthma and in ulcerative colitis patients who did not respond to GC administration. We have tested this hypothesis by investigating correlation between the expression of GR-α, GR-β, and NF-κB proteins in patients with allergic rhinitis and the responsiveness to GC treatment.

Methods: The patient group consisted of 20 subjects with allergic rhinitis showed persistent GC-resistance, resulting in a required surgical removal after the GC treatments for over 6 months. As normal control, nasal tissues were obtained from 10 subjects underwent maxillofacial surgery. They had not been exposed to GC treatment. We have performed immunohistochemical analysis to detect GR-α, GR-β and NFκ-B proteins in nasal tissues.

Results and Conclusion: Compared to normal subjects, whereas the number of GR-α-positive inflammatory cells was decreased, the number of GR-β-positive cells was significantly increased in nasal tissues from patient group. The number of NF-κB-positive cells was at a similar level both patients and normal control. In conclusion, our data suggests that increased over expression of the GR-β relative to GR-α is associated with GC-resistance.

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Effect of azelastine on substance P release into nasal lavage from non-allergic rhinitis patients

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Introduction: Non-allergic rhinitis is a condition which affects 30 to 50% of patients with perennial nasal symptoms, occurring either alone or in conjunction with allergic rhinitis. Neural mechanisms may have a role in this condition. This study assesses the impact of intranasal azelastine on the concentration of substance P (SP) in nasal lavage fluid.

Methods: 28 patients were defined as having non-allergic rhinitis by history, examination in addition to the presence of negative skin prick tests using common inhalant allergens and low total IgE using the Phadia CAP. 16/28 patients were treated with intranasal azelastine two sprays twice daily for 10 days. The control group consisted of 12 patients with non-allergic rhinitis who used a saline nasal spray, 2 sprays twice a day. Nasal lavages were performed before and after a 10 day treatment period with intranasal azelastine or control saline nasal spray. The concentration of SP in nasal lavage fluid was determined by an EIA (Assay Designs Inc., USA) method. All patients recorded their nasal symptoms daily, including rhinorrhea, sneezing, pruritus, and congestion, using a Visual Rating Scale (VRS).

Results: The baseline concentrations of SP in the nasal lavage fluid were similar in the azelastine and the placebo group (86.8 ± 18.2 pg/mL versus 82.3 ± 21.4 pg/mL). Significantly lower concentrations of substance P were noted in nasal lavage fluid after 10 days of azelastine treatment (73.2 ± 16.9 pg/mL) when compared with placebo treatment (83.1 ± 17.8 pg/mL). The differences between the azelastine and the placebo treated group VRS symptom scores (6.4 ± 2.4 versus 8.6 ± 3.2) were also statistically significant demonstrating that the reduction in nasal lavage SP levels occurred in conjunction with improvement in clinical symptoms.

Conclusion: Intranasal azelastine reduces substance P release into nasal lavage fluid of non-allergic rhinitis patients associated with clinical improvement during a 10 day treatment period. The efficacy of azelastine in perennial non-allergic rhinitis may be related to reduction of release of SP and other non-adrenergic non-cholinergic neural mediators into nasal secretions.

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A double blind placebo controlled study on the clinical efficacy and in vivo pharmacodynamics of potassium humate in the treatment of hayfever in patients with inhalant allergies

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Humic acids are macrocolloidal molecules resulting from the decomposition of plants and are natural components of drinking water, peat, soil and brown coal. The beneficial effects of humic substances have been partly ascribed to the ability of humic acids to absorb or chelate toxic compounds or metals and to absorb xenobiotics mutagens and mycotoxins.

Several mechanistic studies were done on potassium humate, derived from bituminous coal, during the last few years. It was established that this product stimulates lymphocyte proliferation by an increased production of the growth factor, IL2.

However, no in vivo studies have been done on the anti-inflammatory effects of humate derived from coal in humans. It was proven that humic acid s extracted from brown coal have no toxic or teratogenic effects.

Purpose: The aim of this study is to investigate potassium humate's anti-inflammatory properties in patients suffering from exacerbations of hayfever during the grass pollen season in South Africa, using clinical symptoms and signs as well as establishing changes in inflammatory markers using established and new laboratory techniques.

A total symptom score over a 12 hour period and a quality of life questionnaire were evaluated.

Potassium humate (1.8g in daily divided doses) was randomly assigned to 20 atopic patients presenting with acute symptoms of hayfever. 20 Patients received a placebo resembling the humate. Treatment period was one month, preceded by a one week run-in period.

Symptoms were scored on a scale of one to four for nasal and non-nasal symptoms.

A global clinical impression was scored by the doctor at end of study: Measurement of surface area of skin prick test was done at baseline and again at the end of the study. It was restricted to the inhalant that indicated the biggest reaction.

Results: No differences in the presence and improvement of symptoms were detected between the 2 groups. Results of the quality of life assessment were also the same for the 2 groups.

Flare size comparison showed some promising results with a significant difference ($p < 0.05$) observed when comparing the before and after results of the treated versus placebo groups respectively.

Conclusion: Potassium humate shows promise in the treatment of hayfever or allergic rhinitis. The study was also conducted on inflammatory markers and showed positive results, the results presented elsewhere. The duration of the study, one month, might have influenced the results.

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Evidence that nasal mucosal hyperreactivity in healthy women is induced by high levels of estrogen

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Background: Pregnancy rhinitis, a common condition, is thought to affect 18–30 % of pregnant women. It is very annoying for many women, may develop at any time during pregnancy and usually disappears shortly after delivery. The cause is not known and it may be due to hormonal factors. Some data have shown that there may be a relation between high estrogen levels and nasal mucosal reactivity. The knowledge of the etiology of this condition is important since no satisfactory treatment is available. The aim of this study was to determine whether nasal mucosal reactivity and microcirculation change with various levels of estrogen in the blood, but constant levels of other hormones.

Methods: 15 women who were undergoing in vitro fertilization (IVF) were included. The examination and measurements of the nasal mucosa were done in the first part when the concentrations of estrogen in the blood are extremely low and then, when the concentrations of estrogen is high. The nasal mucosa was studied with a combination of rhinostereometry and laser-Doppler flowmetry during challenge with histamine. The swelling of the nasal mucosa was recorded with rhinostereometry. This optical, direct, non-invasive method is designed to measure nasal mucosal swelling with a high degree of accuracy. Laser-Doppler flowmetry, a non-invasive method for studying the microcirculation, is providing continuous and instantaneous measurements of nasal mucosal blood flow. The combination of rhinostereometry and Laser-Doppler flowmetry has the advantage of using two non-invasive methods which permit direct and simultaneous measurements of congestion and the microcirculation.

Results: With rhinostereometry we found an increase in nasal mucosal swelling after histamine challenge when the estrogen levels in blood were high. With laser-Doppler flowmetry the increases of the microcirculatory parameters velocity of moving blood cells and perfusion were lower when the estrogen levels in blood were high.

Conclusion: High levels of estrogen in blood during IVF treatment of healthy women may induce nasal hyperreactivity and changes in microcirculation. This suggests that estrogen plays a role in pregnancy rhinitis.

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Effect of a Lactobacillus paracasei on grass pollen allergic rhinitis

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Background: *Lactobacillus paracasei* strain NCC2461 has been found to exert anti-allergic effects in animal models.

Aim: the aim of the present study was to investigate the effect of NCC2461 in patients with allergic rhinitis to grass pollen.

Methods: 31 subjects (18-35 years-old) were enrolled in a randomized double-blind, placebo-controlled cross-over study. The study consisted of a first 4 weeks period in which subjects consumed either fermented milk containing NCC2461 or placebo (acidified milk drink), a washout period of six to eight weeks and a final 4 weeks period cross-over of the first treatment phase. The entire study was performed out of the pollen season.

Clinical symptoms were analysed after a nasal provocation test using grass pollen allergens and immunological parameters (specific immunoglobulins in serum, percentage of eosinophils in nasal washes and cytokines secreted by restimulated PBMC) were compared between the two treatment periods.

Results: A trend to a decrease of nasal itching favouring probiotic treatment was noticed, and a significant improvement of the nasal blockage favouring probiotic treatment was observed. However, no significant change of the nasal reaction threshold was observed between the end of the first and second treatment periods, whatever the sequence (active product before or after placebo) of product consumption. Immunological parameters' analyses are under investigation. No side effects were reported for both groups during the study period.

Conclusion: A decrease of nasal blockage was observed in allergic patients nasally challenged with grass pollens, after NCC2461 consumption. The ongoing analysis of immunological parameters should help understanding the mechanisms leading to the mitigation of respiratory allergy symptoms.

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Fluticasone furoate nasal spray demonstrates consistent efficacy against both the nasal and ocular symptoms of seasonal allergic rhinitis

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Background: Fluticasone furoate (FF) is a novel enhanced-affinity glucocorticoid that has been developed for topical respiratory use. Data from four randomised studies are presented to demonstrate the consistent nasal and ocular efficacy of FF nasal spray (FFNS) in adolescent and adult patients with seasonal allergic rhinitis (SAR).

Methods: Patients aged ≥12 years with confirmed SAR were enrolled in the four studies (n = 1141, combined total) and received once-daily double-blind treatment with FFNS 110 μg (n = 571, combined total) or vehicle placebo spray (n = 570, combined total) for 2 weeks. Individual nasal and ocular symptoms were scored by patients each on a 4-point categorical scale (0 = none to 3 = severe) each morning and evening. Nasal and ocular efficacy was evaluated by the mean change from baseline over the 2-week treatment period in daily reflective Total Nasal Symptom Score (rTNSS; average of AM and PM score totals for nasal congestion, nasal itching, rhinorrhoea, and sneezing) and daily reflective Total Ocular Symptom Score (rTOSS; average of AM and PM score totals for eye itching/burning, tearing/watering, and redness). The AM pre-dose instantaneous Total Nasal Symptom Score (iTNSS) and Total Ocular Symptom Score (iTOSS) were also assessed as a measure of 24-hour symptom control.

Results: Significant and consistently greater improvements in rTNSS were seen with FFNS than with placebo across all four studies (least square [LS] mean difference: −2.012, −0.777, −1.757, −1.480; P≤0.003). In addition, ocular efficacy was demonstrated consistently across studies, with significantly greater improvements in rTOSS in FFNS recipients than in placebo recipients (LS mean difference: −0.736, −0.546, −0.741, −0.600; P=0.008). FFNS showed sustained efficacy against both nasal and ocular symptoms over 24 hours, based on the AM pre-dose iTNSS (LS mean difference vs placebo: −1.885, −0.902, −1.898, −1.370; P<0.001) and iTOSS (LS mean difference vs placebo: −0.734, −0.519, −0.764, −0.560; P≤0.009). FFNS was well tolerated in all four studies.

Conclusion: FFNS once daily consistently demonstrates significant and sustained efficacy against both the nasal and ocular symptoms of SAR in adults and adolescents in prospective analyses performed over a programme of four clinical studies.

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Favourable safety and tolerability with fluticasone furoate nasal spray in children with allergic rhinitis

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Background: The safety profile of the novel enhanced-affinity glucocorticoid fluticasone furoate (FF) administered using a unique, side-actuated device has been investigated in children with perennial (PAR) and/or seasonal allergic rhinitis (SAR).

Methods: Safety was evaluated in an integrated analysis of data from three randomised, double-blind, parallel-group studies in 1224 paediatric patients aged 2–11 years with SAR or PAR. Patients received once-daily FF nasal spray (FFNS) 55 μg (n = 369) or 110 μg (n = 426), or placebo (n = 429) for 2 weeks (SAR study) or 6 or 12 weeks (PAR studies). Assessments included evaluation of adverse events (AEs), clinical laboratory tests, nasal examinations, ophthalmic examinations (12 week PAR study only) and electrocardiograms (ECGs). In the 6-week study, the effects of once-daily FFNS 110 μg on 24-hour serum cortisol (SC) was assessed in a domicile setting in patients with PAR.

Results: No safety or tolerability issues were identified in paediatric patients with PAR or SAR. In the integrated analysis, the most common AEs with an incidence of >3% and more common in FFNS than in placebo were headache, nasopharyngitis, epistaxis, pyrexia and pharyngolaryngeal pain. The incidences of drug-related AEs were similar across the FFNS 55 μg and 110 μg treatment and placebo groups: epistaxis was reported in 4%, 2% and 3% of patients, respectively, and headache in 2%, <1% and 1% of patients, respectively. Data from the 6-week PAR study demonstrate that SC levels following administration of FFNS 110 μg were similar to those following placebo (least squared means, 0.94 vs 0.97; treatment ratio, 0.97; 95% confidence interval, 0.88, 1.07) which suggests that FFNS 110 μg is not associated with any effect on the HPA axis. In addition, plasma levels of FF were non-quantifiable.

Conclusion: Once-daily FFNS 55 μg or 110 μg has a favourable safety and tolerability profile in children aged 2–11 years with PAR or SAR. Furthermore, FFNS shows low systemic exposure and is not associated with hypothalamic-pituitary-adrenal axis suppression in children aged 2–11 years with PAR.

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Long-term safety of fluticasone furoate nasal spray 110μg once daily in adults and adolescents with perennial allergic rhinitis

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Background: Fluticasone furoate is a novel enhanced-affinity glucocorticoid, with unique pharmacological properties suitable for topical use in rhinitis. This study investigated the long-term safety and tolerability of fluticasone furoate nasal spray (FFNS) over 12 months in adults and adolescents with perennial allergic rhinitis (PAR).

Methods: Following a 7- to 14-day screening period, patients aged ≥12 years with PAR were randomised in a 3:1 ratio to double-blind treatment with once-daily FFNS 110μg (n = 605) or vehicle placebo nasal spray (n = 201) for 12 months. Adverse event (AE) data were collected using diary cards and interviews at each study visit; the investigator graded AE severity as mild, moderate or severe, and assessed the relationship of each AE to the administration of study treatment. Safety was assessed by 24-hour urinary cortisol excretion, nasal and ophthalmic examinations, ECGs and clinical laboratory testing. Plasma levels of FF were determined from blood samples. Compliance was measured using diary cards, change in bottle weights, and symptom scores.

Results: 592 patients (73%) completed the study. Demographic characteristics were similar in treatment and placebo groups. FFNS was well tolerated: the incidence of most AEs in the FFNS group was similar to that in the placebo group, with the exception of epistaxis (defined as any observation of blood in or from the nose, irrespective of quantity), which was more frequent in FFNS than placebo recipients (20% vs 8%). There were no other clinically meaningful differences between FFNS and placebo in terms of safety assessments, including 24-hour urine cortisol excretion, mean ophthalmic parameters, ECGs and clinical laboratory tests. Plasma levels of FF were not quantifiable in the majority of patients following administration of FFNS. Compliance with study treatment was high based on diary cards (85% of patients had >90% compliance) and bottle weights. Over the treatment period, improvements in reflective Total Nasal Symptom Score were greater with FFNS (−3.37) than with placebo (−2.49).

Conclusion: Long-term (12-month) administration of FFNS 110μg once daily in adult and adolescent patients with PAR revealed a safety profile typical of intranasal corticosteroids as a class, with no evidence of clinically relevant systemic corticosteroid exposure.

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The prevalence of allergic rhinitis in college students at Kenya Medical Training College-Nairobi,Kenya

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Background: Allergic rhinitis is one of the commonest atopic diseases world wide yet its epidemiology in Kenya remains largely unknown. Currently, there is only one questionnaire based study (International Study of Asthma and Allergies-ISAAC) in children documented in Kenya.
Objectives: The primary objective was to determine the prevalence of allergic rhinitis in Kenya Medical Training College students, aged 18–50 years. The other objectives were; to determine the severity, pattern of symptomatology and the common aeroallergens involved in the study group.
Methods: The study was done in two steps. In stage 1, using a stratified random sampling, 423 students were screened for symptoms of allergic rhinitis based on International Consesus Report definition of rhinitis. In stage 2, the positive responders (63 students) were subjected to a physical examination and a skin prick test to confirm presence of allergic rhinitis.
Results: A point prevalence rate of 13% was found with no sex or age predilection in the study group. The average age of onset was 15.2 years,seasonal peaks were in January, July and December. 81.8% of the students with allergic rhinitis had their daily activity affected to a certain degree. Sneezing (83.6%) was the commonest smptom and hypertrophied inferior turbinates (70.9%) the commonest physical finding. Patients with intermittent disease (73%) were the most, while 36% of the students with allergic rhinitis had a family history of atopy. The commonest aeroallergen was the house dust mite (76.4%) and the least was Aspergillus Niger (1.8%).
Conclusion: Allergic rhinitis affects a significant proportion of the college students and has symptoms which have an impact on the lifestyles of these patients. The common aeroallergens are found within our immediate surroundings e.g, house dust mite, which can be controlled if patients are educated and proper, cheap enviromental control measures are instituted.

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Inflammatory mediators level in nasal polyposis

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Background: Nasal polyposis (NP) often coexists with asthma and rhinitis. Polyp histology typically shows chronic, eosinophilic inflammation including eosinophils, lymphocytes, plasma cells and mast cells. We studied mediator levels and leukocyte values in nasal fluids (NFs) and eosinophil cationic protein (ECP), total IgE levels and eosinophils in the blood in both allergic and non-allergic patients with NP and in patients with allergic rhinitis (AR).
Methods: Forty-six patients with NP and 37 patients with AR as a control group: 15 patients with seasonal AR to grass pollen, 12 with AR sensitive to Parietaria and 10 with AR sensitive to house dust mite (HDM) entered the study. Twenty-one patients with NP were also allergic patients (11 were sensitive to Parietaria and 10 - to HDM), whereas 25 were non-allergic patients. Tryptase and histamine values were assayed in NFs, total IgE was determined in serum. ECP values were assayed both in NF and serum. Eosinophils were quantified both in the blood and NFs.
Results: Tryptase levels were significantly higher in the NFs from patients with NP than in those from patients without NP (3.9 vs. 3.5 U/l, $p < 0.001$) and correlated with symptom scores ($r(s) = 0.36$, $p < 0.0001$). The median levels of histamine in NFs from patients with NP were significantly higher than those in patients without NP (40.0 vs.19.4 ng/ml, $p < 0.001$), but did not correlate with symptom scores. The median levels of ECP in NFs from patients with NP were significantly higher than those in patients without NP (37.4 vs. 17.2 ng/ml, $p < 0.001$) and correlated with symptom scores ($r(s) = 0.33$, $p < 0.001$). With regard to leukocyte counts in NFs, no significant differences were between rhinitis patients with NP and those without NP. With regard to serum ECP and serum total IgE, no significant differences were detected between the two groups. Blood eosinophil levels in patients with NP were significantly higher than those in patients without NP (5.7 vs. 5.5, $p = 0.002$).
Conclusion: Chronic eosinophil mucosal inflammatory disease in NP involves a self-sustaining mechanism independent of allergen stimulation of nasal mucosa. Increased release of inflammatory mediators contributes to the development of NP, determining oedema and an increased recruitment of inflammatory cells. Eosinophils, mast cells also play a key role in this process.

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Physicians' compliance with international guidelines in the treatment of allergic rhinitis

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Background: Allergic rhinitis (AR) is an allergy associated with a high burden of costs. This disease is also considered an important risk factor in the development of asthma. Within a time slot of 10 years, 20–40% of rhinitis patients develop asthma. The ARIA guidelines, introduced in 2001, address this problem and make treatment recommendations for allergic rhinitis based on the concept "one airway, one disease." Several studies have investigated the implementation process of these guidelines and have stressed their suitability for daily use.
Methods: The objective of the present analysis was to estimate the compliance with and the acceptability of international guidelines among ENT specialists and general practitioners when treating patients with allergic rhinitis. For this purpose, we examined data from 122,000 patients using an IPD meta-analysis from seven post-marketing surveys collected from 1998 to 2005.
Results: First, we investigated the data pool as a whole, and the results showed that 38% of the patients who were treated by ENT specialists received therapy according to these international guidelines. In contrast, only 16.3% of the general practitioners heeded the guideline recommendations. Next, we examined the time line in general, regardless of the physician's specialty. We observed that the rate of compliance and acceptability in 2002 was higher than that in previous years. Moreover, in more than 50% of cases the patients with rhinitis and concomitant asthma were treated by their ENT specialists according to the ARIA guidelines.
Conclusion: The results are evidence of the well-structured and successful implementation process of the ARIA guidelines. ENT specialists could apply the recommendations in practice more easily as compared to other guidelines. However, the investigation also shows that the ARIA document has not yet found its way into the daily routine of general practitioners. This finding thus supports the goals set by the "International Primary Care Respiratory Group" for implementing special guidelines for general practitioners in the primary care setting.

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Inhibition of the nasal reaction by second-generation antihistamines in patients with Japanese cedar pollinosis in an artificial pollen exposure chamber (OHIO chamber)

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Background: Pollinosis is seasonal allergic rhinitis due to pollen antigens, and its prevalence is high enough to be called a national disease in Japan. Among the many pollen antigens, Japanese cedar pollinosis is the most common. A pollen exposure chamber (OHIO Chamber) was built in central Tokyo, Japan, in order to study seasonal allergic rhinitis (SAR). Epinastine hydrochloride (epinastine), the second-generation antihistamines, is largely used in the indication of allergic rhinitis in Japan. The purpose of this study was to investigate the protective efficacy of epinastine in patients with Japanese cedar pollinosis in an artificial pollen exposure chamber (OHIO Chamber).
Methods: The study was designed as a double-blind study. After preliminary study, 20 volunteers were initially exposed to a low concentration (4500 grains/m3) of JC for at most 1 hour in this chamber, volunteers were randomized into 2 groups (group A and group B) and allocated to receive

either epinastine 20 mg tablets or a placebo tablets once a day for 2 weeks. At the end of 1 week interventional period, volunteers were exposed to a low concentration (4500 grains/m3) of JC for 2 hours. And then at the end of another 1 week interventional period, volunteers were exposed to a high concentration (6000 grains/m3) of JC for 2 hours again. Subjective nasal and ocular symptoms were recorded at 0, 15, 30, 60, 90, and 120 minutes using personal computer systems and the amount of nasal secretion was measured during the allergen exposure periods.
Results: Total symptom scores (TSS) in epinastine group was significantly lower than in placebo group during low concentration of JC exposed. However during high concentration of JC exposed, significant differences in TSS were not observed.
Conclusion: This is the first clinical study using Japanese cedar pollen under well-controlled conditions in the OHIO chamber. This study showed that 20mg of epinastine once daily reduced the severity of allergic symptoms compared with that once in a week or placebo in pollen season.

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Development of a next-generation delivery system for allergic rhinitis: fluticasone furoate nasal spray

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Background: Patients with allergic rhinitis (AR) consider ease of use to be the most important feature of prescription nasal sprays, with formulation-related attributes (e.g. medication that runs down the throat/out the nose and bitter taste) being key barriers to continued use. Thus, improved delivery systems and formulations may help improve adherence to nasal spray treatment.
Methods: The fluticasone Furoate nasal spray (FFNS) device was designed for optimal ergonomics. Following a review of published data, key dimensions and operational ranges required for device operation were determined, and device concepts were modelled and evaluated by user groups, whose feedback was used to refine and finalise the design. The reliability of device was tested *in vitro*, and its ease of use was assessed in a Phase III clinical study in 302 patients with AR. The delivery system was developed with minimal steps for use, to be suitable for use by a wide patient population, for easier third-party administration and to allow determination of medication remaining in the pack.
Results: *In vitro* tests confirmed that the FFNS device is robust, operates reliably when a force of 25–45N is applied to the side lever, and delivers a consistent dose, even when left unused for 6 weeks. A low-dose volume (50 μL) is delivered as a fine mist which consistently delivers 27.5 μg/spray and minimises the amount of formulation available to run down the back of the throat or drip from the nostrils. In a Phase III clinical study, 84% of 302 patients found the device easy to use, 95% found it easy to carry, and 97% found the drug product comfortable to use. In addition, a review by an independent ergonomist concluded that the FFNS device is more comfortable to hold and easier to operate than current "top-down" nasal spray devices and is suitable for use in children as young as 2 years. The unique side-actuated delivery mechanism also allows for easier third-party administration.
Conclusion: The novel FFNS device is easy to use, delivers a consistent dose, and addresses patient-reported barriers to the use of existing nasal sprays.

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Subsequent development of asthma in allergic rhinitis: a questionnaire based study

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Background: Subsequent development of bronchial asthma (BA) in allergic rhinitis (AR) patients is common. We investigated the clinical aspects of subsequent development of BA in AR patients by a questionnaire based study.
Methods: Total 227 patients, visited us for AR without BA (2001~2004), were enrolled in 2006. We reviewed their clinical data and followed them up with a BA-detecting questionnaire, based on International Primary Care Respiratory Group (IPCRG) guidelines 2005. These results were statistically analyzed.
Results: Among 227 (215 loss to f/u, 12 regular visitors), 91 responded to the questionnaire (85 by phone, 6 via mail). 37 responders, who answered yes to either doctor-diagnosed asthma or any of 4 pivotal IPCRG questions, were suspected as the group with subsequent development of BA (BAS). BAS showed higher female rate (73%) than non-suspected group (NBAS, 54%). In BAS, males were younger and females had higher BMI than NBAS ($p < 0.01$, respectively). Familial allergic diseases (36.8%) and nasal polyp/surgery were more frequent in BAS. BAS had longer AR duration (10.7±10.1 yrs) than NBAS (6.3 ± 4.5 yrs). According to the increase of AR duration, BAS (%) increased linearly ($p = 0.012$). Current AR symptoms were more prevalent in BAS (89.2%, NBAS 64.8%). Atopic tendency, skin test results, serum total IgE, and induced sputum eosinophil (%) were not different between both groups. In pulmonary function test, BAS males showed lower initial FEV1 (L) and FVC (L) ($p < 0.05$, respectively). In initial methacholine challenge test (M-test), airway hyperreactivity (AHR: PC20 < 25 mg/mL) were more common in BAS (93.7%, NBAS 33.3%).
Conclusion: 40.7% of AR patients (mean duration 8.0¡¾7.5 years) were suspected to develop subsequent BA. Such progression might be suggested by female gender, younger age in male, higher BMI in female, longer AR duration, and more frequent AHR positive results in initial M-test. AR patients without lower respiratory symptom should be followed up regularly for early diagnosis of subsequent BA.

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Possible roll of the nitric oxide as an immune marker for the diagnosis of respiratory pathologies in Venezuelan scholar children

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Background: It has been proposed a clear association between exhaled nitric oxide (NO) and asthma exacerbation. We evaluated the NO in nasal mucus and sera, from 144 Venezuelan scholar children (6–12 years old), from low to middle socioeconomic background covering different areas from Caracas, Venezuela, with allergic rhinitis and bronquial asthma.
Methods: All children were evaluated with a validated modified Graffar's socioeconomic questionnaire and an allergic rhinitis, asthma and atopic dermatitis after ARIA, GINA and Hannifin criteria; skin prick testing (ALK-ABELLO) for common food and inhalant allergens plus total IgE (ELISA), complete blood count (COULTER) and serial feces examination for ova and parasites were performed in all children. Pre and post bronchodilator FEV1 and PEF spirometric measurements values were obtained (MICROLOOP) (we obtained the approval by the ethical committee of the Institute of Biomedicine, ratified by the academic council of the medical Faculty of Central University of Venezuela, After an informed consent was signed from parents or guardians). In nasal mucus and sera from children NO were detected by a simple colorimetric method based on Griess Methodology.
Results: According to clinical evaluation children's were classified in four group. Healthy children's, children's with rhinitis and without asthma, children's with asthma and without rhinitis, and children's with asthma plus rhinitis. Healthy children present lower NO value both in sera (X = 34,48μM) and nasal mucus (X = 7,99μM). Rhinitis children had the higher NO levels both in sera than in nasal mucus, finding statistical differences in nasal mucus levels (21.23μM) compare to control group (7,99μM). In asthmatic children NO levels were lower than in rhinitis once, but higher than healthy control group.
Conclusion: In nasal mucus, the study of immune parameter as nitric oxide by a rapid and simple methodology, could clarify the local inflammatory process

related to allergic rhinitis in order to bring better strategies for the diagnosis and also for the evolution of the treatment.

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Inducement of eosinophils to apoptosis by injection of steroid into nasal polyp

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Background: It is wellknown that the basic protein of eosinophils induce respiratory epitherial disorders. The eosinophils which migrate to nasal mucosa do not return again into the circulating blood stream. It has been reported that eosinophils into nasal mucosa and mucus were degranulated by cytolysis. It is preferable treatment of allergy that the eosinophils are promptly excluded nasal mucosa without degranulation. It is aspired that the eosinophils induced to apoptosis not but necrosis.

Methods: After the polyp was taken from the right nostril of the patient in whose mucus many eosinophils were observed, corticosteroid was injected into the polyp of left nostril. Two days after the steroid injecion, the polyp of the left nostril was taken. The both side polyps were fixed with 2% glutaraldehyde for 1 hour and postfixed with 2% osmic acid for 40 minutes and embedded in Epon 812. Ultrathin cross sections were prepared and observed under transmission electron microscope. The total numbers of eosinophils and macrophages with and without phagocytosing eosinophils. In some ulrathin cross section, the localizaion of ss-DNA was examined which was a mark of apoptosis.

Results: The nucleus of eosinophils showed typical characteristics of apoptpsis but apoptoic bodies were not observed after the steroid injection. The ss-DNA was admitted in the part of heterochromatin of the nucleus, which proved apoptosis had been caused. It has been known that the cells are phagocytosed by macrophages as soon as they were induced to apoptosis. There was not a significant difference in the number of total eosinophils and macrophages in the polyp before and after the steroid administration, but the rate of eosinophil phagocytosis by macrophage significantly increased after the steroid administration.

Conclusion: It was clalified that eosinophils were induced to apoptosis after the steroid administration by the feature of nucleus with electron microscope and proof of ss-DNA. Eosinophils which had induced to apoptosis were promptly phagocytosed by macrophages.

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Acoustic Rhinometry in children with allergic rhinitis

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Acoustic Rinometry (AR) is a sound-based techinique by which the reflected wave provides information about volume & area of the nasal cavity. By AR the explorer has the abilitiy to assess nasal patency in patients suffering nasal obstruction.

Goal of Study: To study basal figures of endonasal measurements in subjects with allergic rhinitis by dust mites during winter season in Canary Islands (subtropical climate with high humidity (65–80%) and mild temperatures −20–25°C-) and to compare with subjects without obstruction.

Methods: Patients with clinical history of Rhinitis were assessed. Skin Prick Test (SPT) with mites and German cockroach were performed. Acoustic Rinometry was performed with Rinometer RhinoScan with SER 2000 module (Denmark). Three measurements were made for each nose. The mean area distance curve was calculated for each side of the nose. Curves with artifacts were discarded. Patients without blockage have a normal Minimal Cross-Sectional Area (MCA) at C-notch with values around 0.7 cm2

Results: A clinical history of rhinitis and positive SPT with mites was seen in 82% of patients. In most, the history was a mild/severe persistent rhinitis. In 17% of patients SPT were negative. In most of patients 86% of patients MCA was located in first notch (I-notch), which corresponds with Itsmus nasi and the remainder MCA was located in second notch (C-notch), which corresponds to the head of the inferior turbinate. In all the study, the average distance calculated from nostril to the head of inferior turbinate was 22.0 mm. Average of MCA was 0.54 cm2 in allergic patients and 0.57 cm2 in non-allergic rhinitis patients with no significant differences between groups Conclusion Measurements by AR are similar between allergic and non-allergic rhinitis patients. In most of patients MCA was located in first notch (I-notch), which corresponds with Itsmus nasi AR has major advantages of over other methods for assessing nasal patency since it is a very simple method and requires minimal cooperation.

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Clinical and therapeutic aspects of moderate/severe allergic rhinitis in Transylvania

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Background: Moderate/severe allergic rhinitis (MSAR) is an affection of nasal mucosa induced by allergen exposure and produced by a specific IgE mediated chronic inflammation. Symptoms are present in over 4 days in a week, and over 4 weeks (ARIA guide), and the symptoms interfering with activities and sleep. It is related that MSAR has a 10–25% incidence from all allergic rhinitis, it is more frequent in adolescent and young people (who, usually don't have the financial resources for the therapy: topical corticosteroids, immunotherapy). SMAR patients have a reduce quality of life, and by absenteeism and costs this seems to be an important health problem.

Methods: The aim of this study is to evaluate 40 patients with SMAR that last over a year, for 8 months (age between 9 and 57 years, medium age 33.85). Our patients have completed questionnaires about their nasal symptoms and the effect of the treatment (antihistamines, topical corticosteroids and immunotherapy. They were evaluated by skin prick tests to 10 inhaled allergens, rhinoscopy for nasal mucosa and questionnaires for the symptoms score.

Results: The majority of MSAR have a polisensitization (80%: to dust mites 90.62%, to cat 37.5%, to pollen 37.5%, to cockroach 37.5%, to moulds 37.5%, to dog 18.75%). Only 20% from our patients have monosensitization (to house dust mites). The score of the symptoms was high- over 4 on a day in 4 days from the last 7 days- we evaluated the nasal congestion, rhinoreea, nasal itching and sneeze by 0 to 3. Antihystamines therapy has a lower efficiency- to one patient (3.12%) - in polisensitization patients, and in those with severe congestion has practically no effect, in 26 patients none feels better. The only therapeutic method with higher efficiency was topical corticosteroids, witch make an obvious amelioration, the score of the symptoms decreased between 0 and 2 on a day in 7 days. The financial aspect of the patients with MSAR showed that only a few from them could provide the topical corticosteroid for at least 3 months (12.5%-5 patients).

Conclusion: MSAR is hard to be treated and controlled without topical corticosteroids.

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The effects of olopatadine hydrochloride on rhinitis induced by intranasal instillation of toluene-2,4-diisocyanate in rats

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Background: The main symptoms of allergic rhinitis are sneezing, rhinorrhea and nasal obstruction. In patients with AR, the levels of neurotrophin nerve growth factor (NGF) and neuropeptides substance P (SP) increased. Therefore,

they are considered important modulators in the development of AR. Olopatadine hydrochloride (olopatadine), is an anti-allergic agent with histamine H1 receptor antagonistic action. We reported that olopatadine inhibited the elevated levels of NGF and SP in the mouse model of chronic inflammatory dermatitis.
Objective: To investigate whether olopatadine has an effect on the production of NGF and SP, we used TDI-sensitized rats as an animal model of nasal allergy.
Methods: After the intranasal challenge of TDI, the numbers of sneezes were counted in a blinded way. To determine NGF and SP production in the nasal lavage fluids (NALF), rats were anesthetized and nasal lavages were done. Olopatadine was orally administered orally before the nasal instillation of TDI.
Results: In TDI-challenged rats, nasal allergy-like behavior (sneezing, rhinorrhea and inflammation) was provoked after TDI challenge. The amounts of NGF and SP in the NALF were increased. Olopatadine reduced nasal allergy-like behavior. Moreover olopatadine inhibited the increases of NGF and SP production.
Conclusion: Our findings suggest that the increase of NGF and SP production is one of the mechanisms responsible for nasal allergy-like behavior in TDI-challenged rats. These results suggest that the suppression of neurogenic inflammatory reaction might partially be involved in the improvement of allergy-like behavior by the treatment of olopatadine.

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Do topical steroids reduce subjective and objective measures of nasal congestion in persistent allergic rhinitis?

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Background: Nasal breathing is essential in maintaining the physiologic functions of the upper and lower airways. The predominant symptom of allergic rhinitis is nasal congestion, which also has a significant impact on quality of life and work productivity. In patients who suffer from persistent allergic rhinitis (PAR), a severe drug-resistant hypertrophy and increase in glandular structures of the inferior turbinates may develop, which leads to constant nasal obstruction. Objective methods are strongly recommended for use in the evaluation of pharmacologic agents that are expected to improve nasal airflow.
Methods: This prospective, single-sited study randomized 50 patients with mild or moderate PAR who had substantial bilateral hypertrophy of the inferior turbinates to desloratadine (5 mg/day) or additional mometasone furoate monohydrate nasal spray (MFMNS), (2 sprays per nostril [total dose 200 μg] once daily) treatment groups. Patients with previous treatments, concomitant sinonasal disorders or systemic diseases were excluded. Both objective outcomes evaluated by total nasal resistance at anterior rhinomanometry and subjective outcomes assessed with endoscopic nasal examination and Quality of Life Questionnaire were analyzed before and at least 12 months after treatment.
Results: The median total nasal resistance in patients treated with MFMNS decreased from 0,49±0,17 Pa/cm3/s to 0,39 Pa/cm3/s (p = 0,42), and at least 12 months later. Compared with pretreatment scores, the post treatment scores of these patients significantly improved in both 7 separate domain scores and overall Rhinoconjunctivitis Quality of Life Questionnaire scores (p = 0,004). Nasal symptomatology was reduced 2 months after MFMNS application. No adverse reactions including bleeding, infection, or hormonal disorders were encountered. The patients experienced a lasting benefit from this treatment.
Conclusion: Nasal congestion affects most individuals with allergic rhinitis, and has a notable impact on quality of life, emotional function, productivity, and the ability to perform daily activities. These results suggest that topical mometasone reduces the volume of inferior turbinate at some point while significantly improving the quality of life in patients with PAR. Histopathologic and longer term studies with larger groups will enlighten the potential and mechanism of efficacy of topical steroid in management of inferior turbinate hypertrophy in patients with PAR.

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Topical treatment for perennial allergic rhinitis in children

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Background: Topical corticosteroid is now accepted as safe and most effective in controlling all symptoms of both allergic and nonallergic rhinitis. Mometasone furoate monohydrate nasal spray is a once daily topical corticosteroid preparation.

Objective: To evaluate the efficacy and safety of mometasone furoate monohydrate nasal spray in children 6 to 12 years of age with perennial allergic rhinitis.

Methods: A double-blind, placebo-controlled, parallel group of 112 recruited patients of whom 96 were evaluated. Treatment with once daily mometasone furoate monohydrate nasal spray 100 mcg once daily or placebo for 6 weeks followed by a 3-week follow-up period. Forty-eight patients of each group were treated with mometasone furoate monohydrate nasal spray or placebo by randomized assignment.

Results: There was no statistical significance of the sex, mean age, weight, and height of the two groups. Patients treated with mometasone furoate monohydrate nasal spray showed a significant decrease in total symptom scores rated by physicians at 3 weeks and 6 weeks, respectively ($P < .01$, $P < .05$). The rhinitis symptom scores in treatment group rated by patients (nasal blockage, sneezing, watery rhinorrhea) were significantly decreased at 3 weeks ($P < .05$, $P < .01$). Nasal symptoms as assessed by doctors (turbinate swelling, color of nasal mucosa, secretion, and postnasal drip) also decreased at 3 and 6 weeks, but were not statistically significant, except for the secretion at 3 weeks and postnasal drip at 6 weeks ($P < .05$). There was no evidence of effects on adrenal function by morning plasma cortisol concentration between the two groups.

Conclusion: Mometasone furoate monohydrate nasal spray was safe and effective, well tolerated in children aged 6 to 12 years with perennial allergic rhinitis with incidences of adverse events comparable to placebo.

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Effects of seasonal allergic rhinitis on fatigue levels and mood

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Objective: Many allergy patients complain of fatigue, moodiness, and dysphoria during their allergy seasons. This study evaluated the effect of symptomatic allergic rhinitis on both fatigue level and mood.

Methods: Symptomatic ragweed allergic rhinitis patients on no medications and healthy control subjects completed the Multi-Dimensional Fatigue Inventory and the Positive Affect-Negative Affect mood rating scales in an in-out-in ragweed season research design.

Results: During ragweed seasons, allergic patients reported higher levels of general fatigue and mental fatigue, but not physical fatigue, as well as reduced motivation. Patients described experiencing feelings of greater sadness and reduced pleasurable engagement. Increased anxiety or emotional distress was not reported.

Conclusion: These findings suggest that having allergic reactions to ragweed pollen causes significant fatigue and mood changes in at least a subgroup of patients. Psychoneuroimmunology and medical genetics research suggests that allergic reactions engender biochemical changes that directly affect the central nervous system.

Key Words: allergic, rhinitis, fatigue, depression, mood

Abbreviations: CNS = central nervous system; FSL = Flinders sensitive line; IL = interleukin; MFI-20 = Multidimensional Fatigue Inventory; NA = negative affect; PA = positive affect; TNF = tumor necrosis factor.

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Patient perception of levocetirizine in allergic rhinitis: A multicenter study in Taiwan

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Background: Levocetirizine (Levo) is a potent latest-generation, non-sedating oral H1-antihistamine for which no data in Taiwanese population has yet been published.

Objectives: Primary: patients' perception of Levo in the treatment of allergic rhinitis (AR). Secondary: adverse events (AEs) and incidence of intermittent (IAR) and persistent (PER) AR (as defined by ARIA) in Taiwan.

Methods: A multicenter observational study (6 medical centers) conducted from May 2006 to March 2007 in Taiwan assessing the treatment perception of 236 AR patients on Levo. Runny nose, nasal and ocular pruritus, sneezing and nasal obstruction were measured (0 = absent, 3 = severe). Total 5 Symptom Score (T5SS, sum of the above symptoms (range 0–15)) and asthma symptoms (per GINA): mild (>1x/week, <1x/day); moderate (daily); severe (continuous) were determined. The observational period was 2–4 weeks. The onset of action was rated very rapid (<30min), rapid (>30min, <1hr), and moderate (>1hr). A visual analogue scale (VAS) was used to evaluate global satisfaction of patients and physicians (range: 0–10).

Results: 236 patients were included and 217 completed the study. 19 patients (8%) were lost to follow-up due to AEs, none serious. 56 patients (24%) had concomitant asthma. PER was diagnosed in 191 (81%) and IAR in 45 (19%) patients.

T5SS improved by 56%, from 10 (±3.3) at baseline to 4.4 (±2.3) at end of treatment. Overall efficacy and tolerability were assessed as "good/excellent" by 60% and 66% of patients, respectively. At least 61% of all patients and 75% of those with "moderate/severe" symptoms reported complete recovery or marked improvement of any individual symptom. 50% of subjects reported the onset of action as very rapid or rapid. Levo was reported as better than their previous therapy by 56% of patients. "Good/excellent" improvement in quality of sleep and daily activities was reported by

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Symptom improvements	Rhinorrhea (n = 222)	Nasal pruritus (n = 195)	Sneezing (n = 218)	Nasal congestion (n = 202)	Ocular pruritus (n = 151)	Asthma symptoms (n = 75)
Complete or partial relief	75%	72%	71%	64%	61%	44%
Unchanged	21%	23%	28%	33%	32%	55%
Worse	4%	5%	1%	3%	7%	1%

50% and 53% of patients, respectively. Global satisfaction with Levo treatment was high for both physicians and patients (VAS = 6.9). Most frequently reported AEs were somnolence (7.4%) and fatigue (3.7%).
Conclusion: Results confirm western studies that Levo is potent, effective and well tolerated in Taiwanese patients with PER and IAR. The majority of patients considered it as better than their previous treatments and the global satisfaction with Levo was consistent in both physicians and patients.

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Medicamentous therapy of allergic rhinitis

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Background: To develop algorithms of treatment of allergic rhinitis in different clinical situations and complex scheme of treatment for allergic rhinitis.
Methods: We observed patients with diagnosis of seasonal and all-the-year-round allergic rhinitis. All the patients were evaluated clinical efficacy of medicamentous treatment.
Results: Seasonal or intermittent allergic rhinitis. The treatment of the mild form of rhinitis with episodic symptoms the treatment should be begun with per oral or topic administration of antihistamine (non-sedative) drugs. Other variants of treatment are the topic decongestants (for no longer than 10 days) and per oral decongestants (which are not recommended for children). If eye symptoms are prevailed over the rhinitis symptoms or if they were not stopped with administration per orally of antihistamine preparations then the same preparations may be additionally used as eye drops. In cases of moderate severe and severe forms with episodic symptoms the treatment includes per oral antihistamine preparations with decongestants and topic glucocorticoids.

The all-the-year-round or persistent allergic rhinitis. In cases of light clinical course when the symptoms of disease do not required special treatment and only the measures for elimination of allergen may be performed. Variants of medicamentous therapy include per oral or topic antihistamine preparations, per oral antihistamine preparations with decongestants and topic glucocorticosteroids. Efficacy of therapy performed must be evaluated in 2–4 weeks.

In moderate severe and severe forms of rhinitis the preparations of the first line are the topic glucocorticosteroids. In sharp disturbance of the nasal breathing this treatment may be added with short course of systemic steroid therapy. The effect of this therapy is evaluated in 2 weeks.
Conclusion: The causes of insufficient efficacy of topic glucocorticosteroids may be: irregular dosing of preparation by physician or patients, insufficient administration of preparation into the nasal cavity because of sharp edema of mucous membrane, presence of concomitant pathology (deformation of nasal septum, chronic rhinosinusitis and others), power effect of unremoving allergen and irregularly established diagnosis.

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The impact of Flixonase on quality of life in paediatric patients with seasonal allergic rhinitis

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The aim of our study was to investigate the efficacy of Flixonase in children with seasonal allergic rhinitis and to evaluate the impact of treatment on quality of life. 38 (4–12 years old) outpatient children with seasonal allergic rhinitis were enrolled into the clinical study. We evaluated history, clinical symptom severity (nasal symptom severity – such as sneezing, runny nose, itching, stuffiness; non-nasal symptom severity – such as eye, ear, throat symptoms, chronic cough, and headache). Rhinoscopy (swelling and hyperemia of mucous) and evaluating allergic markers (eosinophyles) in nasal lavage detected diagnosis of rhinitis. We studied blood immunity data (IgE). For the assessment of the quality of life we used the Juniper Rinoconiuctivities quality of life (Pediatric and adolescent) scale. The baseline results showed that eosinophilia in peripheral blood was observed in 35%, patients, IgE hyper production in 72%. There was determined correlation link between IgE hyper production and severity of diseases and eosinophil count in nasal lavage. Flixonaze was administered 50 mcg once a day. The treatment with Flixonase (time of treatment 14 days) showed significant improvement of clinical symptoms and patients condition, reduction of nasal as well as non-nasal severity symptoms. Medication assessment average score was 6, 6 + 0.3. At the same time was observed decrease of eosinophil count in nasal lavage. After the two weeks trial period, the overall RQLQ scores of the treated patients improved by 68% + 7 from baseline.

So, we conclude that using of Flixonazein paediatric patients with seasonal allergic rhinitis improved patient's condition as well as their quality of life.

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Allergic rhinitis: especially of nasal's microflora in the children

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Background: Staphylococcus aureus is proved to play unduestionable role in development of atopic dermatitis. In this connection investigation of bacteria role in dermatitis pathogenesis is of considerable interest. The aim of the present study was to investigate microbial microflora of the nasal cavity in patients with persistent allergic rhinitis (PAR).
Methods: A total of 89 PAR sufferers aged 3 to 17 years have been investigated, bacteriological study of nasal secretion being carried out.
Results: Staphylococcus haemolyticus in diagnosticaly insignificant titer was isolated in every second person in the group of PAR patients (n = 38), Staphylococcus aureus in diagnosticaly insignificant titer being isolated in 23% of patients. High level of dissemination (more than 103–104 KOE/ml) was noted in 10 out of 38 patients: Staphylococcus haemolyticus-15,8% (n = 6); Staphylococcus aureus-10,5% (n = 4). In PAR patients along with atopic bronchial asthma (n = 42) rising Staphylococcus haemolyticus titers-24% (n = 10), Staphylococcus aureus-33% (n = 14) was observed. In 6 out of 14 patients isolated Staphylococcus a ureus strain was combined with Klebsiella, Escherichia coli, Citrobacter spp. Isolation of permissible for normal microflora values was equal to 19% for Staphylococcus haemolyticus (n = 8); and 16,7% (n = 7) for Staphylococcus aureus. Streptococcus haemolyticus was isolated in 3 patients. In the group of PAR patients along with atopic dermatitis (n = 9), Staphylococcus aureus (in 7 patients under study) was the main representative of the nasal cavity microbiocenosis.
Conclusion: Prevalence of staphylococcal found out in the course of study in nasal cavity of PAR patients may suqgest the ability of these microorganisms to support allergic inflammation.

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Efficacy topical desensitization in chronic rhinosinusitis and nasal polyposis with association nonsteroidal anti-inflammatory drugs hypersensitivity

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Background: Subject suffering from aspirin and over nonsteroidal anti-inflammatory drugs (NSAID's) hypersensitivity frequently develop chronic rhinosinusitis (CRS) with nasal polyposis (NP). Previous studies showed that

aspirin-lysine topical desensitization may be effective to CRS and NP treatment and prevent recurrent NP.

The Aim of the Study: To evaluate the efficacy of topical desensitization by sodium diclofenac in patients (SD) with NSAID's hypersensitivity and chronic rhinosinusitis with nasal polyposis.

Methods: Ten subjects (5 male and 5 female, mean age 52.5 ± 2.1 years) after intranasal polypotomy with positive results of nasal challenge with SD underwent topical SD desensitization and daily treatment with SD 20 mg during 1 year. Additionally 5 patients were receiving intranasal corticosteroids in stable dose.

Results: There were significant reductions of the total endoscope count (10.4 ± 2.8 to 5.8 ± 1.9, $p < 0.05$) and nasal symptom score (6.6 ± 2.2 to 3.2 ± 0.8, $p < 0.05$) in the subgroup with combination desensitization and topical corticosteroid therapy. In both subgroups there were improvement of nasal inspiratory peak flow (75,0 ± 13,8 l/min to 86,5 ± 9,2 l/min, $p < 0.05$) and pulmonary function. Using of inhalation corticosteroids were decreased. There were no recurrences of nasal polyposis in all patients after one year observation.

Conclusion: Topical sodium diclofenac desensitization is an effective treatment in chronic rhinosinusitis and nasal polyposis with association NSAID's-hypersensitivity.

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Influences of allergic rhinitis to the outcome of functional, endoscopic sinus surgery

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Background: The exact pathogenesis of nasal poyposis (NP) is still unknown. Empiric data show that patients who suffer from NP and allergic rhinitis (AR), do not benefit as much of functional, endoscopic sinus surgery (FESS) as patients without AR.

Methods: In this study we want to present our expirience in FESS in patients with AR. 120 patients were included and devided into three groups: 1. 45 patients with AR without NP and without FESS 2. 43 patients with AR and NP and FESS a) 32 patients with SIT b) 11 patients without SIT 3. 32 patients with NP without AR. In our protocol we included a score of symptoms, the endoscopis findings and a CCT of the sinuses.

Results: In general, patients in group 3 have a better outcome of FESS than patients in group 2. Group 2a benefits more of FESS than group 2b.

Conclusion: In this investigation, we can show that there is a tendency that patients with NP and AR who are treated by SIT have a better outcome of FESS than patients without AR.

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Expressions of mammaglobins A and B are not different between nasal polyps with and without allergic rhinitis

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Background: Nasal polyposis is a chronic disease of nose and sinuses. Its actual causes remain unclear. Mammaglobins have been implicated in the pathogenesis of nasal polyps. However, their association with the occurrence of nasal polyps in the presence of allergic rhinitis has not been explored.

Objective: The aim of this study was to compare the expression levels of *mammaglobins A* and *B* between the nasal polyps with allergic rhinitis and without allergic rhinitis.

Methods: 31 patients with bilateral nasal polyposis underwent skin prick test to specific aeroallergens. Nasal polyp tissues were obtained from all patients and divided into 2 groups as nasal polyps with allergic rhinitis and nasal polyps without allergic rhinitis depending on the skin prick tests' results. All polyp tissues were analyzed for the levels of *mammaglobin A* and *mammaglobin B* by using real-time quantitative polymerase chain reaction technique (RTQ-PCR).

Results: Of the 16 samples from patients having nasal polyps with allergic rhinitis, only one expressed a detectable level of *mammaglobin A* (1/16). There was no detectable expression of *mammaglobin A* in tissues from the group of nasal polyps without allergic rhinitis (0/15). Expression of *mammaglobin B* was detected in all nasal polyp tissues from both groups. The mean expression of *mammaglobin B* was not significantly different between nasal polyps with allergic rhinitis (0.059; range 0.0002 to 0.343) and nasal polyps without allergic rhinitis (0.133; range 0.003–0.628).

Conclusion: Expressions of *mammaglobins A* and *B* are not different between nasal polyps with and without allergic rhinitis. Our findings suggest that mammaglobins' implication in the pathogenesis of nasal polyps is independent of an underlying allergic rhinitis.

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Characteristics and associated conditions of acute sinusitis in Thai children: a prospective evaluation of 140 patients

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Backgroud: Sinusitis is a common disease in children. Even though new research findings are continually being published on various aspects of this disease, reports of its clinical characteristics and associated conditions are still limited.

Objective: To describe the characteristics and associated conditions of acute sinusitis in Thai children.

Materials and Methods: Our study consisted of 140 acute sinusitis patients who presented to Thammasat University Hospital. Acute sinusitis is diagnosed on the basis of clinical presentation including persistent, severe or worsening nasal symptoms. An informed consent form was obtained from each patient before the study. Radiography has been used to aid in the diagnosis of acute sinusitis. Allergic skin prick test was also performed in this study.

Results: Age range of the 140 patients were between 1.1 to 15 years with a mean (+/-SD) of 5.67 +/-2.85 years. History of allergic rhinitis, confirmed by positive skin prick test or nasal cytology, was found in 64%. The parental history of atopy was found in 50.4 %. Positive history of tobacco smoking was found in 25.9 %. Familial members with diagnosis of a cold or sinus infection during the same time was found in 43.2 %. 69.8% of the patients attended day care or school more than five days per week. 38.1% of the patients had a history of swimming or diving during the past 1 month. 51.8 % of the patients had a past history of sinus disease . The four most common symptoms were nasal blockage (88.5%), rhinorrhea (81.3%), nighttime cough (79.9%) and snoring (73.4%). The three most common signs were swelling of turbinates (75.5%), erythema of turbinates (74.8%) and tonsillar enlagement (48.2%). All paranasal sinuses X-rays were abnormal with maxillary sinus being the most commonly involved sinus (97.5%) followed by ethmoid sinus (58.8%). 58.8% of the patients had involvement of more than one sinus. Adenoid hypertrophy was positive in 36 % of all patients. The skin prick tests were positive in 62.6 % of all patients who the tests were performed.

Conclusion: Nasal blockage, rhinorrhea, nighttime cough and snoring are very common in acute sinusitis. The present study demonstrates that allergic rhinitis and recurrent sinusitis is common in our patients.

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Alteration of paranasal sinus mucosa in children with allergic rhinitis

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The purpose of the study is to determine the alteration of paranasal sinuses in pediatric patients diagnosed as having allergic rhinitis. We have studied 80 patients aged between 8 and 16 diagnosed with allergic rhinitis in the period 2005–2006. The associated diseases have been established: asthma, sinusitis, serous otitis media, hypertrophic chronic pharyngitis. Allergic rhinitis has been found to be perennial in 34 cases and seasonal in 46 cases. Associated sinusitis has been diagnosed in 42 patients, all of this have a high level of Ig E, from whom 28 patients had seasonal rhinitis. Main clinical symptoms: nasal discharge, nasal obstruction, sneeze, headache, eye itching, cough, postnasal drip. Clinical and radiological examination (X-ray) of paranasal sinuses has shown the alteration of maxillary, ethmoidal and frontal sinus mucosa. The mucosa is swollen or with serous effusion. The results of the study demonstrate that: maxillary and ethmoidal sinusitis is predominating affected in patients with seasonal allergic rhinitis. Radiological examination has shown predominant lesions of the type of sinusal swollen mucosa. Postnasal drip and cough associated with allergic rhinitis are indirect symptoms of the alteration of the paranasal sinuses. Allergic rhinitis in children must not be considered an isolated disorder, but must be considered in the context of a systemic allergic disorder. Sinus disorder in children is often misdiagnosed. Nasal mucosal edema at the level of the middle meatus favors associated sinus disorders.

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Presence of eosinophilic inflammation and airway remodelling signs in sinus tissues of patients with asthma and chronic rhinosinusitis

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Background: Chronic rhinosinusitis exists in a high amount of asthma patients. It is still unknown whether a cause and effect relationship is present between the two diseases or they are the manifestations of the same underlying disease process affecting the whole respiratory tract.

Objective: The aim of this study was to find out if sinus tissues in chronic rhinosinusitis have asthma related histological features such as eosinophilic inflamation and airway remodelling.

Methods: The sinus tissues taken from 15 asthmatic patients with chronic rhinosinusitis and who had ungergone functional endoscopic sinus surgery were examined histologically. The patients did not have nasal polyposis. The 'Hematoxylin and eosin' staining was used to detect epithelial damage. Basement membrane thickening of the airway mucosa was evaluated with PAS staining. These two signs were accepted as signs of airway remodelling. Eosinophil semiquantitative scoring system (0–4) was used to detect presence and amount of the eosinophils.

Results: 15 patints with mean age of 37 ±8.19 years were included in the study. 12 patients were female, 3 were male. 9 patients were found as atopic with skin prick test. 9 (60%) patients had epithelial damage with severe damage in 7 (46.6%). 11 (73.3%) patients had basement membrane thickening. 10 (66.6%) patients had eosinophils in the sinus tissue. 6 of these patients were allergic. The rate of allergy was the same in patients who had not eosinophils in sinus tissues.

Conclusion: In patients with asthma and chronic rhinosinusitis without nasal polyposis, the histopathologic signs of asthma can also be seen in sinus tissues of the patients taken with functional endoscopic sinus surgery. This can be a clue of the same pathogenetic event of the whole respiratory tract.

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Alveolar concentration and bronchial output of exhaled nitric oxide in chronic rhinosinusitis

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Background: Rhinitis and chronic rhinosinusitis (CRS), particularly with nasal polyps, are strong risk factors for asthma. Exhaled nitric oxide (FENO), a reliable non-invasive marker of airway inflammation, is known to be elevated in patients with asthma and rhinitis. In this study we investigated inflammatory and functional airway involvement in patients with CRS.

Methods: We recruited 47 patients with CRS, in all of them respiratory questionnaire, spirometry and FENO measurement (at constant flow of 50, 100 and 200 ml/sec) were obtained. Alveolar concentration (FANO) and maximal bronchial output (QbrMaxNO) of nitric oxide were calculated for each patient using a nonlinear model described by Silkoff et al. All patients with asthma-like symptoms were further investigated by methacholine inhalation challenge and/or bronchodilating test.

Results: 19 (40.4%) of our patients met the functional and clinical criteria of asthma, and 34 (72.3%) had nasal polyps. Among the 28 patients without asthma, 11 (39.3%) complained asthma-like symptoms. FENO was higher in patients with asthma (63.9, I.C. 95%: 47–81 ppb) and asthma-like symptoms with normal lung function (64.7, I.C. 95%: 39–91 ppb), compared with those without asthmatic symptoms (29.8, I.C. 95%: 21–39 ppb), $p<0.01$. All the three groups of patients had FENO higher than healthy controls (13.4, I.C. 95%: 6–19 ppb, $p<0.01$). QbrMaxNO was more elevated in patients with asthmatic symptoms, with and without lung function criteria for asthma (3.2 , I.C. 95%: 2.3–4.1nL/s and 3.5, I.C. 95%: 1.5–5.6 nL/s respectively) compared with patients without asthma-like symptoms (1.9, I.C.95%: 1–2.8 nL/s), $p < 0.05$. FANO was higher in patients with asthma-like symptoms but normal lung function (4.5, I.C. 95%: 2.4–6.6 ppb) compared with those with asthma (2.4, I.C. 95%: 1.6–3.4 ppb) and those without asthmatic symptoms (2.8, I.C. 95%: 1.1–4.5 ppb), $p=0.03$.

Conclusion: Patients with CRS showed higher levels of FENO compared to normal controls, suggesting a diffuse airway inflammation. FENO values were particularly elevated, with increased maximal bronchial output of nitric oxide, in patients with asthmatic symptoms, irrespectively of lung function (asthmatics or not), while alveolar concentration of nitric oxide was significantly higher only in patients with asthmatic symptoms, but normal lung function, suggesting a more peripheral inflammation in these patients than in patients with asthma.

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Microbiology of patients with a clinical diagnosis of sinusitis

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The diagnosis of Acute Bacterial Rhinosinusitis (ABRS) involves clinical symptoms and signs, without the use of plain sinus radiographs. We wished to determine the microbiology of sinusitis using these criteria compared to endoscopic culture results, with and without plain sinus films.

Methods: 103 patients diagnosed with acute sinusitis by clinical criteria were studied. Clinical criteria included biphasic illness, signs and symptoms of sinusitis and purulent sino-nasal drainage. After entry, patients had an endoscopic ostiomeatal culture and a Water's view plain radiograph.

Results: The pathogens identified in descending order were: Staphylococcus aureus (48.6%), Streptococcus pneumoniae (22.9%), Moraxella catarrhalis (11.4%), Pseudomonas (11.4%), Enterococcus (8.6%), Haemophilus influenzae (5.7%), Proteus mirabilis (5.7%), and other Strep species (5.7%). We also found 1 Methicillin Resistant Staphylococcus Aureus (MRSA) and 1 Aspergillus niger. The symptoms with the best combination of sensitivity, specificity, Positive Predictive Value (PPV) and Negative Predictive Value (NPV) were colored/purulent nasal drainage, congestion, and facial pain.

Adding a positive x-ray produced a specificity of 68.7%, and NPV of 68.7%, but reduced sensitivity and PPV to 36.4% each (respectively). Utilizing the clinical criteria of colored nasal drainage, congestion and facial pain together (without x-ray) provides the best combination of sensitivity, specificity, PPV, and NPV.

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Mometasone furoate nasal spray in seasonal allergic rhinitis

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Background: Mometasone furoate nasal spray (MFNS) is effective for preventing and treating nasal symptoms in seasonal allergic rhinitis (SAR). Its effects on ocular symptoms have not been investigated. This retrospective analysis examined the effects of MFNS on ocular symptoms in subjects with SAR.

Methods: Ocular symptom data were pooled and analyzed from four randomized, double-blind studies comparing MFNS 200 mcg once daily (n = 494) with placebo (n = 497). Subject-reported ocular itching, redness, and tearing were recorded at baseline and twice daily throughout treatment on a scale of 0 (none) to 3 (severe). Total ocular symptom score (TOSS) was defined as the combined 2-week average symptom scores.

Results: MFNS produced a statistically greater reduction in TOSS from baseline as compared to placebo (-1.33 vs. -0.94, $p < 0.05$). Likewise, mean 2-week reductions in individual symptoms were significantly improved with MFNS ($p < 0.05$ for each symptom). In subjects with TOSS 4 at baseline, MFNS recipients (n = 298) reported a significantly greater reduction in TOSS as compared to placebo recipients (n = 304; -1.97 vs. -1.51, $p < 0.05$), with statistically significant benefits also observed in individual ocular symptoms ($p < 0.05$ for each symptom).

Conclusion: MFNS has a beneficial effect on ocular symptoms, in addition to its established effects on nasal symptoms, in subjects with SAR.

DIAGNOSIS OF ALLERGY

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Identification of major allergens of royal jelly using 2-dimensional electrophoresis and mass spectrometry analysis

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Background: Royal jelly is a health supplement widely consumed in the community and has perceived benefits ranging including boosting the immune system. However, royal jelly consumption has been linked with contact dermatitis, acute asthma, anaphylaxis and death. Case reports of allergy to royal jelly have been reported among atopics. Our previous study among local allergic patients demonstrated that royal jelly sensitivity was found to be present in this country. Several major allergens in the range of 39 to 71 kD have been detected. Thus, the aim of this study is to identify the major allergens of royal jelly using 2-dimensional electrophoresis and mass-spectrometry analysis.

Methods: Extract of royal jelly was fractionated by charge and molecular weight using 2-dimensional electrophoresis (2-D). Immunoblotting of the 2-D profiles were performed to identify the allergenic spots of the major allergens using sera from royal jelly allergic patients. Spots were then excised from the 2-D gel, digested with trypsin and analyzed by MALDI-TOF MS and Q-TOF MSMS. For protein identification, the masses obtained were used to search databases to identify the proteins.

Results: 2-D gel fractionated the royal jelly proteins to more than 50 different protein spots. Out of these, 31 spots demonstrated specific IgE to the sera tested. Digested peptides of most allergenic spots as compared to the amino acid sequence in databases identified the fragments of royal jelly homologus to major royal jelly protein 1 (MRJ1) and major royal jelly protein 2 (MRJ2). Our results confirmed that the major allergenic spots of royal jelly are MRJ1 and MRJ2.

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Identification of IgE-binding proteins of Thunnus tonggol (tongkol) by mass spectrometry analysis

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Background: Fish is one of the most frequent cause of food allergy. Parvalbumins represent the main major allergens and responsible for IgE cross-reactivity between various species of fish. Tongkol belongs to the genus Thunnus that includes several species. Tuna, which is widely consumed worldwide, also belongs to this genus. The aim of this study was to characterize IgE-binding components of Thunnus tonggol (tongkol/ Northern bluefin tuna).

Methods: Uncooked and cooked extracts of the fish were prepared from the fish meat. Protein profile and IgE binding pattern was demonstrated by sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE) and immunoblotting using sera from subjects with fish allergy. The major allergens of the fish were then identified by two-dimensional (2-D) electrophoresis, followed by mass spectrometry analysis of the peptide digest.

Results: The SDS-PAGE of raw extract revealed 26 protein fractions over a wide molecular weight range, while cooked extract demonstrated only nine protein fractions. The 1-D immunoblotting detected 17 IgE-binding proteins, ranging in molecular weight from 151 to ~12 kD. Two protein fractions with molecular weight of ~51 and 42 kD have been identified as the major allergens of this fish. Both proteins were sensitive to heat. The 2-D gel profile of the raw extract demonstrated about >100 distinct proteins spots and immunoblotting detected at least 10 different major IgE reactive spots with molecular masses as expected and isoelectric point (pI) values ranging from 4.1 to 6.1. Comparison of the major allergenic spot sequences of the ~51 and 42 kD proteins with known protein sequences in databases revealed extensive similarity with fish enolase and triose phosphate isomerase, respectively. In conclusion, this study demonstrated that enolase and triose phosphate isomerase are the major allergenic component of this species of fish. Interestingly, our study also detected a heat resistant protein at ~12 kD as parvalbumin, which is similar as Sal s 1. However, this protein was only seen as a minor allergen for this fish.

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The analysis of mite and cockroach allergen in vitro test for allergic disease

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Objective: To investigate the level of specific IgE (SIgE) of mite and cockroach in allergy patients' blood serum, and study their correlation.

Methods: By ELISA test to examine the mite and cockroach allergen's sIgE from the blood serum of 199 outpatients who were recruited during 2004 Oct to 2005 Oct at Respiratory department and Allergy department.

Results: In the mite (dermatophagoides pteronyssinus(d1), dermatophagoides farinae(d2), and Blomia Tropicalis(d201) allergic group, the children showed higher proportion of positive result than the adult group. Some strong positive

results were all recorded from children. In the cockroach allergic patients, the positive results were all under the class IV level. For those patients who allergic to both of mite and cockroach, the mite allergen SIgE in blood was significantly higher then that of cockroach allergen ($p < 0.01$).
Conclusion: Mite and cockroach are both important allergens of allergic diseases, from which mite played a dominant role. For those patients who are allergic to both mite and cockroach, they are proved to be more sensitive to mite allergen.

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Recombinant tropomyosin of american cockroach, Periplaneta americana

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Background: American cockroach (CR), *Periplaneta americana* is an important source of indoor allergens among Thais. Currently, screening and monitoring allergic status of allergic patients are performed by skin prick test using crude CR extract which varies in the allergenic composition from batch-to-batch. Thus, recombinant CR allergens may be a better standardized alternative. Thus, in this study, we produced recombinant tropomyosin (Per a7) which is one of the *P. americana* major allergens. The protein was tested for its ability to bind to IgE in sera of CR allergic Thai patients.
Objectives: To produce recombinant tropomyosin of American cockroach, and to determine the IgE binding ability of the recombinant allergen.
Materials and Methods: Sera of patients with CR allergy who gave positive skin prick test to CR extract and sera of non-allergic counterparts were collected. Total RNA was extracted from adult *P. americana* and first strand cDNA was produced by RT-PCR. Gene encoding tropomyosin was amplified using oligonucleotide primers and the cDNA as a template. The amplified sequences were cloned into a cloning vector and a protein expression vector. The latter was used to transfect an *Escherichia coli* host. *E. coli* transformant was grown and production of tropomyosin protein was induced by IPTG. The protein was extracted and purified from the bacterial lysate.
Results: Gene encoding American cockroach tropomyosin (450 bp) was amplified and cloned into a cloning vector, i.e. pGEM T and a protein expression vector, i.e. pET 20 b+. Amplified sequence showed 100% homology to the tropomyosin gene sequence in the database. Recombinant tropomyosin was produced and purified from whole cell lysis of an *E. coli* transformant grown under IPTG induced-condition. The protein (~16 kDa) was found to bind IgE in sera of allergic Thai patients.
Conclusion and Clinical Relevant: Recombinant tropomyosin of *P. americana* was successfully produced in pure form. The protein binds IgE in sera of CR allergic Thai patients. Thus, the protein may be used as a standard reagent for screening and monitoring of the allergic status of patients.

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Serum allergen profiles related to moth and house dust mite in cockroach-allergic patients in-vitro assay and correlation analysis

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Objective: To look at the serum profiles of specific immunoglobin E (SIgE) related to moth and house dust mite allergens in cockroach-allergic patients and provide evidences for further research on insect hypersensitivity.
Methods: Thirty-three patients who presented to our Respiratory or Allergy Clinics between October 2004 and May 2007, and tested positive of Blattella germanica allergen by both skin prick test and SIgE test, were further evaluated for allergens related to Periplaneta americana, moth and house dust mite using fluorescence enzyme-linked immunosorbent assay.
Results: In patients who showed concurrent hypersensitivity to all of studied allergens, responses to the cockroaches and moth were weak-positive (below Grade IV), with significantly lower SIgE levels than as found for house dust mite ($P < 0.001$). Linear regression showed positive correlations in allergenicity between the two species of cockroaches and moth ($P < 0.001$), but not between house dust mite and moth or cockroaches.
Conclusion: High response to house dust mite was demonstrated in patients with multiple allergies. Allergens from Blattella germanica, Periplaneta americana and moth appeared to share some IgE binding site in common.

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Production and identification of major allergens of two species of local crab: blue/ swimming crab (Portunus pelagicus) and red crab (Charybdis japonica)

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Background: Crab has been recognized as a source of potent allergens especially in countries where seafood forms a large portion of the diet of the community. Blue/ swimming crab (Portunus pelagicus) and red crab (Charybdis japonica) from Family Portunidae are among the most commonly consumed crab in Malaysia. The objective of this study is to characterize the IgE-binding proteins and major allergens of these species of crab.
Methods: Cooked and uncooked extracts of both crab were prepared and fractionated by sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE). IgE-binding proteins were then demonstrated by immunoblotting tests using sera from 99 and 76 patients with positive skin prick test (SPT) to blue and red crab extracts, respectively.
Results: Both species of crab had similar protein profiles. The uncooked extracts of both crab produced 19 protein bands in the range of 15 to 138 kD. Several protein bands between 17 to 25 kD and 50 to 71 kD which were present in the uncooked extracts appeared to be denatured in the cooked extracts. The immunoblotting of blue and red crab revealed 14 and 15 various IgE-binding proteins, in the range of 24 to 138 kD, respectively. The most common IgE-binding proteins were identified in the range of 34 to 46 kD. A 34 kD protein was identified as a major allergen for both crab. A 36 kD protein was also demonstrated as a major allergen in red crab and two proteins at 38 and 46 kD were also seen as major allergens in blue crab. Interestingly, these major allergens at 34, 36 and 38 kD were similar in size with tropomyosin, the main major allergen and responsible in the majority of allergic reactions to various seafood species including crab.

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Skin prick test reactivity to peanut and tree nuts in adult and children with atopy

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Background: Nut allergy (peanut and tree nut) is a common cause of allergy and can even cause severe anaphylactic reactions. Peanut allergy is more common than tree nut allergy, but many subjects develop hypersensitivity to

both peanuts and tree nuts. Peanuts are known as ground nuts, earth nuts and monkey nuts while tree nuts refer to nuts such as Brazil nut, hazel nut, almond, walnuts and cashew nuts. The peanut plant, Arachis hypogaea, belongs to the botanical family Leguminose and do not related botanically to nuts such as Brazil nut, hazel nut or almond. There are no reports of prevalence of peanut and tree nuts allergy in Malaysia neither is there reports of fatal anaphylaxis due to nut allergy.

Methods: A total of 621 children and adults with history of atopy (asthma, allergic rhinitis, hives and food allergy) were skin tested to a panel of peanuts and tree nuts (commercial and in-house extracts).

Results: We found that children have higher skin test positivity to peanuts and tree nuts compared to adults. In children, skin prick test showed 8.5% positivity to raw peanut and walnut, 6% to big peanut and 4.3% to almond, and cashew nuts extracts whilst skin prick test positivity to both hazel nuts and walnut were lower at 2.6% and 1.7% respectively. In adults, the most common skin test reactivities are peanuts, 5% and 4% to cashew nuts.

Conclusion: It appears that skin test reactivity to nuts exist in subjects with no clinical presentation of nut allergy. A large number of individuals have skin test reactivity to multiple nuts and this is probably due to cross-reactivity.

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Allergen-specific IgE-values to inhalant and food allergens in China - comparison between two commercial immunoassays: Dr. Fooke ALLERG-O-LIQ versus Pharmacia CAP-System

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Purpose of the Study: Comparison of two in vitro assays for the detection of allergen-specific IgE (sIgE) and Total IgE (TigE) (Dr. Fooke ALLERG-O-LIQ, Neuss, Germany, www.fooke-labs.de; Pharmacia CAP-System, Upsala, Sweden, www.pharmacia.com) re-analyzing sera with sIgE to common inhalatans and food allergens and TIgE.

Materials and Methods: Allergens: Inhalants: housedust mite D. pter. d1, D.farinae d2, Blomia tropicalis d5, Cat (Epithelia) e1, Dog (Epithelia) e5, German Cockroach i6, Aspergillus fumigatus m3; food allergens (FA): hen's egg f1, cow's milk f2, Crab f23, shrimp f24.

Patients Sera: Sera from allergic patients (bronchus asthma, allergic rhinitis and chronic cough) were collected from Oct. 2004 to May. 2007 at Guangzhou Institute of Respiratory disease out patient department. Clinical data were not evaluated.

Performance: sIgE was measured in single runs according to the recommendations of the manufacturers. Assay features and methodological differences are listed in table 1.

Calculation: Associations between quantitative sIgE-levels of different assays were calculated using non-parametric Spearman-Ranktest and depicted with log scales. Sensitivity, Specificity and Agreement per allergen was calculated, using following formula: 1) Sensitivity (%) = (# of LIQ+ results)/(# of CAP+ result per allergen) 2) Specificity (%) = (# of LIQ- results)/(# of CAP- result per allergen).

Results: 1. Data see the appendix. 2. Quantitative values of both systems are associated to different degrees, depending on the specific inhalant allergen. 3. Dr. Fooke Allerg-O-LIQ has a good sensitivity, specificity and Agreement to the Pharmacia CAP-System.

Comments: Values of sIgE to inhalants detected by different in vitro methods show better correlation than sIgE to FA. Allergens with one dominant major allergen (i.e. e1) seem to be less difficult than complex allergen mixtures (i.e. d1). Identical units (kU/l) are no guarantee for equivalent sIgE results. The data presented do not allow estimates of the true sIgE concentrations or judgement on superiority of one of the tests over the other.

Conclusion: Comparative studies are necessary and helpful to define the concordance of sIgE-values detected by different immunoassays. Lack of concordance (complex allergen mixtures > simple allergens; FA > inhalants) should prompt subsequent improvement of allergens used in test systems detecting specific IgE.

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Cow's milk and gluten allergy in correlation with behaviour in autism children in Malaysia

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Background: Autism is a neuropsychiatric condition that presents with abnormal, bizarre behaviour patterns and accompanied by learning difficulties. Peptides known as casamorphin (from milk proteins) and gliadinomorphin (from gluten protein) are absorbed from the gut into the blood stream leading to the theory of "leaky gut syndrome".

Methods: Eighty-four autism children, divided into 2 groups: younger age (less than 5 years) and an older age group (5 to 19 years) were skin prick tested (SPT) to a panel of food allergens. Blood test for specific Immunoglobulin E (IgE) to cow's milk, wheat, soya and oat, specific Immunoglobulin G (IgG) and Immunoglobulin A (IgA) to alpha-lactalbumin, beta-lactoglobulin, casein and gliadin were carried out. Questionnaires on behavioural pattern of children were filled by parents.

Results: The majority of autism children were Chinese (76.2%) and male (88%). In the younger group, we found that SPT to soya 8.3%, wheat 3.5%, and cow's milk 2.3%. Specific IgE to cow's milk 9.52%, wheat 2.3%, and soya 1.2%. 8.3% had detectable levels to IgG alpha-lactalbumin, 5.9% to IgG beta-lactoglobulin, 1.2% to IgA beta-lactoglobulin and 4.76% to IgG casein. No IgA alpha-lactalbumin, IgG casein, IgG gliadin, IgA casein and IgA gliadin were detected in this group of children. In the older children, 23% has positive SPT to wheat, 17.8% to soya, and 9.5% to cow's milk. Specific IgE to cow's milk and wheat was 9.52% whereas to soya 8.3%. 11.9% had detectable IgG alpha-lactalbumin, 7.1% to IgG beta-lactoglobulin, 3.5% to IgG gliadin while 2.3% had detectable levels of IgG casein, IgA alpha-lactalbumin, beta-lactoglobulin, gliadin and 1.2% to IgA casein. As to behavioural pattern, the study showed that 70.6% of younger group and 64.2% of older group presented with unawareness of the world, 88.4% in younger group has poor eye contact compared to 61.2% in older group. There is not much difference in

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	Dr. Fooke Allerg-O-LIQ	Pharmacia CAP-System
Test principle	fluid phase reversed enzyme-immuno-assay (R-EAST)	solid phase fluorescence-enzyme-immuno-assay (FEIA)
Allergens	fluid phase allergens	solid phase allergens
Test procedure	1. IgE binds to anti-IgE. 2. Allergen binds to sIgE	1. sIgE binds to allergen. 2. Anti-IgE binds to sIgE
Markers	biotinylated allergens	β-galactosidase coupled anti-IgE
Detection	Photometry	fluorophotometry
Units	kU/l	kU/l
Classes	0 – 6	0 – 6

general learning ability and antisocial behaviour observed in both groups. In conclusion this study showed children 5 years and above had a higher SPT positivity to wheat and cow's milk. Casein was found to be low in both age groups and no gliadin was detected in the younger group. Behavioural pattern seemed to be improving as they became older and has no correlation with cow's milk and gluten allergy.

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Indigenous allergens in Saudi Arabia: Efficacy of diagnostic kits

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Background: In order to diagnose allergic patients with more relevant allergenic species which they are directly exposed to, indigenous species were collected and prepared commercially along with other allergens under a collaborative project with M/s Inmunotek, Madrid, Spain. These allergens were tested in allergic patients in the region to evaluate the efficacy and reactivity.

Methods: A total of 30 allergenic extracts with 50% indigenous species were included in the Diagnostic Panel. These allergenic species were selected after comprehensive aerobiological studies in many parts of the country and their growth and availability in the region. Glycerinated extracts were prepared by Inmunotek under a brand name of Allergotek. Standard prick tests with histamine and saline controls were conducted on 541 allergic individuals in the Middle East and Africa. The method is considered to be convenient and economical providing nature of sensitizing allergen(s) and an opportunity for a possible successful immunotherapy.

Results: A total positive reactions for all allergens (Mild, Moderate and Severe) were recorded; Jeddah 1123 (n = 194) Riyadh 731 (n = 108), Dammam 478 (n = 103, Khamesh 244 (n = 40), Hofuf 163 (n = 40) UAE 53 (n = 6) and Sudan 397 (n = 50). An overall reactivity pattern of 41% (1273 positives) was recorded with outdoor allergens and 59% (1916 positives) with indoor allergens. Maximum combined reactivity with pollen allergens for all sites were recorded with Cynodon dactylon (11.8%), Chenopodium murale (10.8%), Phoenix dactylifera (10%) Salsola imbricata (9.2%) Prosopis juliflora (8.9%) and Lolium perenne (8%). The maximum indoor allergens reactivity was recorded with D. farinae (15.7%), D. pteronyssinus (15.7%), Felis domesticus (12.5%) Periplaneta americana (10.4%), Blatella germanica (9.7%) and Blatta orietalis (8.4%).

Conclusion: The results of this efficacy trial of indigenous allergens revealed that majority of these allergens were effective with moderate to severe reactions. Asthmatic and allergic individuals were found to be comparatively more sensitive to indoor allergenic species than the outdoor allergens. While the cultural habits and climate appear to have played a role, socioeconomic conditions did not influence the overall sensitization pattern. It is further suggested that, if possible, regional species and/ or allergenic material should be included in the diagnostic test panel.

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Prevalence and sensitization to weeds pollen in Saudi Arabia

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Background: In an aerobiological study, airborne pollen grains from weeds emerged to be the most dominant and prevalent pollen type in Saudi Arabia. Therefore, we conducted Skin Prick Test (SPT) on 500 allergenic individuals to examine IgE mediated sensitization level to a number of weeds allergens.

Methods: SPT on 500 individuals having asthma and other allergic manifestations attending allergy clinics in six different regions was conducted using commercial extracts. The selection of allergens was made after an extensive nation wide aerobiological survey using Burkard Volumetric Spore Traps. The major pollen components of the Kingdom's environment were identified as *Amaranthus viridis., Atriplex polycarpa, Chenopodium album, Cyperus rotundus, Rumex crispus and Plantago spp*.

Results: The SPT data revealed a comparatively higher degree of sensitization to weeds pollen. In the south, mountainous region (Abha), 21.8%, while in an agricultural setting (Gassim) 75.5% patients reacted to weeds pollen. In an another location in the Eastern region (Hofuf) 16.7% of the patients while close to Red Sea region (Gizan) 9% of the patients reacted positively to various weeds pollen, which included *Atriplex polycarpa, Chenopodium album, Salsola tennifolia* and *Rumex crispus*. Individual pollen releaved *Chenopodium album* with maximum reactivity (81.8%) in agriculture setting (Gassim) followed by *Salsola tennifolia* (75.5%), (25% Al-Hofuf), *Rumex crispus* 27.3% (Gassim) and 18.1% (Gizan). Apart from *Cynodon dactylon*, a grass pollen and *Prosopis juliflora*, a tree pollen, highest skin reactivities were recorded by members of the *chenopodiace* weeds in all regions.

Conclusion: The study indicates that sensitization and exacerbation of symptoms in patients during pollination season may be caused by desert weeds growing in the Kingdom, and may possibly be a major contributor of respiratory allergy in Saudi Arabia.

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Specific IgG antibodies (total and subclasses) to saffron: a study of their correlation with specific IgE and immediate skin reactions

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Saffron (crocus sativus) cultivation is one of the most agricultural products in Iran. Saffron contains an aeroallergen that causes reactive respiratory allergic reactions in exposed subjects. To investigate the role of specific IgE and IgG in this type of allergic reaction, saffron specific IgE and IgG subclasses were determined in the sera of 38 exposed subjects (test group) and 20 normal subjects (control group) by ELISA. Immediate skin reactions were assessed in exposed subjects. The mean optical densities of saffron-specific total IgG and IgE antibodies were significantly higher in the test group than the control groups. Specific IgG4 antibodies were lower in the test group. Other IgG subclasses did not have any significant difference in the two groups. The immediate skin reactions were positive in 80% of the test group. The specific IgE antibody levels correlate with the specific total IgG antibody levels and the positive skin reactions (R = 0.67 and R = 0.42, respectively). A reverse correlation was found between the specific IgE antibody levels and the specific IgG4 antibody levels (R = 0.03).

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Can specific IgE testing in primary care be cost saving?

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Background: Allergy reduces quality of life and places a considerable burden on society. Specific immunoglobulin E (sIgE) testing can improve management and potentially reduce costs. The purpose of the study is to assess the cost-effectiveness of allergy testing for patients with respiratory and/or skin problems in primary care in Italy.

Methods: A cost-minimization analysis was carried out in which the costs of the two treatment alternatives were compared and assessed using a decision model that includes costs for pharmaceuticals, physician visits in primary care, and the costs of allergy testing. In the no-test-strategy part, patients were classified as allergic, non-allergic or uncertain, on the basis of clinical judgment without the test. In the test-strategy part, classification of patients was also based on sIgE results.

Results: Agreement between physicians' classification and test results was substantially elevated in the test-strategy part (0.88) vs the no-test-strategy part (0.52). Costs per patient over 2 years decreased from 784 in the no-test strategy to 535 in the test strategy, owing mostly to reduced medication costs. Sensitivity to the prevalence of allergy and price of medications was demonstrated.

Conclusion: Allergy testing in primary care is cost-saving compared with not testing. Decrease in false-positive or uncertain diagnoses is the main component of cost reduction and enables more appropriate patient management.

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Study of a clinical cohort of patients tested for allergy in a diagnositic immunolocgy clinic

Heidemarie Saxarra, Glenn Reeves, and Karla Lemmert. *John hunter Hospital Haps, Immunology Haps, Newcastle, Australia.*

A large number of patients (500) were studied and their allergy resuts collated. The specific allergen tests to dust mite, grass mix, food mix, animal epithelial mix, weed mix, latex and peanut were tabulated. This group of patients was then analysed for trends in any of the following parameters: (clinical notes and medical records were used) age, sex, medication, general health, symptoms of allergy and severity of the symptoms.

The study was limited in that all of the parameters were not available for the cohort studied.

No significant trends noted. However useful guidelines established for future reviews of a large patient cohort in the diagnositic laboratory.

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How can the royal college of pathologists of australasia immunology qap help your laboratory?

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Why does in vitro allergy testing survive? despite generally being less sensitive, slower and more expensive than skin testing?

1. Applicable when:
• skin disorders (e.g. eczema, dermatographism)
• medications (e.g. H1 anti-histamines, tricyclic antidepressants)
• risk of anaphylaxis (e.g. initial testing for drug allergy; extreme nut allergy) would prevent or inhibit skin testing
2. Does not require clinician to carry an expensive range of allergens
3. Lends itself to quality control, standardisation and regional collaboration

Why quality control in vitro allergy testing?

1. Some errors are clinically crucial; e.g. false negative result in nut allergy
2. Credibility of laboratory reagents, equipment and performance depends on satisfactory performance in QC
3. Understanding discrepancies can enhance products and performance
4. Meaningful dialogue depends on comparability of results

The RCPA immunology quality assurance program (QAP) is an internationally accredited (ILAC G13:2000) provider of External Quality Assurance (EQA) in Clinical Immunology. The Program has been based at Flinders Medical Centre in Adelaide since 1988. It offers 18 different programs, 5 of which are based on Allergy:

• Total IgE
• Tryptase
• Specific IgE to a range of common inhalant and food allergens, allergen mixes and Bee Venom

Participating Laboratories: Enrolled laboratories are drawn from Australia, New Zealand, Malaysia, United Arab Emirates, Hong Kong, South Africa, Singapore, France and Bahrain

Specimens: Serum specimens are obtained from patients with well characterised allergic disorders and clinical histories are provided

Reports: Reports are returned within 2 weeks of the closing date and provide a direct graphic comparison of individual laboratory results with all results received and with participants using the same methodologies.

Assessment is by assignment of 'Acceptable Scores' which are based on clinical criteria and set by a panel of clinicians, using skin prick test results if available. Quantitative results are not assessed, but are displayed graphically for comparative information purposes.

Education - Case Studies: Three informative clinical case studies provided by clinicians are included free of charge each year to participants. Clinical notes and results of preliminary investigations are provided. A discussion with references is provided by expert clinicians.

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Appropriateness of reducing the number of pollen allergens to three

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Background: Skin prick testing (SPT) is an essential tool in the diagnosis of allergic disorders. The optimal number and type of allergens used in different settings remains undefined. We aim to describe SPT in our clinical practice and propose the appropriateness of reducing the number of pollen allergens to three. The aim is to improve cost-effectiveness and reducing time spent on allergen testing, particularly in the community setting.

Methods: Consecutive patients who attended the private rooms of 2 immunologists and the allergy/immunology clinic in a tertiary referral hospital who required skin prick testing were evaluated from June 2006 to November 2006. Statistical analysis was undertaken with the use of Pearson's Chi-square and Fisher's Exact test to assess for significance. Multivariate analysis was also performed.

Results: There were a total of 273 skin prick test sets performed. There was no significant difference between the rates of SPT positivity with common pollen allergens between the two clinics. A positive SPT to Perennial Ryegass (*Lolium perenne*), Timothy (*Phleum patense*) or Bermuda grass (*Cynodon dactylon*) had a sensitivity of 100% to Bent or Orchard grass (*Dactylis glomerate*), with sensitivities of 97%, 96.3% and 94.8% to English Plantain (*Plantago lanceolata*), Bahia grass (*Paspalum notatum*)and Dock/Sorrel (Rumex sp) respectively. Use of Perennial Ryegrass, Timothy or Bermuda grass also detected tree pollen sensitivity with sensitivities in Birch mix (Betula sp) of 92.6%, Acacia (96.4%), Casuarina (89.1%), Platanus sp (92.3%) and Privet(Ligustrum sp) (88.6%).

Conclusion: The use of 3 common grass pollen allergens in SPTs (*Lolium perenne, Phleum pratense and Cynodon dactylon*) detected 90% of atopic individuals with sensitivity to many pollen types. This information may be useful in defining the most appropriate allergens to determine pollen hypersensitivity in community settings.

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Clinical evaluation of a new allergy lateral flow assay for professional and home use

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Background: Specific immunoglobulin E (sIgE) is a hallmark in the diagnosis of type I allergic reactions and atopic diseases. A new allergy screening test (Allergy Lateral Flow Assay; ALFA^{TM}) for qualitative detection of sIgE in human whole blood, serum or plasma is based on a test device, allowing linkage to a variety of allergens. Objective of our study was the evaluation of ALFA^{TM} for professional and home use.

Methods: Untrained volunteers (n = 96) performed ALFA^{TM} Seasonal Screen (S) [Birch (t3), Bermuda Grass (g2), Rye Grass (g5), Timothy Grass (g6), June Grass (g8), Cultivated Rye (g12), Mugwort (w6) and *Alternaria alternata* (m6)] and ALFA^{TM} Perennial Screen (P) [*D. pteronyssinus* (d1), *D. farinae* (d2), cat (e1), dog (e2), *Aspergillus fumigatus* (m3) and *Aspergillus niger* (m33)] with capillary blood. Each serum was tested for specific IgE to all single allergens contained in both ALFA^{TM} tests by ALLERG-O-LIQ. Furthermore, skin prick tests (SPT, Allergopharma) were performed. Volunteers were defined allergic if patient's history was concordant with SPT and sIgE in-vitro results. ALFA^{TM} results and patient's diagnoses were analyzed by kappa agreement, Chi-square test, positive (PPV) and negative predictive value (NPV) and diagnostic efficiency (DE).

Results: ALFA^{TM} results were obtained from 91 (S) and 83 volunteers (P). 33/91 (36.3%) volunteers, were defined as allergic for seasonal and 16/83 (19.3%) for perennial allergens. Of those, 25/33 and 6/16 showed positive test results in ALFA^{TM} S and P. Agreement between the ALFA^{TM} results and doctor's diagnosis was 94.5% (kappa = 0.75, $p<0.0001$, $\chi^2 = 49.3$) for S and 91.6% (kappa = 0.46; $p<0.0001$, $\chi^2 = 17.3$) for P. Overall agreement was 93.1% (kappa = 0.67; $p<0.0001$, $\chi^2 = 79.1$). Sensitivity, specificity, PPV, NPV and DE were 75.8%, 96.6%, 92.6%, 87.5% and 89.0% (S), 37.5%, 98.5%, 85.74%, 86.8% and 86.7% (P) and 63.3%, 97.6%, 91.2%, 87.1% and 87.9% (combined).

Conclusion: Results, particularly for seasonal allergens, are in good agreement with doctor's diagnosis. Therefore, ALFA^{TM} offers the opportunity for primary care physicians and patients to perform a screening test for early type-I allergy diagnosis.

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Sensitization to five common aero-allergens in children suffering from atopic eczema as examined by atopy patch tests, skin prick-tests and specific IgE

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Background: Although the role of allergy in atopic eczema (AE) is still controversial, some patients with atopic eczema suffer from exacerbation of skin lesions after contact with or inhalation of aeroallergens. From the histological examinations of the skin after contact with aeroallergenes is known that the delayed-type hypersensitivity reactions mediated by allergen-specific T cells can take a part in pathogenesis of atopic eczema. Atopy patch tests (APT) represent a useful tool for detection of such hypersensitivity.

Methods: We examined hypersensitivity to common aero-allergens (birch pollen, grass pollen, cat dander, house-dust mites) using APT, skin prick-tests (SPT) and specific IgE in 27 children suffering from atopic eczema. Results of all methods were then compared.

Results: Delayed-type hypersensitivity was found out (using APT) in 16 patients (59%), immediate type of hypersensitivity was found out (using SPT) in 13 patients (48%), using specific IgE in 15 patients (55%). Only immediate type of hypersensitivity was proved in 5 patients (18%), only delayed-type hypersensitivity in 6 patients (22%). Both types of hypersensitivity occurred concomitantly in 11 patients (41%). In 32 cases the type of hypersensitivity differed in the same allergen. A significant ($p< 0.0005$) positive correlation was found between SPT and specific IgE. Correlation of clinical symptoms of AE and positivity of tests was in 7 patients (26%) in IgE mediated hypersensitivity and in 10 patients (37%) in delayed-type hypersensitivity.

Conclusion: Various aero-allergens can influence substantially the course of atopic eczema not only via specific IgE, but as well by specific T cell-mediated

reactions. Therefore testing for hypersensitivity to aero-allergens both using SPT and/or specific IgE, and atopy patch tests could be useful.

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Complex diagnosis of IgE mediated allergy by in vivo and in vitro methods

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Background: The aim of the study is to assess the diagnostic potential of two in vitro methods for IgE diagnosis and to compare them with the skin-prick test (SPT) as a gold standard.
Methods: 131 patients with positive case history and SPT to grass pollen, house dust mite, moulds, bee and wasp venom suffering of bronchial asthma or allergic rhinitis /hay fever and 10 clinically health controls were studied. The in vitro quantity of serum allergen-specific IgE (UniCap, Pharmacia) and the percentage of allergen-specific basophil's degranulation (FasImmune, BD) were evaluated. The correlation and the percent of coincidence of the results from the three methods were analysed (Statistica 5.5).
Results: Significant statistical correlation between the resuls from the three methods in patients sensitized to grass pollen and house dust mite were found. Strong positive correlation (Spearman, $p<0.05$) between the SPT and the quantity of specific IgE- R=0.67 and R=0.61, between the in vivo test and FasImmune - R=0.66 and R=0.62 and between the both in vitro methods - R=0.67 and R=0.53 were determined. Data from patients, allergic to insect's venom, showed a high percent of coincidence between the three methods - from 70% to 90%. Respectively a cincidence of 60% between the SPT and the quantity of specific IgE in the group sensitized to moulds was established.
Conclusion: The results from the invitro methods represent positive correlation and coincidence with SPT, especially for the allergens of grass pollen, house dust mite, bee and wasp venom. Their application ensures more preceise diagnosis of patients and contributes to the complex assessment of IgE mediated allergy.

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Development and optimization of quartz crystal microbalance immunosensor for the detection of total IgE

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Background: Allergic disease have a significant impact on clinical practice due to their high prevalence. The total IgE quantification is one of the important steps in the classic atopic disease diagnostics.The most widely used methods for IgE detection are time-consuming and complex. Biosensors are interesting tools offering certain operational advantages over standard photometric methods, notably with respect to rapidity, ease-of-use, cost, simplicity, portability, and ease of mass manufacture.
Methods: QCM work as sensors based on the relationship between frequency change and mass loading on the surface of the crystal according to Sauerbrey equation (Sauerbrey, J Phys, 1959). When antigens react with coated antibodies on the surface, a frequency shift occurs and this change is proportional to the mass loading.
Results: The monoclonal anti-IgE were successful immobilize in Nafion polymeric matrix on silver electrodes of piezoelectric quartz resonator. The optimal conditions for anti-IgE immobilization procedure and for piezoelectric immunoassay have been determined. Only 10 microlitres of serum and 45 minutes reaction time is required to measure total IgE. It was found that biosensor is capable to differentiate blood serums of patients with low, intermediate and high level of IgE.
Conclusion: The quartz crystal microbalance immunosensor offers a number of significant advantages over the currentlu available in vitro techniques for the detection of total IgE. It is supposed that such biosensor can be used in laboratory practice for IgE determination.

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Systemic reactions to percutaneous and intradermal skin tests

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Background: The purpose of this study is to determine over 12 months, 2/1/06–1/31/07, the rate of SRs to both P and ID ST, the symptoms reported, and the response to immediate treatment with epinephrine IM.
Methods: A retrospective review over a one year period was conducted to evaluate SRs to P and ID ST to 20 to 50 allergens (trees, grasses, weeds, animals, molds, foods, medications, and Hymenoptera) in 1,456 subjects. A standard form was used to record symptoms, signs, and treatment. No vasovagal reactions were included. Nurses as instructed by the attending physicians administered epinephrine 1:1000 v/v, 0.2mL IM as soon as any signs or symptoms of anaphylaxis occurred.
Results: 52 patients (3.5%) had SRs, 43 (83%) female and 9 (17%) male. The average age of the patients with SRs was 40.6 years (range 13–70, median 35.5 years). 17/52 (33%) had asthma. Symptoms reported: pruritic eyes, nose, and/or pharynx (40%), worsening cough (27%), sensation of difficulty swallowing (17%), worsening nasal congestion (15%), rhinorrhea (13%), chest tightness and/or shortness of breath (13%), generalized pruritus (12%), sneezing (12%), urticaria (4%), and wheeze (4%). No severe asthma, shock, hypotension, unconsciousness, or late phase responses occurred. Treatment: 52 (100%) patients received epinephrine (average dose, 0.2 cc, 1:1000 IM), 48 (92%) oral prednisone, 9 (17%) oral prednisone to take 6 to 8 hours after reaction, 50 (96%) oral antihistamine (H1) , and 6 (12%) nebulized beta agonist.
Conclusion: SRs occurred in 3.5% of patients skin tested and readily responded to early intervention with epinephrine. This early administration of epinephrine by nurses appears to prevent more serious and late phase reactions.

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Clara cell protein in irritating and sensitizing effects of inhaled benzalkonium chloride in rats

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Background: Benzalkonium chloride (BAC) is a bacteriostatic agent used in the pharmaceutical industry as a preservative and is known to cause bronchoconstriction in asthmatic subjects. The aim of our study is qualification of results of inhalation exposure to BAC in rats, with particular reference to the effect on the remodelling of the respiratory system. Conditioned allergic reactions that impair lung function, such as allergic asthma, can be evaluated by specific lung biomarkers. It is known that the irritant fumes affect nonciliated epithelial Clara cells, which release anti-inflammatory and immunosuppressive Clara cell protein (CC16) into the respiratory tract.
Materials and Methods: Female Wistar rats were exposed to BAC aerosol at 30 mg/m3 for 5 days (6h/day) and on day 16 were re-exposed to BAC for 6 h.

After the exposure, bronchoalveolar lavage fluid (BALF) was collected. BALF concentration of total protein, CC16, IgE, MMP-9, hyaluronic acid (HA), IL-6, TNF-α, MIP-2 and activity of lactate dehydrogenase (LDH) were determined. CC16 as the marker of bronchiolar epithelium was assessed by latex immunoassay. In the lung, histological examinations were done and the activity of glutathione S-transferases (GST) was determined. Additionaly total and differential cell number (lymphocytes, neutrophils and macrophages) were measured.

Results: Benzalkonium exposure after challenge induced statistically significant increases of BALF cytokines, LDH and IgE in BALF and serum. CC16 level in BALF was significantly reduced. Significant negative correlation of CC16 concentration in BALF with mediators (IL-6) of inflammatory processes was seen. Huge increase of LDH correlated with the level of total protein, MIP-2 and IgE in serum. Negative relationship was shown to occur between CC16 and LDH. IgE in serum and BALF correlated with MMP-9. In histopathology examination, focal agglomerations of alveolar macrophages were noted as well as proliferation of peribronchial lymphatic tissue.

Conclusion: CC16 play a protective role in allergic inflammation and take part in remodelling effects of low molecular weight sensitizers. CC16 can be used as a diagnostic marker for early detection of impaired respiratory function.

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Pulmonary irritation after inhalation exposure to benzalkonium chloride in rats

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Background: Benzalkonium chloride (BAC) is a quaternary ammonium compound in which the alkyl groups have a chain length from C8 to C18. BAC exerts toxic effects on microorganisms. This property has been utilized in the cosmetic industry and medicine, where it is used as an effective germicide and preservative agent. Various BAC-containing preparations used by people may produce a number of adverse effects on the human body. Bearing in mind that BAC is widely used in different branches of the national economy, its toxic effect may constitute a major health problem.

Materials and Methods: Female Wistar rats IMP: WIST of body weight 165–185 g were exposed to BAC aerosol at the target concentration of 30 mg/m3 in the dynamic inhalation chamber for 6 h and 3 days (6 h/day). After the exposure and 18 h after termination of exposure to BAC aerosol, bronchoalveolar lavage fluid (BALF) was collected from each animal and BALF concentrations of total protein, Clara cell protein, matrix metalloproteinase-9 (MMP-9) hyalurnic acid (HA), immunoglobulin E (IgE) and cytokines (TNF-α, IL-6 and MIP-20) and the activity of lactate dehydrogenase (LDH) and GSH-S-transferase (GST) were determined.

Results: All the rats survived inhalation exposure to 30 mg/m3 BAC. A significant reduction of body weight was noted in the animals exposed repeatedly by inhalation to BAC. Lung weight, total protein, HA level and LDH activity in BALF were higher in rats after single and repeated exposure to BAC, compared to control. Decreased concentrations of CC16 in BALF of rats were observed after the single and repeated inhalation exposure. A significantly higher level of IL-6 and IgE were noted in the BALF from the animals exposed to the single and repeated dose. Concentrations of MMP-9, TNF-α, and MIP-2 in BALF of rats exposed to BAC were similar to those found in the control animals.

Conclusion: BAC showed a strong inflammatory and irritating activity in the lungs of the rats already after 6 hours of inhalation exposure. BAC stimulates the dynamic patterns of IL-6 and IgE production and infiltration of protein from blood circulation system to BALF. Continued exposure resulted in changes involving cellular destruction, statistically increase of LDH activity and a continuous reduction of CC16 concentration in BALF.

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Allergic bronchopulmonary aspergillosis in an asthma clinic using essential minimal criteria

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Objective: Allergic bronchopulmonary aspergillosis (ABPA) occurs in cases of atopic asthma and may result in important lung disease. Early diagnosis is essential as this disease is responsive to corticosteroids. However, there is still no consensus about the diagnostic criteria, because patients in different stages of ABPA may not fulfill the criteria. In this study, we evaluate the prevalence of ABPA or ABPA-like disease in an asthma clinic using essential minimal diagnostic criteria.

Methods: A prospective evaluation of patients with bronchial asthma for ABPA from July 2006 onward. ABPA was diagnosed using essential minimal criteria ?asthma, skin prick testing (SPT) positivity to Aspergillus fumigatus (Af), elevated serum total IgE (CAP), elevated serum Af-specific IgE (CAP), and central bronchiectasis on CT scans.

Results: Ninety consecutive patients with bronchial asthma were enrolled. Forty-four of 90 patients were atopic (49.0%), 7 of 44 (18.0%) were positive to SPT to Af. Five of 44 patients (11.0%) showed only elevated serum Af-specific IgE without positive response to Af on SPT. A secure diagnosis of ABPA, satisfying all essential minimal criteria, was evident in 4 of 12 patients (33.3%).

Conclusion: There is high prevalence of ABPA in asthmatic patients presenting our hospital. Further evaluations are required to differentiate ABPA from asthma patients sensitizing to Af without ABPA. The role of serum Af-specific IgE as a screening tool in diagnosis of ABPA should be redefined.

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Clinical presentation in 12 patints with allergic bronchopulmonary aspergillosis

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Purpose: Allergic bronchopulmonary aspergillosis (ABPA) is an immunologically mediated lung disease charecterise by a Complex hypersensitivity reaction in patients with asthma which occurs when bronchi become colonized

	No. of patients Average	Average
Age (years)		25.3
Cough, wheezes and dyspnea	12	
A history of asthma	12	
Immediate skin test reactivity to Aspergillus antigens	12	
Serum total IgE concentration greater than 1000 ng/mL	10	
Peripheral blood eosinophilia >500/mm3	12	2180
Lung infiltrates	12	
Proximal bronchiectasis	4	
High ESR mm in the first hour	12	64.4

by Aspergillus. Repeated episodes of bronchial obstruction, inflammation, and mucoid impaction can lead to bronchiectasis, fibrosis, and chronic lung disease. Our aim of the study is to increase awareness of this disease.
Methods: We described a study of 12 cases with Allergic bronchopulmonary aspergillosis Twelve patients (6 men and 6 women) were diagnosed in chest department at King Hussien Hospital between 1993–2003. The main criteria for the diagnosis were A history of asthma, immediate skin test reactivity to Aspergillus antigens, Serum total IgE concentration greater than 1000 ng/mL, peripheral blood eosinophilia more than 500/mm3, Lung infiltrates and proximal bronchiectasis.
Results: Demographic data for 12 patients with Allergic bronchopulmonary aspergillosis.
Conclusion: ABPA is a rare disease, diagnosis is depending upon certain criteria.
Clinical Implication: We have to think about the diagnosis of ABPA in any patient with a history of asthma, lung infiltrates and peripheral blood eosinophilia.

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The significance of the diagnostic profile of the ophthalmic allergies in excluding mimicking clinical conditions(MC)* which may pose therapeutic difficulties

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Introduction: Patients with (AC) with/without concomitant allergies in some cases is a therapeutic dilemma.
Methods: In the series of patients, ages 10–35 years usually with intermittently red. eyes(shot redness), intractable itching of eyes, tearing (stringy discharge) with/without seasonal association.

On laboratory investigations, is found raised tears & blood eosinophils counts, total eosinophils counts. Total serum IgE measurement in most of the cases had been higher than 200 to 300 kU/l. supported by, the. rise in titers of allergen-specific IgE, by the radioallergosorbent test (RAST) method, Skin prick test with a mixture of allergenic extracts had a conclusive evidence of an allergy cause for the red eye.

On ophthalmoscope examination. Found pinkish papillae, with a central vessel & characteristics, serous, watery conjunctival secretion.

Conjunctival scrapings, and tear cytology performed after topical ocular allergen challenge to sensitized subjects have shown significant increases in neutrophils and eosinophils, and their presence evidenced a positive diagnostic criterion.
Results: Confirmation of a suspected allergic sensitization by skin prick test for the diagnosis of immediate hypersensitivity is the most sensitive, fastest and cheapest method to confirm an allergic sensitization. However, it carries a small but a significant risk of systemic anaphylaxis.
Conclusion: Challenge tests are the only way to relate the specific allergen to the triggering of ocular symptoms. but with a variable degree of systemic anaphylaxis.

*Bacterial, chlamydial and viral conjunctivitis, superior limbic, phlyctenular, conjunctivitis, keratoconjunctivitis, rosacea-associated conjunctivitis, erythema multiforme, eoiscleritis/scleritis, and ocular cicatricial pemphigoid.

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Electrodiagnostic study of phrenic nerve function in patients with systemic lupus erythematosus

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Objective: 21 patients with SLE were screened for the presence of Phrenic nerve neuropathy and to determine whether neurophysiologic findings correlate to clinical respiratory signs, spirometric abnormalities or serological examination in patients with Systemic lupus erythematosus.
Methods: A total of 21 patients(18 female & 3 male)with systemic lupus erythematosus (SLE) (age range, 16–36 yr) were included and studied by physical pulmonary examination, chest radiography, respiratory function tests, as well as serological examination and bilateral transcutaneous phrenic nerve conduction studies.
Results: 14(66.6%) patient complained of dyspnea ,only one patient showed paradoxical abdominal movement .Pulmonary function tests showed proportional reduction of the forced vital capacity (FVC) and forced expiratory volume in 1 second (FEV1), suggesting a restrictive process which was severe in 23% of patients. All patients were on corticosteroids, only 10 (47.6%) patients were on immunosuppressive medication to include methotrexate or cyclophosphamide. Phrenic nerve evaluation using transcutaneous stimulation studies showed delayed latencies of RT, LT & both phrenic nerve in 17 (81%), 19(90%) and 17 (81%) patients respectively confirming a demyelinating neuropathy. Also Phrenic nerve stimulation evoked a low-amplitude response from the right, left and both in 17 (81%), 15 (71%) and 14 (66.6%) of patients respectively confirming axonal neuropathy. There was no significant correlation between electrical phrenic nerve stimulation and serum immune markers, except there was decreased action potential amplitude in SLE group with positive results for Anti DNA as14 (66.6%) of patients had Anti DNA + ve, all showed reduced amplitude of rt phrenic nerve & 13(93%) of them showed reduced amplitude of lt phrenic nerve. Fourteen (66.6%) patients presented with dyspnea and all of them showed abnormal phrenic nerve conduction studies. While 11 patients showed abnormal CXR with small but clear lung fields, no evidence of major parenchymal lung or pleural disease was found. There was no significant correlation between electrical phrenic nerve stimulation and CXR abnormalities.
Conclusion: Diaphragmatic weakness in patients with SLE is both common and is very likely to be caused by a phrenic neuropathy with evidence of bilateral involvement.

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The search engine as a diagnostic tool in complex immunological and allergic case reports: is google useful?

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Background: Web-based search-engines have become an important source of knowledge and communication. Google is the most popular search-engine (64% of all web-searches in March 2007) whilst Yahoo accounted for 21%. Recently Google's value to guide doctors to a correct diagnosis in case records of the New England Journal of Medicine was reported.
Objective: To evaluate the utility of searching clinical information with Google in order to obtain a correct diagnosis of complex immunological and allergic (CIA) case reports.
Study Design: Comparative cross-sectional study.
Methods: Firty-five CIA case reports were randomly selected by an independent investigator from peer-viewed medical journals. Clinical data was presented separately to three observers blinded to final diagnosis. Observer A is a Consultant in Internal medicine and Allergy with an expert knowledge of these fields and basic computing skills. Observer B is a Registrar in Internal medicine and Allergy. Observer C is a research nurse. Both observers B and C had a more familiar knowledge of the regular use of computer search engines. An internet-based search using Google was conducted. In order to perform this, the observers individually studied each text and independently selected five search terms, of their own choosing, from each case record to enter into the standard Google search engine. The observers then recorded for each case the single most prominent diagnosis that

was evident from within the first three results pages of the conducted Google search. Since Google does not necessarily include diagnoses within the search results page itself, observers were permitted to select the diagnosis that best fitted the case record from information after opening each direct results link only. The independent investigator then compared the diagnoses obtained by each observer with the definitive diagnoses as published in the Journals. The main outcome measure of this study was the percentage of correctly obtained diagnoses achieved by each observer.

Results: Observer A identified the definitive diagnosis in 30/45 cases (66%, 95%CI 52–79). Observer B in 39/45 (86%, 95%CI 76–95) and Observer C in 29/45 (64%, 95%CI 50–77). Most diagnostic inaccuracies for both observers were those related to primary immunodeficiency or pediatric cases.

Conclusion: This Google-based search was useful to achieve an appropriate diagnosis in CIA cases. Computer and Internet-based search skills could influence the results.

PHARMACOTHERAPY

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The anti-allergic properties of potassium humate

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Background: Although the anti-inflammatory properties of humate derived from peat, sapropeles and mumie have been described, no clinical studies has been done on the anti-inflammatory effects of humate derived from coal. Leonardite humate compared favourably with prednisolone in suppressing contact hypersensitivity in a rat model. According to a report by the European Agency for the Evaluation of Medicinal Products on toxicity studies (Feb 1999), humic acids extracted from brown coal has no toxic effects on rats in a chronic study at oral dosages as high as 1g/kg BW, whereas the LD50 in rats, after oral administration of humic acids, has been reported to be greater than 11g/kg BW. This report has recently been confirmed by a separate study.

The objective of this study was to establish the safety and therapeutic efficacy of oral potassium humate in reducing the signs and symptoms of hay fever in atopic patients during the grass pollen season.

Methods: In this parallel double-blind placebo controlled phase II study potassium humate was randomly assigned, at a dosage of 1.8g in divided doses/day, to atopic patients (n = 40) presenting with acute symptoms of hay fever. The blood and nasal samples were used to determine the safety and the effects of potassium humate on basophil activation, cytokine levels and eosinophil migration. A skin prick test was used to determine its anti-allergic effects. An in vitro neutrophil adhesion test was used to determine the effects of the product on the adhesion of human neutrophils to ICAM-1expressing baby hamster kidney cells.

Results: A significant decrease in the skin prick test results (presented elsewhere) and eosinophil counts was observed. No significant differences were observed with regard to neutrophil adhesion nor were there any differences observed with regard to the stimulation of basophils. However decreases were observed in the expression of IL-4, IL-5, IL-8 and IL-1â after treatment, although not reaching statistical significance. The product had no effect on neutrophil adhesion to ICAM-1.

Conclusion: This study confirmed, without doubt, that this product possesses anti-inflammatory as well as anti-allergic properties possibly due to a decreased recruitment of eosinopils to the site of inflammation.

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Anti-inflammatory effect of LipoPGE1 on therapeutic intervention for intractable skin ulcer

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Background: Intractable skin ulcer is common in our daily practice. Although patients' QOL is severely impaired by this skin lesion, until now effective treatment protocols have not been established. Wound healing process can be separated into inflammatory phase, proliferating phase, and remodeling phase. Recent reviews have described that prolonged inflammatory phase might partly affect the pathogenic mechanisms of intractable skin ulcer. During the inflammatory phase, various kinds of inflammatory cell infiltrates ware observed in the affected area, and recruited macrophages secrete some inflammatory cytokines including IL-6 and VEGF. Prostaglandin E1 (PGE1)-treatment is believed to be one of the promising treatment for skin ulcers, and LipoPGE1 also demonstrated improvement in a drug delivery system. To note, it has been reported that infiltrated macrophages uptake LipoPGE1. Taken these results together, we hypothesized that LipoPGE1 might have an anti-inflammatory effect and thus contribute to the improvement of intractable skin ulcer.

Methods: Patients with various kinds of intractable skin ulcers were administered with intra-venous injection of 10 microgram/day LipoPGE1 (Palxus®) for two weeks, and the size of the ulcer area and serum concentration of CRP, IL-6, VEGF, and sICAM1 were measured before and after treatment.

Results: LipoPGE1 effectively reduced the size of the ulcer area and the serum inflammatory markers after two weeks of LipoPGE1-treatment.

Conclusion: These results indicated that anti-inflammatory effect of LipoPGE1 might contribute to the improvement of the intractable skin ulcers.

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Comparative efficacy of levocetirizine, desloratadine, clemastine, kvifenadine and sekvifenadine on histamine prick test induced weal reaction, blood perfusion evaluated by laser Doppler flowmetry. Randomized, double-blind, placebo-controlled, crossover design study

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Background: Evaluation of weal reaction and laser Doppler flowmetry are valuable methods for evaluation of efficacy of different pharmacological agents. The aim of our study was to compare the influence of different H1-antihistamines on histamine induced weal reaction, increase of skin blood perfusion and sedation.

Methods: Histamine prick test induced weal area in mm2, percentage of blood perfusion change and area under curve during peak perfusion period (AUCmax) was measured with Periflux System 4000 (Perimed AB, Sweden) 2 hours after intake of 5 mg levocetirizine, 5 mg desloratadine, 1 mg clemastine, 50 mg kvifenadine, 50 mg sekvifenadine and placebo. Sedative effect was measured in mm by visual analogue scale (VAS).

Results: Results were expressed as mean ± 95%CI. Mean weal reaction area was 6.9 (−3.9;+10.7); 17.5 (−12.6;+23.1); 20.2 (−14.9;+26.2); 18.1 (−13.1;+23.9); 17.8 (−12.8;+23.5) and 29.0 (−22.6;+36.1) mm2 respectively. Statistically significant difference was observed between active treatment and placebo ($p<0.05$), and levocetirizine and other H1-antihistamines ($p<0.001$). Increase of blood perfusion was 393.1% (−221.3;+613.8); 626.2% (−403.0;+898.3); 756.5% (−508.8;+1053.2); 741.0% (−496.2; +1339.1); 1001.5% (−712.8;+1339.1) and 1033.2% (−739.6;+1375.8) respectively. Significant decrease of augmentation of blood perfusion was observed after pre-treatment with levocetirizine and desloratadine vs. placebo ($p<0.05$) and levocetirizine vs. kvifenadine, sekvifenadine and clemastine ($p<0.05$). AUCmax was 1298.7 (−781.2;+1947.0); 2197.3 (−1504.5; +3020.9); 2454.3 (−1718.3;+3321.0); 2633.2 (−1868.6;+3528.6); 2551.7 (−1800.0;+3434.2) and 3166.2 (−2321.4;+4141.7) U*s. AUCmax was significantly lower after pre-treatment with levocetirizine vs. placebo and other antihistamines ($p<0.05$). Sedative effect was 24.5 (−17.9;+32.1); 21.1

(−15.1;+28.1); 28.2 (−21.2;+36.2); 17.6 (−12.0;+24.1); 15.1 (−10.0;+21.0) and 19.9 (−14.1;+26.7) mm of VAS. Significant difference of sedation we observed between levocetirizine vs. sekvifenadine, clemastine vs. kvifenadine, sekvifenadine and placebo ($p<0.05$).

Conclusion: Levocetirizine induced significant decrease of weal and flare reaction and skin blood perfusion compared to placebo and other H1-antihistamines. Influence of kvifenadine and sekvifenadine on weal reaction area was similar to desloratadine and clemastine. Sedative effect of clemastine was more pronounced than kvifenadine and sekvifenadine.

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Characterising the pharmacological properties of fluticasone furoate, a novel enhanced-affinity glucocorticoid

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Background: Fluticasone furoate (FF) is a novel enhanced-affinity glucocorticoid developed for topical respiratory use. Its distinct pharmacological properties have been investigated in several in vitro and in vivo studies.

Methods: Binding affinity of FF for the human lung glucocorticoid receptor (GR) was determined by elucidation of association and dissociation rate constants. Molecular interactions of FF with the GR were identified by X-ray crystallography of the GR ligand-binding domain. Cellular onset of action was determined by measuring nuclear translocation in human lung epithelial cells. Measurement of human progesterone (PR), mineralocorticoid (MR), androgen (AR), and oestrogen receptor (ER) activities was used to assess the steroid hormone selectivity of FF. Cellular protection of FF to elastase or mechanical wounding was determined in 16HBe human lung epithelial cells and anti-inflammatory effects of FF in the lung were determined using the Brown Norway Ovalbumin Rat model.

Results: FF has very fast association with, and slow dissociation from, the GR, with a relative receptor affinity (RRA) of 2988±135 with reference to dexamethasone (RRA: 100±5), higher than all other currently available clinical glucocorticoids: mometasone furoate (MF) 2244±142, fluticasone propionate (FP) 1775±130, beclomethasone-17-monopropionate 1345±125, ciclesonide active principle 1212, and budesonide 855. FF has hydrogen bond interactions with the GR through the 3-keto (with Gln570 and Arg611) and the 11β-hydroxy (with Asn564) groups; the 17 β-fluoromethylthioester group also forms a favourable electrostatic interaction with Asn564. FF induces a rapid translocation of GR into the nucleus (<20 minutes to maximum effect) and has high selectivity for GR (30-, 790-, >330,000- and >330,000-fold) versus PR, MR, AR and ER, respectively. FF confers substantial protection against elastase-and mechanically induced damage, with more potent protection than budesonide, FP and MF. FF completely prevents lung eosinophilia, an effect greater than that with FP.

Conclusion: FF has enhanced affinity for the GR compared with other available glucocorticoids, which translates into more potent protection against cellular damage and lung inflammation. Coupled with its fast cellular onset of action and high selectivity for the GR, these properties may contribute to a favourable clinical efficacy and safety profile for FF.

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Downmodulatory effects of cetirizine and levocetirizine on cytokine/chemokine production and CD54 expression in keratinocytes

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Cetirizine is an antihistamic drug of the second generation. Besides its anti-histamic activity various actions have been reported in this anti-histamic. In epidermal keratinocytes, cetirizine inhibits the expression of co-stimulatory molecule ICAM-1 and the MHC class II molecule HLA-DR. Moreover, it exerts anti-inflammation actions by suppressing the production of cytokines and chemokines in various immunocompetent cells. Levocetirizine (L-cetirizine) is the optical isomer of cetirizine and widely used for the treatment of allergic disorders in European countries. In this study, we investigated whether there are differences between cetirizine and levocetirizine in the cytokine and chemokine production by normal human epidermal keratinocytes (NHEK).While NHEK were stimulated with interferon-γ(IFN-γ) and tumor necrosis factor-α (TNF-α) cetirizine or levocetirizine was added to the experimental cultures. Three day-culture supernatants were measured for the concentrations of IL-1α, IL-8, RANTES, Mig, I-TAC and MDC. The IFN-γ/TNF-α-augmented levels of IL-1α, IL-8 and I-TACK were significantly suppressed by the addition of either cetirizine or levocetirizine to the culture in dose-dependent manners (10^{-9} – 10^{-7} M). RANTES, Mig or MDC was not suppressed by cetirizine. To examine the effects of these two reagents on the expression of CD54 (ICAM-1) molecules, NHEK were incubated with IFN-γ with or without cetirizine or levocetirizine for 48 hrs. Cetirizine and levocetirizine at 10^{-8} M downmodulated the expression of CD54 molecules at similar levels to each other.This study demonstrates that cetirizine and levocetirizine have comparable effects on the immunological function of keratinocytes. It is noted that levocetirizine has slightly but significantly stronger effects than cetirizine in the production of RANTES and Mig.

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Efficacy and safety of levocetirizine 5mg as continuous or on-demand treatment for persistent allergic rhinitis over 6 months

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Background: We aimed to document the efficacy and safety of levocetirizine 5mg as continuous (CT) or on-demand treatment (OnD) of persistent allergic rhinitis (PER), as defined by ARIA.

Methods: This was a single-center, randomised, open-label study comparing CT vs. OnD treatment of PER patients with levocetirizine 5mg, once daily, over 6 months. Patients were allowed to have mild asthma treated with a short-acting beta-agonist. Sneezing, rhinorrhea, nasal and ocular pruritus (T4SS=sum of these 4 symptoms) and nasal congestion were measured daily on a 0 (absent) to 3 (severe) scale. Rhinasthma questionnaire (RQ) was used to assess subjects' quality of life (range 0–100; 30 item questionnaire: 1=not bothered at all, 5=bothered very much). Quality of sleep was reported on a VAS scale (0=worst; 10=best).

Results: 31 patients were enrolled per group; 18 in the OnD and 22 in the CT group completed the study. No patients discontinued for drug-related serious adverse events (AEs).

Improvement from baseline in T4SS was significantly higher in favour of CT during months 5 ($p<0.01$) and 6 ($p<0.03$). The maximal T4SS improvement was 80% for OnD and 87% for CT. The maximal improvement in nasal congestion was 75% for OnD and 85% for CT. Quality of sleep considerably improved at end of study: from baseline VAS=5.77 (OnD) and VAS=5.63 (CT) to VAS=7.82 (OnD) and VAS=7.25 (CT). No serious (AEs) were observed. 3.3% of subjects in the CT and 9.4% in the OnD groups reported drug-related treatment-emergent AEs.

Conclusion: Our study confirms previous data that, when taking a potent antihistamine, like levocetirizine, PER symptoms (including nasal congestion) are effectively controlled over a 6-month treatment period. In addition, most of the individual symptoms were controlled significantly better when treated continuously. Regardless of the regimen, levocetirizine improved the patients' quality of life and sleep, and was very well tolerated with fewer patients

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Change from baseline		Month 1 (Adj. Mean)	Month 2 (Adj. Mean)	Month 3 (Adj. Mean)	Month 4 (Adj. Mean)	Month 5 (Adj. Mean) (p-value vs. OnD)	Month 6 (Adj. Mean) (p-value vs. OnD)
Sneezing	OnD	−0.97	−1.22	−1.28	−1.14	−0.95	−0.77
	CT	−0.74	−0.86	−1.26	−1.31	−1.50 (0.004)	−1.41 (0.008)
Rhinorrhea	OnD	−1.32	−1.47	−1.62	−1.51	−1.27	−1.05
	CT	−0.90	−1.07	−1.39	−1.54	−1.75 (0.023)	−1.63 (0.019)
Nasal pruritus	OnD	−1.11	−1.38	−1.63	−1.46	−1.30	−1.14
	CT	−1.02	−1.10	−1.26	−1.47	−1.74 (0.022)	−1.57 (0.081)
Ocular pruritus	OnD	−0.97	−1.11	−1.28	−1.32	−1.18	−1.06
	CT	−0.90	−1.01	−1.16	−1.37	−1.61 (0.016)	−1.51 (0.055)
Nasal congestion	OnD	−1.08	−1.36	−1.54	−1.23	−1.19	−1.03
	CT	−1.11	−1.26	−1.41	−1.55	−1.66 (0.059)	−1.39 (0.210)

		Baseline Mean	Month 1 Mean	Month 2 Mean	Month 3 Mean	Month 4 Mean	Month 5 Mean	Month 6 Mean
RQ Upper Airways	OnD	52.91	30.26	21.53	22.83	29.87	30.56	21.14
	CT	53.11	33.62	29.68	21.71	19.67	22.47	23.86
RQ Lower Airways	OnD	25.19	15.45	11.24	10.54	11.54	11.03	9.40
	CT	29.78	19.19	18.50	12.87	11.92	13.81	14.53

reporting AEs in the CT group. Our results support the long-term continuous treatment of PER with a potent and well-tolerated antihistamine.

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Oxatomide-treated children with atopic dermatitis complicated by food allergy and prevention of asthma development

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Background: Recent epidemiology suggests the increasing prevalence of allergic diseases in the industrialized countries including Japan, which necessitates the analysis of the mechanisms of allergic diseases and development of the effective treatment. Oxatomide (OXM), an antihistaminic drug, has been shown to be clinically effective for the treatment of hypersensitivity and childhood asthma. Its mode of action has been elucidated to increase IFN-g activity as well as anti-histaminic reaction.

Objective and Methods: Peripheral blood mononuclear cells were obtained from 41 patients with atopic dermatitis allergic to hen-egg ranging from 2 months to 2 years 10 months in age. The patients had recurrent eczema, pruritus and positive skin reactions to egg white and/or cow's milk. Patients also had positive responses to the oral provocation test to raw hen egg and or cow's milk. Diagnostic criteria for atopic dermatitis was based on the criteria of Hanifin and Rajk. To clarify the mode of action whereby OXM ameliorates the conditions of the children with food allergy-complicated atopic dermatitis and whether development of bronchial asthma is prevented, OXM-related alterations of the clinical symptoms and examination, seen in patients during the course of 8 months' to 6 years and 10 months' treatment was evaluated.

Results and Discussion: Scores for itching, sleep disturbance and skin lesions (inflammation, lichenification, cracking) was improved from 10 to 2.7 (mean) during the course of 8-16 weeks' treatment with 2mg/kg of OXM in addition to elimination diets, treatment of skin care (shower, Isodine[R], non-steroid ointment), administration of hydroxyzine and/or oral sodium cromoglicate. In further study, OXM efficiently suppressed incidence of asthma to approximately 9% of the patients (control:42.9%) and both of total IgE value and peripheral eosinophils count was not elevated after OXM-treatment and were lower than those in age-matched asthma patients un-treated with OXM.

Conclusion: OXM, which is a significant candidate for one of the therapeutic modalities against children with food allergy-induced atopic dermatitis, and based on the clinical study, was also found to be effective prophylaxis for development of childhood asthma.

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A comparison between intramuscular dexamethasone and fluticasone propionate inhaler in treatment of croup

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Introduction: Croup is a common viral disease in children under 6 years old with incidence rate of 2–6%. The mainstay of treatment is airway management. Treatment focuses on respiratory distress, using cold mist, epinephrine, heliox and corticosteroids. In this study we tried compare the effectiveness of Fluticasone spray with intramuscular Dexamethasone.

Materials and Methods: In this clinical trial, 107 children with croup randomly assigned into two groups. The study group was treated by Fluticasone Propionate and the control group was treated by intramuscular Dexamethasone. Croup scoring was performed at the 6th and 12th hours from initial administration according to Westley croup score.

Results: Improvement was observed in 83% of the study group and 66% of the control group, 6 hours after initiation of treatment. In both groups 10% of the patients didn't respond to treatment ($p = 0.03$). 12 hours after treatment the study group response was 85% and the control group response was 90% ($p = 0.4$).

Conclusion: We found that Fluticasone Propionate and Dexamethasone have similar efficacy in treatment of respiratory distress, considering the simple method of using Fluticasone spray, it can be suggested as a good treatment for croup.

Key words: Croup, Fluticasone Propionate, Dexamethasone, Westley Croup Score.

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Bioequivalency of single doses of desloratadine administered as syrup and tablet formulations in healthy volunteers in China

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Background: Desloratadine (DL) is an oral, non-sedating, selective and potent H1 receptor antagonist that is indicated for the treatment of allergic

rhinitis and chronic idiopathic urticaria in pediatric and adult populations. The clinical and pharmacological profiles of DL have been extensively investigated and DL is available in a number of formulations, including tablets and syrup. The bioequivalence of DL tablets and syrup has not been studied to date in a Chinese population.

Methods: This randomized, open-label, single dose crossover trial studied the pharmacokinetics of DL 5mg administered as a 5mg tablet or 10 ml of 0.5 mg/ml syrup in 24 healthy adult male Chinese subjects. After providing written informed consent and undergoing screening, subjects were admitted to a clinic for baseline assessments. Subjects were randomized to receive one of the two DL formulations in a fasting state. Blood tests for pharmacokinetics were taken over 5 days (subjects remained in the clinic for the first 24 hours), and after a 14 day washout period the subjects were crossed over to the other DL formulation and underwent identical pharmacokinetic analyses. The main pharmacokinetic variables for the two formulations were the log-transformed AUC(I) and the Cmax for DL and 3-OH-DL. Biochemical and hematological tests, ECG data and vital signs were also assessed during the study and adverse event (AE) reports were collected.

Results: DL was safe and well tolerated when administered in the tablet or syrup formulations; no AEs were reported. The Tmax, T1/2, Cmax, and AUC(I) values for DL and 3-OH-DL were similar for both formulations. There were no statistically significant differences between the tablet and syrup DL formulations on the basis of log-transformed Cmax and AUC(I) values for DL and 3-OH-DL (P>0.05). The 90% CIs of AUC, and Cmax were 91.61–103.97% and 86.04–99.92% respectively for DL, and 94.22–101.71% and 88.01–101.35% respectively for 3-OH-DL. The relative bioavailbility of the DL syrup was 99.4% for DL and 98.79% for 3-OH-DL, which met the criteria for bioequivalence of the two formulations.

Conclusion: Both syrup and tablet formulations of DL 5mg were safe and well tolerated. When administered as a syrup formulation DL was bioequivalent to the tablet form of DL in healthy Chinese subjects.

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The pharmacokinetic and safety profiles of desloratadine in healthy korean volunteers

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Background: Desloratadine (DL) is a non-sedating, selective and potent H1-receptor antagonist that is effective and well tolerated in the treatment of subjects with allergic rhinitis and chronic idiopathic urticaria. The pharmacokinetics (PK) of DL have not been studied in a Korean population, to date.

Methods: This was a double-blind, dose escalation study of the PK and tolerability of single doses of DL 5mg, 10mg and 20mg in 36 healthy male Korean subjects. In each dose group 10 subjects received DL and 2 received placebo. Safety was demonstrated at the 5mg DL dose before escalation to the next dose group. Subjects were screened for eligibility during a 3 week pre-baseline period. Subjects were confined on Day -1 and baseline blood/urine tests and ECGs were performed. DL was administered fasting at 9 am on Day 1 and subjects remained confined until Day 3. Blood sampling was performed for biochemistry, hematology and PK from DL administration until Day 8 and the Tmax, terminal T1/2, Cmax and AUClast for desloratadine and it 3-OH-DL metabolite were calculated. Vital signs, physical examinations and ECGs were performed regularly and adverse event (AE) reports were collected.

Results: No clinically relevant changes in vital signs, biochemical, hematological or ECG results occurred. Nine subjects reported 13 AEs during the study. Only dizziness, gingival bleeding and a flu-like illness (all mild in severity) were deemed potentially related to study drug. One subject developed a hemopneumothorax on Day 6 after receiving DL 5mg. This was deemed as being of unknown relationship to study drug. The mean terminal T1/2 for DL was 23.7–31.1 hr across the dose range, while the mean Tmax ranged from 1.75 to 2.0 hr. The Cmax ranged from 2.4 μg/L (DL 5mg) to 9.9 μg/L (DL 20mg); the AUClast ranged from 36.5 μg.hr/L (DL 5mg) to 170.3 μg.hr/L (DL 20mg). The mean AUClast and Cmax values for DL demonstrated moderate intersubject variability (50.6% and 51.1%, respectively) at the 5mg DL dose. The intersubject variability for AUClast and Cmax was less at the 20mg DL dose (21.4% and 25.3%, respectively). The mean AUClast and Cmax for DL and 3-OH-DL demonstrated linear PK.

Conclusion: Single dose DL was well tolerated in healthy Korean volunteers across a dose range from 5mg to 20mg and is consistent with results in other populations. With increasing doses of DL, AUClast and Cmax demonstrated linear PK.

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Cardiac safety evaluation of loratadine in the treatment of allergic rhinitis in elderly patients

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Background: In elderly patients with allergic rhinitis, the second-generation H1-antihistamines have not been adequately studies, although they are widely used and assumed to be safe.

Objective: To evaluate cardiac safety of loratadine in the treatment of allergic rhinitis in elderly patients.

Methods: A total of 40 patients with perennial allergic rhinitis were enrolled in the study. There were 25 males and 15 females, aged 50 to 88 years (mean, 64.4-years-old). 17 cases (42.5%) had a history of cardiovascular diseases and/or presented abnormal ECG parameters, but had no prolonged QT-interval. The subjects received loratadine 10mg once-daily for 30 days. A series of baseline ECG recordings was obtained before treatment. ECG effects of the treatments were then compared with the baseline ECGs.

Results: There were no changes in sinus rhythm in all patients after 30 days treatment by loratadine. No statistically significant difference was found between the heart rates, P durations, PR and QRS intervals at baseline and end-point ECGs (P >0.05), with no significant prolongation of the QT as well as QTc corrected for heart rate using Bazett formula (P>0.05).

Conclusion: The results suggest no cardiotoxicity of loratadine, at the usual recommended dose, in long-term treatment of allergic rhinitis in the elderly.

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Propranolol cytotoxicity on human leukemic MOLT-4 cell line

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Background: Propranolol, a beta-adrenergic blocker has been used for treatment of a large number of cardiovascular diseases. This drug is also an inhibitor of phosphatidic acid (PA) phosphohydrolase and phosphatidic acid biosynthesis. Phosphatidic acid is a growth factor for tumor cells. In addition, the inhibitory effect of Propranolol on the development of a tobacco-induced pulmonary adenocarcinoma and also its cytotoxicity on rat and human lung macrophages and human lung tumor cell line has been reported. The widespread and long-term use of propranolol in lots of heart diseases as well as its cytotoxicity against some tumor cells, prompted us to investigate its cytotoxic effect on a human T leukemic cell line (MOLT-4).

Methods: The MOLT-4 cells were cultured in complete RPMI medium and then incubated with different concentrations of Propranolol (0.0004 –0.4 mM) for 10 and 20 hours. The cytotoxicity was then assessed by 3-[4,5-dimethyl thiazol–2,5-diphenyltetrazoliumbromide (MTT) reduction and also trypan blue dye exclusion methods.

Results: Propranolol induced a significant dose dependent cytotoxic effect on human MOLT-4 cell line in less than 10 hours compared to untreated control cells.

Conclusion: The results showed that human T leukemic cell line was dose dependently sensitive to Propranolol. Further studies investigating the in vivo effect of Propranolol on leukemic patients and also other leukemic cells are warranted.

INFECTION & IMMUNITY

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Human coronavirus infections in Hong Kong children: epidemiology, disease spectrum and relationship with childhood wheezing illnesses

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Background: Human coronaviruses (HCoVs) are enveloped viruses with a large plus-strand RNA genome. Five serologically distinct groups of HCoVs have been described - HCoV-229E, HCoV-OC43, HCoV-NL63, HCoV-HKU1 and SARS-CoV. The clinical disease spectrum by HCoVs in our population is not clearly defined. Preliminary studies suggested that HCoVs might be related to childhood wheezing. This prospective study investigated the epidemiology and clinical features of HCoV infections in Hong Kong children.
Methods: Nasopharyngeal aspirate (NPA) samples were taken from children who were hospitalised in our university teaching hospital between April 2005 and March 2006. The clinical features, diagnoses and laboratory investigations in these subjects were prospectively collected, and laboratory staff blinded to these details performed low-stringent reverse transcription-polymerase chain reaction (RT-PCR) assays using 12 pairs of primers that detect constant regions of HCoVs (i.e. pancoronavirus).
Results: 1139 subjects (57% males) were recruited, with mean (SD) age being 5.1 (3.6) years. The main discharge diagnoses were pneumonia (n=239), upper respiratory infection (URI; n=227), asthma (n=191), seizure (n=107), bronchiolitis (n=105), roseola infantum (n=98), croup (n=31), and others (n=141). Twenty-eight (2.5%) of these NPA samples were positive for HCoVs. The clinical diagnoses associated with these HCoV isolates included asthma (n=7); seizure (n=6); URI (n=5); bronchiolitis, pneumonia, tonsillitis and roseola infantum (n=2 for each); and croup and otitis media (n=1 for each). HCoV infection was not related to age, highest respiratory rate and maximal temperature (P>0.3). HCoV infection was not associated with wheezing illnesses as defined by 'asthma', 'bronchitis' or 'bronchiolitis' (2.7% *versus* 2.4%; P=0.870) or with lower respiratory infections (the above three plus 'pneumonia'; P= 0.341). HCoV cases were more likely to suffer from seizure (5.6% *versus* 2.1%, P=0.040). Complete blood count and C-reactive protein were not related to HCoV infections (P>0.15).
Conclusion: HCoVs are uncommon yet important pathogens causing seizure disorders in local hospitalised children. On the other hand, HCoV infections are not associated with wheezing illnesses in Hong Kong children.

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Are there predominant strains of Staphylococcus aureus in atopic dermatitis patients? : Genotypic characterization of staphylococcus aureus isolated in adolescent and adult patients with atopic dermatitis

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Background: The colonization of *Staphylococcus aureus* is one of the most important aggravating factors of atopic dermatitis. Until now, the importance of *S. aureus* in atopic dermatitis and a positive correlation between colonization with *S. aureus* and clinical severity / skin barrier function has been demonstrated. Qualitative analysis, especially a genotypic characterization of *S. aureus* isolated from atopic patients, however, has rarely been reported.
Methods: This study aimed to find the genotypic characterization of *S. aureus* from atopic dermatitis patients. We performed newly-developed typing methods - spa typing, multi-locus sequence typing (MLST) and toxin gene assay, by a multiplex polymerase chain reaction, with 165 isolates of Staphylococcus.
Results and Conclusion: The results showed that there was no predominant clone of *S. aureus* with a high heterogenicity of spa typing and MLST. A toxin gene assay showed very interesting results that all *S. aureus* strains had at least two kinds of toxin genes; sea and tsst-1 being the most prevalent.

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Role of primary and secondary low-grade rhinovirus infection in allergic airway inflammation in a murine model of allergic asthma

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Background: Rhinovirus respiratory syncytial virus (RSV) infection is known to develop and exacerbate asthma in young children. In adult, RSV causes recurrent but asymptomatic infections. However, the impact of asymptomatic RSV infection on adult asthma is yet to be determined. The aim of this study was to determine the effects of primary and secondary low-grade rhinovirus infections on allergic airway inflammation in a murine model of allergic asthma.
Methods: A low-grade rhinovirus (2 x 10(3) plaque-forming units/mouse) was inoculated, and this caused neither pulmonary inflammation nor symptoms but induced significant IFN-gamma production in thoracic lymph nodes. To investigate interaction between low-grade virus and Dermatophagoides farinae (Df), airway hyperresponsiveness, lung inflammation and cytokine production from thoracic lymph nodes were compared after primary and secondary low-grade rhinovirus infections in four groups of mice; control, Df allergen-sensitized, rhinovirus-infected and Df-sensitized rhinovirus-infected mice. A direct comparison between low- and high-grade rhinovirus infections was also performed in primary infection. To investigate the role of IL-5 during secondary rhinovirus infection, anti-IL-5 monoclonal antibody (anti-IL-5 mAb) was injected in mice and similar parameters were compared in four groups of mice.
Results: Primary high-grade rhinovirus infection increased allergen-induced airway inflammation, while primary low-grade rhinovirus infection attenuated allergen-induced airway inflammation concomitant with significant IFN-gamma production in lung-draining lymph nodes. In marked contrast, secondary low-grade rhinovirus infection increased both IFN-gamma and IL-5 production, resulting in exacerbation of allergen-induced airway inflammation. Anti-IL-5 mAb treatment in secondary low-grade rhinovirus infection and Df allergen-sensitized mice attenuated virus and allergen-induced airway inflammation.
Conclusion: Low-grade rhinovirus infection per se does not cause pulmonary inflammation, whereas it induces a significant immunological response in the allergen-sensitized host. These results indicate that subclinical and recurrent rhinovirus infection may play an important role in exacerbation and maintenance of asthma in adults, wherein IL-5 is critically involved.

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Serum zinc levels in young children with recurrent wheeze

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Background: Zinc is one of the dietary antioxidants. Previous studies have shown that zinc is crucial for normal development and function of cells mediating non-specific immunity. Recently, zinc supplementation was reported to reduce acute lower respiratory infections and prevent severe pneumonia in children. Our purpose was to examine zinc levels in the serum of the young children who had recurrent early wheeze and evaluate the clinical and laboratory findings in relation to zinc status.

Methods: Seventy-three patients (aged from 8 months to 6 yrs) admitted with acute respiratory infection with wheezing were enrolled. All children had experienced more than 3 episodes of wheezing before admission. Zinc levels were measured in serum samples collected on admission using inductively coupled plasma-optical emission spectrometry (ICP-OES) and the value of < 64 mg/dl was defined as zinc deficiency. Clinical and laboratory findings in the children with zinc deficiency were examined and compared with in the children who had normal values. Zinc levels in sixteen age-matched controls were also studied.
Results: Median value of zinc levels in the patients was significantly lower than in controls ($P<0.001$). 36 patients were found to have zinc deficiency (49.3%), which was significantly higher than in controls (12.5%). Zinc deficiency was observed in 56% of the patients = 2 yrs of age and 40.6% of >2 yrs of age. There was no significant difference in total WBC count, lymphocyte count and atopic status in relation to zinc status in the patients. CD4/CD8 ratio was significantly lower in the patients with zinc deficiency ($P<0.05$), however, other immune profiles were within normal limit.
Conclusion: This study showed that median value of zinc level was significantly lower and zinc deficiency was more frequently found in the patients with recurrent early wheeze compared with in age-matched controls. Our results suggest that zinc deficiency may be associated with frequent respiratory viral infections, a likely trigger for recurrent early wheeze in the young children.

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Persistent cough in patient with infection for mycobacterium avium intracellulare infection

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Aim: Mycobacterium avium intracellulare (MAV) is the atypical Mycobacterium most commonly associated with human disease. The pulmonary disease is the most frequently clinical presentation and appears with higher prevalence in immunosuppressed patients.
Materials and Methods: We present the case of a 46 years old woman, nurse as profession , with cough and dysnea for a period of nine years. No wheezing, fever nor constitutional syndrome were referred. Skin prick tests with common aero-allergens and latex, spirometry and bronchodilatation test were performed. Total IgE, complement study, proteins electrophoresis, immunoglobulins determination, cellular immunity study, HIV, X-ray study and thoracic CT-scan, mantoux, zielh and sputum culture were done.
Results: Positive skin prick test for pollens and dog and cat epithelia were obtained. The patient had normal spirometry values and a negative bronchodilatation test. All the laboratory tests were in normal levels. Determination of total IgE was 483 KU/l. The chest X-ray showed cavities in both lungs with interstitial infiltrates. The CT-scan confirmed these findings. Mantoux, Zielh with 50 BAAR/field and MAV culture were positive. Mycobacterium tuberculosis was excluded by CRP. Cellular immunity, complement, proteins electrophoresis and immunoglobulins determination were in normal range. HIV test was negative.
Conclusion: We present the case of a patient with rhinoconjuntivitis due to pollens hypersensitivity and persistent cough with pulmonary infection for Mycobacteium Avium associated. The Mycobacterium Avium was not described as human pathogen until 1950, when many series described pulmonary infections for MAV. This mycobacterium mainly attacks immunosuppressed patients. This infection is less frequent in patients with normal immunity. Our patient did not have immunosuppression nor risk factors. At the present time she is being treated with antibiotics (ethambutol and claritromicin) and she is in good general condition with no need of hospitalisations.

IMMUNODEFICIENCY

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Molecular diagnosis of Chronic Granulomatous Disease in Iran

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Chronic Granulomatous Disease (CGD) is an inherited phagocytic disorder caused by mutations in NADPH oxidase subunits. Patients with CGD have life-threatening bacterial and fungal infections. Childrens Medical Center at Tehran University is the referral center for immunodeficiency in Iran. During two years of study forty five families with clinically diagnosed CGD were referred to this center. Neutrophil functional assays performed for affected children and their mothers; no activity or residual activity was detected in affected neutrophils. PMN (Poly Morpho Nuclear) oxidative burst revealed mosaic pattern in 12 mothers. Western blot analysis revealed gp91° phenotype in all their sons. Mutation screening in CYBB gene using SSCP analysis followed by sequencing, showed 9 different mutations including one novel mutation. Western immunoblot subtyping of patients whose mother showed no mosaic pattern by DHR123 revealed 24 patients with p47 null expression, 7 and 2 patients with p22 and p67 defect, respectively. ΔGT screening in Ncf1 gene for p47° patients, revealed 8 patients with this mutation. Mutation analysis for the rest of Ncf1 gene for these patients is understudy. CYBA mutation analysis revealed 6 different mutations including three novel mutations in p22° patients. Overall, the number of autosomal recessive patients with CGD in Iran is high and it seems consanguineous marriages is one of its causative factors.

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First HIV/AIDS vaccine clinical trials in Russian Federation: cohort formation

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Background: The first clinical trials of HIV/AIDS vaccine in Russia (phase I) – VICHREPOL, recombinant protein antigen (comprised C-terminal p17, full length p24 and gp41 immunoreactive fragment) conjugated with polyoxidonium adjuvant – started in 2005 (HVRF-380-131004, IAVI List of ongoing trials of preventive AIDS vaccines). Volunteers recruitment was the first experience of cohort formation for clinical trials of HIV/AIDS vaccines in Russia. Willingness, reasons to participate and reasons of refusal were estimated also.

Methods: 15 volunteers (HIV non-infected healthy persons, 20–50 y.o.) were planned to enroll into phase I trials. Recruitment started by the wide spreading of information on clinical trials of VICHREPOL through media, medical centers and NGOs (predominantly in Moscow and Moscow region). All appropriate papers and information were presented to volunteers. Motivation, understanding of vaccine properties and trial procedure, risk behavior, ability to perform the trial protocol were estimated. Volunteers enrolled in trials after confidential medical investigation and informed consent undersigning.

Results: 65 persons of 298 enquiries were screened as potential volunteers. The main reasons for participation: help to HIV-infected people (65%) including persons in nearest surround (wife, husband, friend, child) (41%); contribution in development of HIV/AIDS vaccine (35%); possibility of immune protection against HIV (18%), free insurance and/or medical care (4%). 20% (13 of 65) of persons refused to participate in trials. Reasons of refusal: doubts in vaccine safety and fear to receive HIV-infection (61%), fear of side effects (30%), fear of vaccine-induced HIV seropositivity (15%), impossibility to perform the trial protocol (7%), low compensation (2%), unexplained (15%). 30 persons not refused to participate in clinical trials, but decided to do it in future (clinical trials phase II). Reasons: planned pregnancy or temporal impossibility to perform the trial protocol. 17 volunteers (14 men, 1 woman, age 22–31) were enrolled in trials. 2 of them leaved trials after 1st vaccination (unexplained reason), 9 successfully passed the trials protocol and 6 are at final step of trials.

Conclusion: The first cohort for the clinical trials of preventive HIV/AIDS vaccines was created in Russia and partially enrolled in clinical trials of VICHREPOL vaccine. Cohort is stable (88%), expandable and may be included into international multicenter trials.

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A novel mutation within exon 12 of the CYBB gene resulting in severe form of x-link chronic granulomatous disease

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Chronic granulomatous disease (CGD) is a primary phagocytic disorder with defective superoxide formation and intracellular killing. The most common form, X-linked CGD (X-CGD) resulted from the mutation of CYBB gene on chromosome Xp21.1. We evaluated a Thai boy who had multiple Salmonella septicemia, Aspergillus pneumonia and brain abscess. His nitroblue tetrazolium (NBT) test was reportedly abnormal. The dihydrorhodamine (DHR) flow cytometry assay was performed and the fluorescence pattern upon stimulation was compatible with typical X-CGD. CYBB analysis revealed a novel complex mutation atggacg→ ttca in exon 12 (base pairs 1532–1538). As a result, 3 amino acids Tyr 511, Gly 512 and Arg 513 were deleted and replaced by 2 amino acids, Phe and Gln. The DHR and mutation analysis of his mother showed normal DHR pattern and no mutations in exon 12 of CYBB gene. Over 300 CYBB mutations have been registered in an internationally maintained X-CGD database. Most mutations are distributed throughout the 13 exons or at exon/intron boundaries, and almost 200 of these mutations are unique. We reported a novel mutation within exon 12 of CYBB gene which was on the nicotinamide adenine dinucleotide phosphate (NADPH)-binding domain in a CGD patient. Functional defect was demonstrated by almost absence of fluorescence upon stimulation of granulocytes on DHR histogram. This defect leads to a severe form of X-CGD.

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Nutrition status in Iranian patients with primary antibody deficiency

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Background: Nutrition is an important factor that influences immunity, and nutritional deficiency can impair resistance to infections.Malnutrition is the most common cause of immunodeficiency worldwide. Trace elements such as zinc, selenium, iron, and copper can influence several components of immunity. Primmary antibody deficiency disorders are a group of disorders characterized by an unusual susceptibility to infections and malnutrition. Impaired nutritional status has been reported in immunodeficient patients. The aim of this study was to determine anthropometric indices and trace elements status in these patients.

Methods: Thirty-eight children (28 males, 10 females, aged 2–18 years) with primary antibody deficiency referring to Children,s Medical Center of Tehran University of Medical Science were enrolled in this research. Primary immunodeficiency disorders consisting of CVID, XLA, IgA deficiency, IgG subclass deficiency, and hyper IgM were assessed. Anthropometric indices, comprised of height, weight that were measured and body mass index (BMI) was calculated. Height-for-age (HAZ), weight-for-height (WHZ) and weight-for-age (WAZ) were determined according to Z-score to study mild, moderate and severe malnutrition. Serum copper, zinc, selenium and iron levels were measured by an atomic absorption spectrometer.

Results: The most common disorders were CVID 52.5% and X-linked agammaglobulinemia 27.5%. Based on BMI measuredment 21.1% of patients had malnutrition. According to HAZ, 13.2%, 13.2%, and 36.8% had severe, moderate and mild malnutrition, respectively. According to WAZ, 10.5%, 18.4 %, and 28.6% had severe, moderate and mild malnutrition, respectively. Regarding to WHZ, 14.3%, 28.6 %, had moderate and mild malnutrition, respectively.Low selenium levels and high copper levels were obsevd in 37.5% and 70.3%, respectively.

Conclusion: Anthropometric data showed that the frequency of malnutrition in these patients was higher than the CDC standard. Low serum selenium levels and high serum copper levels were observed, suggesting further research is needed on these parameters. Most of the patients had serum zinc and iron levels within the normal range. It is recommended that clinical immunologists and nutritionists should make a collective effort to provide these patients with standard or specialized diet so as to decrease the risk of infection.

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A registry of primary immunodeficiencies in a University Hospital in Thailand: an 18 year-review

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Background: Primary immunodeficiencies are group of rare diseases which are difficult to diagnose and manage. Most presenting symptoms were infections from uncommon organisms leading to high morbidity and mortality.

Objective: To evaluate clinical characteristics of pediatric patients with primary immunodeficiencies in Thailand.

Methods: Medical records of all patients diagnosed and treated for primary immunodeficiency in the past 18 years, at Siriraj Hospital, Bangkok, Thailand, were reviewed. Patients with secondary immunodeficiencies and chromosomal abnormalities were excluded.

Results: A total of 85 pediatric patients (58 males and 27 females) were registered. The earliest onset of symptom was in the newborn period. IgG subclasss deficiency was the most common primary immunodeficiency diseases (47%), followed by severe combined imuunodeficiencies (17.6%), specific antibody deficiencies (14%), agammaglobulinemia (7.5%), common variable immunodeficiencies (4.5%) and chronic granulomatous disease (4.5%). The most common presenting symptoms of antibody deficiencies were upper respiratory tract infections (49/56, 87.5%). In patients with T-cell immunodeficiencies, most common presentations were PCP pneumonia (9/15, 60%) with septicemia being the most of common presentations among patients with phagocytic defect (2/4, 50%). There is an increasing trend of early detection of primary immunodeficiency and decreasing diagnostic lag month over the past 18 years. History of death in family is the important information for rapid diagnosis.

Conclusion: Establishment of registry of primary immunodeficiencies may provide the information for early detection and proper treatment to improve the prognosis of these patients in Thailand.

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Paucity of adverse events associated with administration of ivig

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Introduction: Intravenous immunoglobulin (IVIG) is a valuable treatment for many immune-mediated disorders and it is vital management for those with humoral immunodeficiencies. These individuals receive regular, long-term therapy with IVIG. Adverse events associated with administration of IVIG have been reported. In our specialist unit at Westmead, hospital, Sydney, approximately 850 courses of IVIG are administered annually.

Aim: As we have a large experience of IVIG administration, we have performed a retrospective analysis of adverse events (AE) experienced by patients attending our unit.

Methods: Retrospective chart review of all IVIG infusions delivered in the last four years was performed. Factors examined were patient demographics,

diagnosis, previous administration of IVIG infusion and symptoms of AE. IVIG products utilized were Intragam-P (CSL Bioplasma,Melbourne,Australia.), Sandoglobulin (ZLB Bioplasma A G, Bern, Switzerland.), Octagam (Octapharma AG, Lachen, Switzerland.). Patients receiving IVIG were not routinely pre-medicated with either antihistamines or steroids.
Results: In our centre 3,320 infusions of IVIG were given in a 4 year period to 112 patients (46 males - 66 females). 49 of the 112 patients had PID. 63 had various autoimmune conditions or treatment induced hypogammaglobulinemia. 50 of the patients were long standing recipients of IVIG and 62 were naïve to any IVIG products. During the 4 years there were 7 AEs observed. 3 of these patients were naïve to any IVIG products. 4 were long-term recipients. 3 patients had PID and the remainder had autoimmune conditions.
Discussion: Our review showed that the incidence of AEs associated with IVIG infusions is very low (0.4%) for 3,320 infusions, which are similar to figures reported elsewhere. All AEs may have been preventable as three of the patients were naïve to IVIG and may have benefited from pre-medication and three of the reactions occurred when recommended maximum infusion rates were exceeded. The other reaction occurred due to patient delay between infusions. All patients whom experienced AEs have continued on IVIG without any reoccurrence of AEs and only one of these is routinely pre-medicated. Therefore, in conclusion, with due regard to infusion rates, timing between infusions and use of pre-medication IVIG is a safe treatment for humeral immunodeficiences and autoimmune disorders.

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Complement levels in hereditary angioedema

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The diagnosis of hereditary angioedema (HAE) is based on complement tests, however the relationship between the clinical symptoms and the complement levels is poorly studied. In our study we compared complement values in 90 patients with HAE and 212 patients with angioedema of unknown origin. In addition the complement parameters (CH50, C1q, C3, C4, antigenic and functional C1-INH) tested at the time of diagnosis were correlated with age, sex, severity of the disease in 99 patients with HAE type I and 7 patients with type II. Unlikely to the previous findings we have found that out of the complement parameters tested the functional C1-INH had the highest specificity, but the lowest sensitivity in the diagnosis of HAE, while highest specificity was observed with the antigenic C1-INH assay. We did not find correlation between complement levels and age at the time of diagnosis. No association was found between the complement levels and sex or HAE type. We found significant association of baseline functional C1-INH (p=0.0144), and CH50 (p=0.054) levels with the severity of disease. As a conclusion we demonstrated that both antigenic and functional C1-INH is required for the correct diagnosis of HAE and testing of C4 highly validates the results. Determination of funcional C1-INH and C4 may have clinical significance. Regular evaluation of these parameters can be a useful tool in the strategy of long term prophylaxis, however further studies are required to confirm these associations.

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Attacks of hereditary angiooedema (HAE) treated with icatibant in the open-label phase of the fast-2 study

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Background: HAE is characterized by recurrent edematous swellings of the subcutaneous or submucosal tissues. Bradykinin plays an essential role in the development of angioedema. This study was conducted to investigate the effects of the selective bradykinin-B2-receptor-antagonist Icatibant in the treatment of attacks in patients with HAE due to C1-inhibitor deficiency.
Methods: During an open-label extension phase of a randomized, double blind, controlled phase III study (FAST-2) patients with cutaneous and/or abdominal attacks of hereditary angioedema were treated with Icatibant. Within 6 hours after the onset of a moderate/severe attack they received a single injection of 30 mg Icatibant subcutaneously. Symptoms were assessed by patient (VAS, symptom score) and by physician (symptom score global assessment, clinical global impression).
Results: 6 angioedema attacks in 2 patients with HAE were treated with Icatibant. The affected sites were twice the extremities and once the genitalia in Patient #1. Patient #2 experienced two abdominal attacks, while another attack affected the gastrointestinal tract and one extremity. Clinical symptoms of all attacks improved quickly. Time to first improvement of symptom was 10–23 minutes. The duration of complete resolution of symptoms was shorter in abdominal attacks compared to attacks involving the subcutaneous tissues (180–240 min versus 480–720 min). Drug related adverse events and relapses were not experienced. Local skin reactions at the injection site were tolerable and resolved within 4 hours.
Conclusion: Icatibant–which differs from earlier drugs concerning both the mode of action and the method of administration as well–has been found effective and safe in the treatment of angioedema attacks in patients with HAE. The possibility of subcutaneous administration of Icatibant may improve the daily life of patients suffering from HAE. In the present cases repeated use of Icatibant did not result in reduced efficacy. The potential of Icatibant for the treatment of other bradykinin mediated angioedema has to be tested in further studies.

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Leukocyte adhesion deficiency: (LAD-1) Report of two cases

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Leukocyte adhesion deficiency-1 (LAD-1) is a genetic immunodeficiency disease characterized by life-threatening infection results from the mutations in the leukocyte integrin, CD18 molecule with severe and moderate phenotype. We report a case of severe LAD occurred in a 1 month old girl and a case of moderate LAD which was presented with aggressive periodontitis.
Case 1: Clinical features include delayed separation of the umbilical cord, omphalitis, severe bullose and /ulcerative skin eruption which mimic picture of staphylocolal scaled skin syndrom in neonatal period and persistent leukocytosis. The immunological workup showed pathological values of CD11b, CD18 (2.3%, 0.3% respectively). The patient is waiting for BMT now.
Case 2: A 4/5 year old boy with aggressive periodontitis in the primary dentition which pathologic examination showed actinomycosis ginigivits and normal separation of umbilical cord. He had CD11b __ 4/6% and CD18 __ 3/7% in peripheral blood flow- cytometery compatible with moderate phenotype of LAD1. He is receiving antibiotic prophylaxis. Conclusion: Although LDA is a rare form of congenital immuno -deficiency, severe LDA1 should be considered when delayed wound healing and recurrent bacterial skin infections are present in a newborn and moderate LAD1 in aggressive periodontitis with normal wound healing.

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Clinical and molecular characteristics of Thai families with X-linked chronic granulomatous disease

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Department of Pediatrics, Division of Medical Genetics and Metabolism, Bangkok, Thailand.

Rationale: X-linked chronic granulomatous disease (X-CGD) is an immunodeficiency disorder characterized by defective intracellular killing of microorganisms due to neutrophils' inability to generate superoxide ions. Although it is caused by mutations in the same *CYBB* gene, clinical and molecular characteristics vary among different ethnic backgrounds.

Materials and Methods: Two unrelated Thai boys presented with severe persistent pulmonary infections at the age of two months. Their abnormal DHR assays supported the diagnosis of X-CGD. Mutation analysis was performed by polymerase chain reaction (PCR) amplification and sequencing of the entire coding regions of *CYBB*. Mutations identified were confirmed by restriction enzyme analyses.

Results: PCR-sequencing of the entire coding regions of *CYBB* identified nonsense mutations, 271C>T (R91X) in exon 4 and 456T>A (Y152X) in exon 5, in probands of each family. Both of the probands' mothers were found to be carriers.

Conclusion: This observation supports that *CYBB* is the gene responsible for X-CGD across different populations and nonsense mutations are associated with severe phenotypes.

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Immune Reconstitution Inflammatory Syndrome, right sided hemiparesis, in a Thai HIV infected girl with severe immunosuppression

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Background: Inflammatory responses to prior infections due to improved immune responses to latent pathogens with paradoxical clinical worsening observed in HIV-infected patients initiating potent antiretroviral therapy. No case report of cytomegalovirus (CMV) associated CNS IRIS in children.

Methods: We report a case with serious atypical adverse events after initiate ARV.

Results: A 7 years old Thai girl, CDC C due to wasting syndrome, presented with history of fever and oral candidiasis with CD4% 6%, CD4 count 17 cells/ml, viral load 163,000 copies/ml. After 2 weeks cotrimoxazole for PCP prophylaxis, she started antiretroviral therapy (ARV), stavudine, lamivudune and nevirapine, at Chulalongkorn hospital. One week later, she developed right sided hemiparesis without any history of headache, vomiting and loss of consciousness. The neurological exam revealed right sided weakness with motor power grade IV at both upper and lower extremities, normal reflexes and clonus. The differential diagnoses were cerebral toxoplasmosis, brain abscess and tumor. The CT brain with contrast media revealed generalized brain atrophy without mass or ring enhancement. Lumbar puncture was performed. The cerebrospinal fluid (CSF) was colorless and clear with WBC 2 cell/mm3, no RBC, protein 35.9 mg/dl and CSF sugar/ plasma sugar 70/96 mg/dl respectively. CSF bacterial culture was no growth but CSF cytomegalovirus (CMV) was positive through PCR method. MRI brain was shown hypodensity lesion at the right cerebellum. Ophthalmologic examination was normal.After 2 weeks of ARV, her CD4%, CD4 count and viral load were 17%, 202 cells/ml and 440 copies/ml respectively. The diagnosis was Immune Reconstitution Inflammatory Syndrome (IRIS). However, we did not prescribe any steroids because of the bacteremia. During this period, she continued to take ARV as usual. One month after her hospitalization, she was reexamined to be fine with full motor power. After a total of 6 months of ARV, she was fine with CD4% 13%, CD4 count 356 cells/ml, viral load 50 copies/ml.

Conclusion: We reported the CMV IRIS, right hemiparesis, after a week of initiating antiretroviral therapy in an HIV infected girl with severe immunosuppression. The diagnosis of atypical CNS symptoms after initiate ARV with rapidly increased CD4 and decreased viral load should consider as IRIS.

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Usefulness of quantification of C-reactive protein (CRP) for diagnosis of bacterial infection in the febrile neutropenic child

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Background: Diagnosis of bacterial infection in a neutropenic child with fever becomes difficult due to a poor inflammatory response. Currently, a method reliable and fast to identify patients with bacterial infection is not available. CRP has been evaluated in these patients with controversial results.

Objective: To determine the sensitivity, specificity, predictive values and likelihood ratios of CRP for diagnosis of bacterial infection in neutropenic children with fever.

Design: Cross-sectional study.

Methods: We included patients less than 16 years of age with fever and severe neutropenia (<500 AN/mm3). Blood cultures and CRP were taken in the initial evaluation. All of them, had empirical antimicrobial treatment; their evolution was followed up until discharge. Blood cultures were processed by the automated system BACT-alert; CRP by nephelometry. Patients were classified in 4 groups: Group I: Clinical and bacteriological infection, Group II: Infection clinically defined, Group III: Fever due other causes than infection and Group IV: Patients with cancer, neutropenia without fever. Diagnostic test analysis, receiver operating curves (ROC) and likelihood ratios were performed.

Results: One hundred twenty seven episodes were included from 113 subjects. Leukemia was the most frequent disease (61%). Twenty nine, 47, 20 and 31 episodes were included in groups I, II, III and IV respectively. We found microbiological isolation in 29 episodes, Staphylococcus aureus and Escherichia coli were the most common isolated germs (27.6% and 17.2%). Median for CRP levels (IQR) was of 282 mg/L (174–385) in group I; 205 mg/L (119–267) in group II; 27,3 mg/L (12.3–55) in group III and 5,1 mg/L (2.4–13) in group IV ($p<0.001$). By COR curves a CRP level higher than 60 mg/L showed a sensitivity (S) of 94%, specificity (E) 94%, positive PV (PPV) 96% and negative PV (VPN) 92% when we compared groups I and II vs. III; at this cut level, likelihood ratio for a positive result was 15,6 and 0,06 for negative result. Among those patients with diagnosis of solid tumor, S, E, PPV and NPV were of 96, 100, 100 and 95%. We found that none patient with neutropenia, fever with negative blood cultures and without focal infection, with a CRP level lower than 30 mg/L had not bacterial infection. All patients with CRP levels higher than 100 mg/L were infected.

Conclusion: CRP is a useful, fast and economic test to identify bacterial infection in the neutropenic patient with cancer and fever.

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Hyperimmunoglobulinemia E (Job) syndrome

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Background: The hyperimmunoglobulinemia E (Job) syndrome is a rare immunodeficiency disorder. The only one patient is present in Kharkiv Region.

Methods: The patient was observed by traditional clinical and paraclinical methods of investigation.
Results: Clinical signs: 1) Recurrent furunculosis and episodes of skin cold staphylococcal abscesses. An onset of streptostaphylodermia is 2 month age. It was the first clinical manifestation of disease. A frequency of exacerbation is 2–4 times per year. 2) Recurrent bronchitis since 5 month. 3) Recurrent pneumonia complicated pleuritis and pneumatocele since 4 year old. The child had 1–3 pneumonias per year. The child was operated at the age 4 year - lobectomy of upper right lung lobe; at the age 8 year - lobectomy of upper left lung lobe; and at the age 9 year - toracotomy and sewing up of the broncho - pleura fistula of left lung. 4) Recurrent abscesses in abdominal cavity since 8 year. The child was operated many times - at 8 year due to acute appendicitis complicated peritonitis; at 10 year - a huge abscesses of big omentum; at 11 year - an abscess in a region of afteroperation scar; in 3 month - an abscess in left underdiaphragm region and an abscess of upper pole of spleen. 5) Presence atopic dermatitis since 3 month. The eczematous skin eruption does not have a seasonal variation and is present, to some degree, at all times. 6) Presence skeleton anomalies: scoliosis, generalized hyperextensibility of joints, facial abnormalities, retention of 4 primary teeth with unresorbed roots, absent 2 lower premolaries, 1 upper premolaries, 2 lower and 2 upper molaris. Lab Findings: high eosinophilia (7 – 24 %), lymphocytosis (till 42 %), than - neutrophilia (till 86 %), shift to the left (band neutrophils 6 – 11 %), eleveted ESR (25 – 43 mm/hour), high level IgE (60498 IU ml). Imaging Studies: dental abnormalities - delay of primary teeth shedding owing to lack of root resorption, absent of some permanent teeth (X-ray); thin-walled cysts in both lungs (X-ray); cysts in the liver and in the lungs (Ultrasonic, Computer tomography). The patient was treated by prolonged course of antibacterial therapy, fluconazole for mucocutaneous candidiasis, onychomycosis and topical steroids for eczematous dermatitis.
Conclusion: This clinical case gives us an experience in observation and treatment of children suffering from a rare primary immunodeficiency disorder.

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The evaluation of clinical and paraclinical in twenty CVID patients and detection Of IL-2 as a significant cellular function in these by invitro way

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Subjective: Common variable immunodeficiency is the primary immunodeficiency disease. CVID is a heterogeneous group of immunologic disorder of un known etiology, characterized by impaired antibody responses, associated with number and function cells defect including lymphopenia,anergy,impaired lymphocyte proliferation and deficit cytokine secretion that the most important of them is IL-2.most prevalent of the
Objective: The goal of this study was evaluation of the clinical and paraclinical characterized 20 CVID patients,interlukine 2 production and compaired these to healthy control group.
Methods and Materials: We done a randomized cross sectional study in 20 CVID patients from department of immunology and allergy in children medical center and 10 healthy control. The evaluation of the patients was done by questioner and peripheral blood mononuclear cells of the twp groups were cultured with PHA and supernatant were collected for quantitation of IL-2 by ELISA.
Results: Mean of age the patients was 15.9+/−11.23 and distribution of gender was 9 female and 11 male. There were high frequency hospitalization in 80% of cases (over the two time).The most frequent of infection was respiratory tract infection. There were opportunistic infection in 5–10% of them, autoimmunity in 40% of cases,un response DTH in 65% of cases and IL-2 production was zero in15 patients and the 5 patients had level of IL-2 lower than corresponding levels in the healthy control (P value<0.005).
Conclusion: In our the study, all of the patients showed that T cells of them exhibit deficient production of IL-2 and manifestations of this deficiency were high frequency of autoimmunity,granolumatose disease, bronchectasia and recurrent infection in spite of given monthly IVIg in patients.

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A 3 years old female with SCID T(−)B(−)Nk(+) treated with halogenic bone marrow transplantation. Case report

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A 3 years old female is the second of two children born to non-consanguineous mexican parents after an uncomplicated, term pregnancy. Birth weight was 3900 g, length 53 cm.

She had breast feed until 2 months of age. She didn't receive immunizations. She has history of a 9 years old brother who suffered totipotent cell transplantation because severe immunodeficiency. He cursed a neurologic infection and died. At age 45 days she had abundant, liquids, and blood stools. She received ceftriaxone treatment with partial improvement. After that a stool cultive test was positive for campylobacter yeyuni so she was treated with claritromicyn for 14 days. She also had oral and esophageal candidiasis treated with miconazol. Because the brother with immunodeficiency, when she arrived to our hospital the following studies were done: cd3 0.2%, cd 20/19 0%, cd4 0.1%, cd8 0.1% cd 53 83%, igm 4 mg/dl (20–40) igg 415 mg/dl (310–852), iga 6.6 mg/dl (3.5–67). A severe combined immunodeficiency diagnosis was made with t (−) b (−) nk (+) features. We supposed deficiency in enzimes of receptor's recombination. We restituted totipotent cells with halogenic bone marrow transplantation. The hla compatibility was:

patient: a68 (28) y a2, b39 (16) y b61 (40), dr4 y dr 11(5), dq8 (3), dq7 (3) patient's father: a68 (28) y a2, b61 (40) homozigous, dr4 y dr 11(5), dq8 (3) y dq7 (3).
High resolution study for dr
patient: drb1 0407
patient's father: drb1 0407.

We decided to admistered only partial hidrolized formula because the posible risk to adquired infections by breast feeding. She was treated with immunosupresor as acondicionated treatment with busulfan 4 mgkgday 4 days, day −8 to −6, cyclophosphamide 60 mgkgday day −5 to −4. In september 19 2003, our patient received hallogenic bone marrow trasplantation with prior medication with h1 blockers and steroids. We adminitered 120 ml of aferesis products with a total count of 189,000 × 10 3. 98.5% mononuclears.

The patient presented host againts disease with papules, petechias in abdomen. She was treated with metotrexate, ciclosphorine and methilprednisolone. She had citomegalovirus blood culture test positive and was treated succesfully with ganciclovir. Nowadays she has normal blood citology, immunoglobulin levels, lymphocyte subpopulations and she has completed her immunization schedule two and a half years after the stem cell transfusion.

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Is staphylococcal alpha-toxin a critical pathogenetic factor of atopic dermatitis in highly atopic individuals?

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Background: *Staphylococcus aureus (S. aureus)* is a well known trigger factor of atopic dermatitis (AD). Besides staphylococcal superantigens alpha-toxin (α-t) which is produced by one third of skin-colonizing S. aureus strains in AD may influence the cutaneous inflammation. α-t can be detected both in the upper epidermis and in the dermis of AD patients. Sublytic concentrations of α-t have been shown to induce T-cell proliferation and secretion of T-cell cytokines.

Objective: To explore the association of sensitization to inhalant allergens and α-t producing skin colonizing S. aureus in AD. Methods: We investigated 127 adult patients with AD according to their skin colonization after treatment with antiinflammatory and antiseptic substances, medical history, severity of AD and sensitization to different allergens.

Results: 48 out of 127 patients where colonized with S. aureus. S. aureus colonized patients suffered from a more severe AD (SCORAD: S. aureus positive patients: 46, S. aureus negative patients: 33) and showed a higher sensitization level to different inhalant allergens. Surprisingly, they also suffered more frequently from allergic asthma (S. aureus positive patients: 69%, S. aureus negative patients: 44%). 30 of the 48 S. aureus skin-colonizing strains produced α-t. The severity of AD was similar in patients being colonized with α-t negative S. aureus (α-t- pts) compared to patients with α-t producers (α-t+ pts). α-t+ pts had a significally higher specific IgE to birch pollen (median: α-t+ pts: 100kU/l, α-t- pts: 32kU/l) and a trend of higher total IgE values (median: α-t+ pts: 6849kU/l, α-t- pts: 2215kU/l) and specific IgE to timothy grass pollen (median: α-t+ pts: 80kU/l, α-t- pts: 12kU/l).

Conclusion: Following topical treatment the colonization rate was lower (38%) than expected. But there was a high rate of patients who were skin colonized with α-t producing S. aureus. Colonization with S. aureus was associated with a higher severity of atopic dermatitis, higher sensitization, and a higher frequency of allergic asthma. Cutaneous colonization with α-t producing S. aureus was associated with a higher sensitization to saisonal inhalant allergens in AD. If this is a consequence of direct chronic cutaneous T-cell stimulation by sublytic doses of α-toxin penetrating constantly into the skin needs further investigation.

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Trichophyton allergy: review of 98 cases

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Background: The association of mycosis or onicomycosis by dermatophyte fungi and allergic diseases including urticaria/angioedema, rhinitis and asthma has been described for over 70 years but large series of confirmed cases have not been published.

Methods: From 9087 patients seen in the last few years 98 (1,07%) 20 to 66, 61 males (62,2%), 37 females (37,8%) had a clinical history and previous or actual visible skin or nails infection and positive tests to Tricophyton. Skin tests have been done in all the patients by prick in a first step and by intradermal with 1/1000 and 1/100 dilution in a second step. Specific IgE has been looked by UNICAP for Tricophyton rubrum in 78 cases. Treatment with antifungal itraconazol has been tried as a first option in all the patients. When allergic symptoms subsisted specific immunotherapy by parenteric route has been started with a slow release vaccine during a minimal period of 3 years.

Results: Fungal infection presentation was tinea pedis in 66 patients (67,3%), onicomycosis in 36 (36,7%), tinea cruris in 27 (27,6%) and remaining pruritus after clear infection in 3 (3,0%). Dual infections were present in 33 patients (34,4%). Total of clinical mycosis 129. Allergic disease: urticaria/angioedema in 64 patients (65,3%), rhinitis in 14 (14,3%), asthma plus rhinitis in 11 (11,2%), eczema in 8 (8,2%) and conjunctivitis in 1 (1,0%). Fifteen patients (15,3%) all rhinitic had also positive skin tests to house dust mites. Prick tests for Tricophyton were positive in 61 patients (62.2%). Intradermal tests were always positive. Specific IgE was positive in 52 (66,6%) cases with a range of 0,50 to more than 100 Ku/l. Treatment with antifungal cleared allergic disease only in 20 (20,4%). Specific immunotherapy has been tried in 60 patients with very good results (complete clearing) in 23 (38,3%), good in 36 (43,3%) and moderate or nil in 11 (9,4%).

Conclusion: Tricophyton IgE mediated allergy play a role in the pathogenesis of Urticaria/angioedena, asthma and rhinitis in most patients. Fungal skin and nails infection must be searched, skin tests and specific IgE used for diagnosis. Sensitivity and specificity of skin prick tests and specific IgE are roughly comparable. Specific immunotherapy must be tried if anti fungal therapy failed with good results in most cases.

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Profile of inflammatory cell in erythema nodosum leprosum patients, treated with minocycline-prednisone combination drugs and prednisone

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Background: A clinical trial study was conducted with " pre-post treatment" design, to know effectiveness minocycline-prednison combination drugs and prednisone only in the erythema nodosum leprosum (ENL) patients by profile of cells inflammation.

Methods: Sixty ENL patients released from treatment (RFT) were included in this study. They were devided into 2 groups, 30 patients were treated with minocycline-prednison combination drugs (group A) and the other 30 patients were treated with prednisone-placebo (group B). To evaluate result this study, we performed histopathology examination by skin biopsy, haematology test by peripheral blood sample that obtained from cubital venous puncture, and the changes of clinical feature.

Results: The results, that the ENL patients group A were shown more improve histopathologically ($p<0.05$) haematology finding not showed difference between 2 groups ($p>0.05$). There was more improve change clinical feature in the patients get combination therapy.

Conclusion: Combination therapy, prednisone-minocycline, better than just prednisone only in ENL patients, so that it may be alternative drug to reduce steroid dependency in the ENL patients.

Key Words: neutrophil, ENL, minocycline, prednison

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Level of bacterial IgG and IgM antibodies in multiple sclerosis patients

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Multiple sclerosis is one of demyelinating diseases based pathophysiologically on changes in vasculo-myelin system. Immunological basis of these diseases is failure of immunological tolerance for brain antigens with development of autoimmune cellular reactions. Important role in this failure and development of demyelinating process is attributed to bacteria possessing antigenic determinants common with myelin basic protein (MBP) molecule.

Therefore we performed in multiple sclerosis patients study of IgG and IgM antibodies against Clebsiella pneumonia, E. coli, St. aureus, H. influenzae, and pertussis anatoxin.

The study was performed with serum from 78 patients with different types of multiple sclerosis. Titer of antibodies against Clebsiella pneumonia, E. coli, St. aureus, H. influenzae, and pertussis anatoxin was detected by enzyme immunoassay. Results were expressed as 1–5 classes of reaction.

The study showed that in recurrent type of multiple sclerosis exacerbation is accompanied by increase of IgM antibodies level (to 4–5 class) for all abovementioned bacteria. Remission is marked by lowering of IgM level compared to exacerbation one, to 2–3 class, and by increase of IgG level to 3–4 class. In progressive type of multiple sclerosis level of IgG as well as IgM antibodies increased progressively (to 3–5 class). If course of multiple sclerosis lasted more than 5 years, levels of IgG and IgM antibodies were the highest (mostly 4–5 class). Based on data obtained, we concluded, that degree of manifestation of antibacterial immunity reflects the severity of multiple sclerosis and can be considered as negative prognostic factor.

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Local cytokine status and clinical morphological parameters of chronic viral c hepatitis

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The current research is aimed at investigating the levels of local cytokines in liver histology examination samples of the CCH patients taking into account clinical morphological parameters as well as biochemistry and virology investigations. Cytokine investigation in homogenizers of liver histology examination samples was performed in the period of CCH clinical manifestation accompanied with cytolysis. As a tool of control the samples of the hepatic tissue belonging to 5 donors who did not have any chronic diseases and markers of being infected with viruses of parenteral hepatitis were used to investigate cytokine levels in homogenizers of liver histology examination samples. In the process of investigation of liver histology examination sample homogenizers in CCH patients it was established that the they have the authentic increase of IL-1α, IL-4, TNF-α proportion and decrease of IL-2, IFN-γ proportion. The considerable fluctuation in proportion levels of each of them from the minimal to the maximal indices was noted and the total distribution differed from the normal one, which hampered the adequate assessment of the investigated indices. The patients were divided into two groups that differed from each other by the median of the low (V0,25) and upper (V0,75) percentiles of the quantitative contents of the local cytokin levels. The first group whose levels of practically all investigated cytokines in liver histology examination samples were authentically different from those of the second group. IL-4 proportion in the second group nearly twice exceeded its index in the first group. The increase of TNF-α level in liver histology examination samples of CCH patients of the second group six times as high as that of the first group was noted. The considerable decrease of the local proportion of IL-2 and IFN-ã in the second group (0,6 ± 0,02 pg/ml versus 16,0 ± 0,6 pg/ml, $p < 0,001$ and 4,2 ± 0,65 pg/ml versus 18,9 ± 2,7 pg/ml, $p < 0,0001$ correspondingly) was noted. No difference in IL-1á levels in investigated groups was noted ($p > 0,05$). In accordance with the local cytokine levels along the median of the extreme percentiles (V0,25 and V0,75) two CVH patient groups were singled out, the groups having the authentic difference in clinical, biochemical and morphological parameters.

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Modern representation of immunopathogenesis of hemorrhagic fever with renal syndrome

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HFRS is one of the clinical forms of Hantavirus infection that is widely spread on Eurasian continent. In Europe HFRS is predominantly associated with Puumala, Dobrava - Belgrad Hantaviruses (HV), in Asia - with Hantaan, Amur, Seoul. Another clinical form, which is common for the American continent is Hantavirus Pulmonary Syndrome (HPS), associated with Sin - Nombre, Andes serotypes. On the south of Far East region, where three serotypes of HV are collected, the similarity of the main pathogenical and clinical aspects of HFRS and HPS were revealed. It is established that in the pathogenesis the important role plays either HV, as the initiator of the process, or the reactions of cellular immunity. The more virulent HV (Sin - Nombre, Andes, Hantaan, Amur) and lass virulent (Puumala, Seoul) are known. The studying of immunopathogenesis in HFRS patients revealed the dependence of characteristics of immune response from the serotype of HV. On the onset of the disease the level of misbalance of serum cytokines (high levels of IFN - α, IL -1α, IL12p70, IL12p40, IL-8, IL-10 and low levels of IFN-γ) and also the number of cytotoxic lymphocytes CD8 correlated with the severity of clinical symptoms and the serotype of HV. In HFRS, associated with Hantaan, Amur serotypes severe and complicated manifestations with hemorrhagic syndrome, hyperhydration of kidneys, respiratory tract, liver tissue and multiorganic insufficiency. Severe forms of Seoul - infection were less common and the characteristics of immune and cytokine status were not changed a lot. The studying of the levels of cytokines and metabolites of nitric oxide in urine and exhaled air condensates showed the absence of correlation of their production locally and systemically. That fact reveals the independent synthesis of the immune response mediators and allows supposing the respiratory tract, as the kidneys to be the target - organ of HV. Viremia begins after the respiratory infection of alveolar macrophages and then the central cells of pathogenesis - endothelial cells of the lungs, kidneys and other organs. So, the results of the study of cytokines in different biologic substrates (serum, urine, exhaled air condensates) in the onset of the disease showed the prevalence of immune reactions in the pathogenesis of HFRS without any dependence with the Hantavirus serotype.

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The comparison of allergenicity between L3ESP and L4ESP and the cytokine production profile in experimental infection of rats with Anisakis simplex

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Background: *Anisakis simplex* is the marine parasite which belongs to ascaroidea. This worm normally live within marine mammals, however frequently induces allergy in the infection of mammals on land by third stage larvae (L3) carried by marine fishes. When L3 infect mammals, these worms develop into fourth stage larvae (L4). Each state larva produces different excretory-secretory products (ESP) in main protein constituents. This investigation compared ESP from L3 (L3ESP) with L4ESP in antigenicity and allergenicity to develop on efficient method to diagnose the allergy by L3, and analyzed cytokine profile in reinfection of L3, which is critical period in allergy development.

Methods: The kinetics of specific antibody production of sera harvested from rats infected by L3 was analyzed by indirect ELISA using either L3ESP or L4ESP as antigen immobilized on ELISA plate. The cytokine kinetics of

IL-4 and IFN-r was analyzed with rat sera harvested for 5weeks from L3 reinfection.

Results: The kinetics of antibody production showed that specific antibody level against L4ESP was higher in antigenicity than the level against L3ESP in IgG1 IgG2b IgG2c and IgM, and very similar in IgG2a. However, the latter had evidently higher allergenicity than the former in specific IgE level. In L3 reinfection sera, the level of IL-4 was relatively constantly maintained. In contrast, that of IFN-¥γ; was decreased continuously.

Conclusion: These results indicated that L3ESP be better than L4ESP for allergic state analysis and Th2 cytokine be comparatively dominant in *Anisakis simplex* larva reinfection.

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Effects of jungle honey on immune functions

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Introduction: Jungle honey (JH) is collected from timber and blossom by wild honey bees that live in the tropical forest of Nigeria. This is used on traditional medicine for cold, skin inflammation and burn wound as well as health care. However, the effect of Jungle honey on immunomodulatory activity is not yet clearly. We have investigated the effect of Jungle honey on immune system and anti-tumor activity in mice.

Methods: JH was provided by Nihon origins. JH was dissolved with distilled water, and freezed dry, and then adjusted with PBS at each concentration. JH was fractionized to Fraction (Fr.) 1, 2, 3, 4 and 5 by HPLC size chromatography. Female C57BL/6 mice were injected with JH at dose of 1mg/mouse/day, 7 times intra-peritoneal. After 7 times injections, peritoneal cells (PC) were obtained by peritoneal lavage with PBS. Total numbers of PC were counted with homocytometor. Cell differentials were observed by microscope after Giemsa stain. Expressions of surface antigens (CD3, CD11b, CD19, NK, and Gr-1) on PC were analyzed by FACS. H_2O_2 production of PC was assayed by FACS using DCFH-DA. Chemotactic assay was analyzed by TAXIScanTM using neutrophils from peripheral blood of human or guinea pig. IL-1β mRNA expression of PC were analyzed by RT-PCR. Anti-tumor activity was assayed by using Lewis Lung Carcinoma/2.

Results: Total numbers of PC were increased in JH-injected mice compared with control mice. In Dot Plot analysis by FACS, neutrophils were increased in JH-injected mice. Percent of Gr-1 surface antigen positive cells and intensity of Gr-1 antigen expression were also increased and H_2O_2 production of PC was increased by JH. Chemotactic activity for Neutrophil from human or guinea pig was expressed at concentration of 1, 5mg/ml or 1, 5, 10mg/ml of JH. IL-1β mRNA expression of PC was increased by JH or Fr.2. Molecular weight of Fr.2 was approximately 261. Inhibitor of tumor growth was showed by JH.

Conclusion: Chemotactic activity for neutrophils, increase of neutrophil counts and H_2O_2 production of PC by JH may prevent bacterial and viral infections.

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A case report of 11-month-old boy infant with Toxocara canis eosinophilic meningitis

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An 11-month-old boy infant was referred due to prolonged fever for 4 weeks, drowsiness and bulging at the anterior fontanelle. The clinical was no response with high dose cefotaxime for 2 weeks that treated as bacterial meningitis. Neurological examination revealed meningeal irritation and bulging at the anterior fontanelle. Marked increase of absolute eosinophils count (1,893/mm^3) was presented. A lumbar puncture showed 270 leukocytes/microliters with 60% of eosinophils; protein 50 mg/dL; CSF culture was no growth and negative PCR for TB. The immunoblotting assay was positive for *Toxocara canis* in both serum and in CSF. The CT scan of brain showed communicating hydrocephalus. The patient was close contact with cats with poor sanitation. A diagnosis of eosinophilic meningitis due to *Toxocara canis* was made and started treatment with albendazole 200mg oral twice a day for 4 weeks, prednisolone 2 mg/kg/day and tape off within 8 weeks. The clinical was improved and discharged at 5 days after start both medications. Follow up monthly for clinical symptom and monitored for absolute eosinophil count. No abnormal neurological symptoms were detected. The parents had got rid of the cats out of the household area. Serum absolute eosinophil count and lumbar puncture results turn to normal within 11 months after treatment. The serological follow up 8 weeks after treatment for *Toxocara canis* was positive in both serum and in CSF by the use of immunoblotting assay but decrease in intensity. He had normal growth and development. Then long term follow up was done and aware for reinfection in this patient. Reinvestigation for immunoblotting assay and follow titer for *Toxocara canis* will be done.

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Activity of allium ascalonicum (shallot) and myrtus communis extract as two novel antibacterial agents against acne vulgaris

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Background: Acne vulgaris is a common skin disease. Its etiology and pathogenesis is not well known, and treatment is not satisfactory, therefore study for novel drugs to replace to common drugs will be necessary.

Methods: From 136 patients who suffered from acne lesions, swabs were obtained. Each swab was cultured on 2 blood agar plates and stored aerobically and anaerobically condition. Sensitivity test from 2 herbal extracts against bacterial isolations done by disk diffusion method, and were compared with tetracycline disks.

Results: From 176 bacterial strains isolations, 113 (64.2%) Coagulase Negative Staphylococci (CONS), 52 strains (29.5%) *Propionibacteria spp.*, 7 strains(4%) *Diphtheroids*, 2 strains (1.13%) *E. coli* and 2 strains (1.13%) *Streptococci spp.* were isolated. 53% of CONS and 10% of *Propionibacterium* isolates were resistant to tetracycline, whereas only 8% of CONS and14% of them was resistance to Shallot and Myrtus communis water extract respectively. No *Propionibacterium* strains and only 4% of *Propionibacterium* were resistant to shallot and Myrtus communis water extract respectively. Also 11% of CONS Isolates were resistant to shallot alcohol extract and 12% of them were resistant to the Myrtus communis alcohol extract. Two percent of *Propionibacterium* strains were resistant to shallot alcohol extract and 3% of them were resistant to the Mmyrtus communis alcohol extract.

Conclusion: Our finding revealed that bacterial agents of acne vulgaris are highly resistant to tetracycline and this antibiotic is not efficient for treatment of the disease. It is suggest that these herbal extracts may have potential for acne treatment.

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Galactomannan antigen detection in the diagnosis of invasive pulmonary aspergillus

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Objective: The invasive aspergillus occurs almost exclusively in immunocompromised host. Aspergillus fumigatus is now the leading cause of infectious mortality in many hematology bone marrow and allograft transplantation. Galactomannan is hetropolysaccharid present in the cell wall of aspergillus species. A rapid and important method for invasive aspergillus diagnostic could be the circulating galactomannan aspergillus (GM) measurement in the serum and BAL (Bronchoalveolar lavage) samples of the patients. The aim of this study was determined the value of galactomannan detection in serum to diagnosis of invasive aspergillus.

Methods and Patients: Fifty three patients (with recurrent pulmonary infection suspected to aspergillus) were included in our study group. None of the selected patients had received any anti fungal therapy prior to the study. Direct microscopy and fungal culture of BAL (Bronchoalveolar lavage) was done and galactomannan estimation in serum and BAL were measured.

Results: 32 patients (>60 %) were considered galactomannan test positive from a total 53 cases (30 male and 23 female) and 28 of these patients were isolated aspergillus in microscopy and culture.

Conclusion: The GM EIA assay had greater sensitivity than culture and microcopy in detection of aspergillus spp in BAL fluid in experimentally induced invasive pulmonary aspergillus. GM positivity also allowed the anticipation of invasive aspergillus diagnosis (from 3 to30 day before mycological culture).

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Revisiting the hygiene hypothesis: Cross-reactivity studies between house dust mites and Ascaris lumbricoides

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Background: The causal relationship between allergy and ascariasis remains a matter of controversy. Despite growing evidence for protective immunity exerted by helminth infections against allergic diseases, the molecular and immunological mechanisms involved needs to be elucidated. In this study, cross-reactivity of allergens from house dust mites (HDM) *Blomia tropicalis* (Blo t), *Dermatophagoides pteronyssinus* (Der p), and *D. farinae* (Der f) with antigens from *Ascaris lumbricoides* (Asc l) was determined.

Methods: Enzyme-linked Immunosorbent Assay was performed to determine the allergenicity of HDM extracts and antigenicity of Asc l extracts to allergic (n=100), ascariasis (n=60), and healthy control (n=100) patients. Inhibition assays and Western blot analyses were done using positive sera from allergic (n=15) and ascariasis (n=15). Specific IgE levels of allergic (n=50) and ascariasis patients (n=50) to a recombinant paramyosin peptide (Blo t 11-fD) was determined.

Results: The allergenicity of HDM extracts was significantly higher among allergic patients (Blo t=81, Der p=72, and Der p=80% positive reactions) than ascariasis patients (20, 20, and 28.3%, respectively) while the antigenicity of Asc l extracts among allergic patients (70%) was significantly lower than ascariasis patients (87%). Cross-inhibition assay showed that Asc l antigens can inhibit up to 92% of the IgE reactivity of allergic patients to HDM allergens while up to 54% of the IgE reactivity of ascariasis patients to Asc l antigens was inhibited by HDM allergens. Western blot analysis showed multiple sensitizations of allergic patients to HDM allergens with molecular weights ranging from 14–240 kDa and of ascariasis patients to Asc l antigens ranging from 15–250 kDa. Positive reactions to rBlo t 11-fD was observed among allergic (80%) and ascariasis (46%) patients.

Conclusion: Multiple cross-reactive antigens are present in HDM and Al extracts and may play a role in the complex immunological relationship between allergy and ascariasis. These antigens might share homology or similarity in epitope recognition sites and must be identified in future studies. The potential role of paramyosin as a specific cross-reactive allergen present in HDMs and *A. lumbricoides* was described. These findings provide support for the Hygiene Hypothesis and may serve as basis for novel forms of treatment and diagnosis of allergic diseases and helminthic infections.

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Functional model of in vitro infection of murine macrophages by F. tularensis LVS

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Background: In this work, we focused on mutual interactions between intracellular parasitic bacteria *Francisella tularensis* LVS and murine macrophage-like cell line J774.2, particularly on immunobiological effect of infection. We established a functional model of infection and stimulation by murine interferon gamma (IFNγ) and lipopolysaccharide (LPS). This model allows us to define basic principles of pathogenesis of infection on cellular level. We are able to define precausions of effective macrophage activation leading to subsequent infection elimination.

Methods: We followed up the expression of specific macrophages surface markers: CD 86, CD 54 (ICAM-1), CD 16/32 (FcgIII/IIR) and CD 25, including specific isotypic controls. The phenotypic changes expressed as mean fluorescence intensity (MFI) or percentage of positivity, respectively, were evaluated by flow cytometry. Morphologic changes were documented by immunofluorescence microscopy. Murine macrophage-like cells (J774.2) were incubated in cultivation flasks (2x106 cells/10ml of medium Dulbecco's MEM with Glutamax-1 with 10% BSA). The cells were activated with 5, 10,50 ng of LPS per 1ml of medium or with 10, 100, 1 000, 10 000 I.U. of IFNγ per 1ml of medium respectively and infected by *F. tularensis* LVS with multiplication of infection 1:100 in particular time schemes.

Results: Infection of *F. tularensis* does not result in any activation of host cells, as seen from stable phenotype profile of infected cells in time. In contrary, stimulation of J774.2 cells by IFNγ or LPS results in predictable, time and dose dependent phenotypic changes. The phenotype profile of IFNγ activated cells is characteristic. These cells display a harmonic increase in CD86 and CD16/32 surface expression in time. In contrary, activation by LPS results in isolated CD16/32 expression elevation without increase in CD86 expression. Moreover, prior *F. tularensis* infection of macrophages prevents subsequent activation, especially by LPS. We demonstrate an essential role of IFNγ in infection control. Minimal concentration of IFNγ resulting in active infection elimination is 1000 IU/ml.

Conclusion: We assume, that *F. tularensis* infection is able to interfere with intracellular signaling triggered especially by LPS, less significantly by IFNγ. This interference leads to immunosupression and seems to be the efficient escape mechanism of *F. tularensis.*

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Effects of cigarette smoke exposure on immune functions in alveolar macrophage

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Purpose: Cigarette smoke is inhaled into lung. In the lung, AM are known to play an important role in lung immune system. In previous studies, we

reported the inhibition of antibody production by alveolar macrophages (AM) from cigarette smoke (CS) exposed mice. However, the mechanism of immune suppression by CS on AM functions is not clearly understood. Therefore, we investigated effects of CS on phagocytosis and antibody production, expression of surface antigens, IL-1βmRNA in AM associated with immune functions.

Methods: Female C57BL/6 mice were exposed to 20 cigarettes /day during 10 days. After 10 days, AM were obtained by bronchoalveolar lavage (BAL). Phagocytosis activity was analyzed by FACS using FITC labeled seep red blood cell (SRBC). Expression of surface antigens (Class II, B7.1, CD11b, CD16/32, CD14, TLR-2) on AM were analyzed by FACS. IL-1βmRNA expression of AM was analyzed by RT-PCR. Antibody production was analyzed by plaque forming cell (PFC) assay using SRBC antigen.

Results: Phagocytosis of AM was significantly decreased in smoked mice (SM) compared with non-smoked mice (NSM). Surface antigens positive cells in AM were decreased in SM compared with NSM. IL-1βmRNA expression of LPS non-stimulated AM was increased, while in case of LPS stimulated AM was decreased in SM compared with NSM. Antibody production was significantly decreased by AM at induction phase, but not expression phase in SM compared with NSM.

Conclusion: These results suggest that the inhibition of antibody production is caused by the inhibition of phagocytosis and expression of surface antigens in AM. Such inhibition of AM functions may be increased the risk of bacterial and virus infections.

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Analysis of signaling pathways involved in peptidoglycan-induced RANTES production by murine Langerhans cells

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Background: Atopic dermatitis (AD) is a chronic inflammatory skin disease with immunopathologic features that vary depending on the duration of the lesion. The lesional skin of AD patients shows an increased number of eosinophils in the dermis and superficial *Staphylococcus aureus* colonization. Our previous study showed that percutaneous application of peptidoglycan (PEG) from S. aureus induced eosinophil infiltration in murine skin through RANTES production by epidermal Langerhans cells (LCs). Although it is well known that PEG is an agonist of Toll-like receptor (TLR)-2, it is unclear whether other TLR agonists are able to induce RANTES production by LCs. In this study, therefore, RANTES production by murine LCs in response to different TLR stimuli, and the signaling pathways involved, were investigated.

Methods: PEG was applied to barrier-disrupted abdominal skin of mice every 5 days. Twenty days later, eosinophils in the abdominal skin were detected. LCs were purified from epidermal cells by the panning method using anti-IA^d monoclonal antibody. RANTES production in the skin and by the cultured LCs was investigated by RT-PCR, immunohistologic analysis and ELISA. Analysis of the signaling pathways responsible for RANTES production by LCs was performed by ELISA using N-acetyl-L-cysteine, SP600125, PD98059 and SB203580, which are specific inhibitors of NF-e?B activation, JNK, ERK and p38 MAPK, respectively, and was finally confirmed by Western blot analysis.

Results: The results of RT-PCR and ELISA showed that PEG, poly(I:C), LPS and CpG DNA, whose signals are transduced through TLR-2, TLR-3, TLR-4 and TLR-9, respectively, strongly induced the production of RANTES. Although treatment with an inhibitor of NF-e?B activation inhibited PEG-, poly(I:C)-, LPS-and CpG DNA-induced RANTES production, treatment with a JNK inhibitor did not inhibit CpG DNA-induced RANTES production. Furthermore, treatment with a p38 MAPK inhibitor affected only PEG-and LPS-induced RANTES production, and the inhibition of RANTES production was correlated with that of p38 MAPK phosphorylation.

Conclusion: These results suggest that the signaling pathways involved in RANTES production by murine epidermal LCs in response to different TLR stimuli are not necessarily the same, and that inhibition of p38 MAPK may be a more specific therapeutic strategy for eosinophilic inflammation in AD patients with *S. aureus* colonization.

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Eczema herpeticatum recidivans

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Purpose: Eczema herpeticum (EH) is defined as disseminated Herpes simpelx Virus (HSV) infection of an eczematous skin disease, which in clinical reality is almost exclusively atopic dermatitis (AD). High serum IgE and early onset of the underlying AD are established risk factors. Many patients have one episode of EH only (EHP), whereas some patients suffer from securrent EH (EHR). This multicenter study was performed to identify clinical risk factors for EHR.

Methods: A total of 217 patients with a serum IgE of 3450 ± 6821 kU/l and a SCORAD of 44 ± 18, diagnosed from 1996 to 2006 with EH, were included in the analysis. Intrinsic AD was diagnosed in one patient only.

Results and Discussion: Herpes blisters were present within the eczematous skin lesions only, indicating a need for clinically active AD lesions to develop EH. The frequency of EHR (26%) was markedly higher than reported in previous decades, which demonstrates a change of the clinical disease spectrum. The EHR patients showed a higher serum IgE, more eosinophils and an earlier onset of the underlying AD as compared to EHP. In addition, they used steroid ointment less frequently.

Conclusion: In conclusion, EHR patients exhibit the established risk factors for EH (severe, uncontrolled AD) in a more pronounced manner and may be described as being even 'more atopic' than EHP patients.

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Shallomin, a novel antimicrobial agent against bacteria and fungi

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Background: Studies in past decade confirm that the growth of both gram positive and gram negative bacteria, yeasts and mold can be inhibited by garlic, onion, cinnamon, cloves, thyme, sage, and other spices. The Latin name for shallot is *Allium ascalonicum*. Shallots belong to the lily family. The name Allium is known to be derived from the Celtic word Allium (pungent), whereas the name ascalonicum could either be derived from its original site of cultivation, Ascalon, an old Palestinian town, or from the French name Echalogne.

Methods: In this study the extract of shallot was extracted by organic solvents and purified with help of column chromatography and TLC. The pure compound was named shallomin. Antimicrobial activity of this compound was tested against 23 strains including pathogen and non pathogen microbes using modified E test and broth dilution technique.

Results: All fungi and bacteria tested were sensitive to shallomin. The MIC values of shallomin for microorganisms tested ranged from 2–10 μg/ml. fungi were more sensitive than bacteria to this novel antimicrobial compound. MLC values were slightly greater than MIC indicate cidal nature of antimicrobial at low concentration.

Conclusion: Shallot is commonly used as a folklore medicine, and used to cure earache, fever, antidote for snake venom and also as an aphrodisiac. In this study antimicrobial properties of shallot were investigated for discovery of a new antibiotic. Based on this shallomin can be an effective medicine for treatment of dermatomycosis and other infectious diseases.

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Hydatid disease of the lung

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Human hydatid disease is caused by metacestode of Echinococcus granulosis. Hydatid disease of the lung appears more frequently in childhood. The infection may result in asymptomatic to severe disease which may be fatal. The conventional treatment of hydatid disease is the surgical intervention. Several reports proved that the patients who have widespread disease and no surgical resection is possible, the medical treatment remains the only oportunity. Percutaneous aspiration and treatment of hydatid cysts was believed to be contraindicated due to anaphylaxis and spillage of scolices, however several investigators have reported neither anaphylaxis nor dissemination. The authors present a successful treatment in pulmonary hydatid disease with bronchoscopic investigations combined with mabendazole/albendazole therapy. A nine year old boy was admitted to a county hospital because of intense black vomit without any severe disease in his previous medical history. He lost 8 kgs within a month and once he had bloody split earlier. Chest X-ray showed decreased left side basal transparency. CT scan suggested a tumor and he was referred to the 2nd Department of Pediatrics of Semmelweis University. Laboratory data indicated the possibility of lung infection and we started antibiotic therapy. The patient improved after few days and bloody vomit was stopped. Repeated CT scan did not prove any improvement and bronchoscopy was required. Cysts were found in the samples removed in the course of bronchoscopy. Histological and serological investigations confirmed the diagnosis of pulmonary hydatid disease. Repeated bronchoscopy, lavage and 50 mg/ bwkg/day mabendazole then 10 mg/bwkg/day albendazole therapy were continued. After 10 months of combined therapy the patient is in good medical condition (he gained weight, he regulary performs physical activities). Control bronchoscopy and CT scan showed mild bronchiectasis on left side in the S6 segment. After one year from the beginning of the hydatid disease the patient recovered nearly completely. Conclusion: The authors demonstrated the safety and efficacy of combined medical and bronchoscopic treatment in pulmonary hydatid disease.

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Prevalence and clinical characteristics of recurrent wheezing infants in the south of Brazil

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Background: There are a few data regarding recurrent wheezing in infants around the world. The aim of study is to verify the prevalence of recurrent wheezing in infants from south of Brazil.

Methods: This is a cross-sectional study. A standardized questionnaire was applied to parents of infants with 12 to 15 months-old attending Health Centers for regular immunization between August/2005 and December/2006. This instrument was previously validated and had questions about clinical characteristics, wheeze, respiratory infections and risk factors. Among 107 Immunization Centers 35 were randomly selected in order to maintain a homogeneous selection of population sample.

Results: Three thousand and three parents answered the surveys. Fourty five percent have had at least one wheezing episode in the first twelve months of life, starting with 5.5 ± 3.1 months of age, and 678 (22.6%) have had 3 or more wheezing episodes. In this group, 84.6%, 18.5%, 24.3% and 5.4% have used β_2-agonists, inhaled steroids, oral steriods and antagonists of leukotriene receptors, respectively. Night-time symptoms, breathless and emergency room visits were as frequent as 58.9%, 46.2% and 57.6% in all of wheezing infants, 12.7% had hospitalization for asthma and in 10.9% asthma was diagnosed. Night-time symptoms, emergency room visits, severe symptoms, asthma hospitalization and diagnosis of asthma were more frequently among wheezers with equal or more than 3 episodes ($p < 0.001$).

Characteristics of wheezes	≥ 3 episodes n = 678 (%)	< 3 episodes n = 682 (%)
Night-time symptoms	(36,6)	(22,3)
Emergency room visits	(34,7)	(22,9)
Severe symptoms	(29,6)	(16,7)
Hospitalization for asthma	(8,6)	(4,1)
Doctor asthma diagnosis	(8,1)	(2,8)

Conclusion: The prevalence of recurrent wheezing in infants in the south of Brazil is high, it start early and carriers on great morbidity. Infants possibly represent a high number of persistent asthmatics.

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Pediatric asthma severity score "PASS" and pulmonary score "PS" as clinical tools compared to others instrumental measures in acute asthmatic children

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Background: International guidelines for treatment of acute asthma call for measurement of peak expiratory flow rate (PEFR) or other objective tests of pulmonary functions. Such measures, however, are frequently difficult to obtain in young children and those unfamiliar with the technique of PEFR. Therefore a variety of clinical scoring systems have been developed for evaluating the severity of acute exacerbations of asthma in children.

Aim: To cmopare between both pediatric asthma severity score "PASS" and pulmonary score "PS" as clinical tools to others instrumental measures as peak flow meter, pulse oximetry and arterial blood gases for assessing the severity of acute asthmatic attacks in children. Also, our aim to find if clinical assessment tools are of value for rapid intervention in treatment of acute asthma, compared to laboratory measurements.

Patients and Methods: These clinical study was done on 100 ashtmatic children attending ER with acute attacks of asthma. Their age ranged from 5–16 years during the period from January 2005 to January 2006. Clinical assessment of severity was done by using both "PASS" and "PS". The results of assessment were compared to "PEFR", oxygen saturation and arterial blood gases.

Results: There was significant association between "PASS" and "PEFR" before treatment, 20 minutes and 24 hours after treatment.There was also significant association between "PS" and "PEFR" before treatment, 20 minutes and 24 hours after treatment. There was significant association between both "PASS" and "PS" scores and oxygen saturation, after 24 houres of treatment.

Conclusion: "PASS" and "PS" are of clinical utility in assessing the severity of asthmatic attacks especialy if the patiant is unable to use the peak flow meter or when pulse oximetry or the peak flow meter are not available.

Key words: Acute asthma, assessment, pediatric asthma severity score, pulmonary score, peak flow meter, pulse oximetry, arterial blood gases

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Some clinical and immunologices characteristics at children with the pathology of the biliary systems proceeding on the background of allergic diseases

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Background: To study features of immune infringements at children with a pathology biliar systems proceeding on a background of allergic diseases.

Methods: Under supervision there were 36 children with bronchial asthma (BA). To all children determined a level of general IgE, absolute quantity of basofils in a peripheral blood.

Results: At all surveyed children deformations of a bilious bubble (S-figurative, prezense of excesses in the field of a body, a bottom and cervix) have been revealed. Thus similar features of a structure of a bilious bubble frequently were found out in parents of surveyed children. Clinical displays were characterized by an abundance and polymorphism of complaints. The incidental nausea and vomiting was observed at 21 % surveyed with BA. Dispepsia frustration were observed at 7 %, locks at 28 %, meteorism at 10 %, abdominal a painful syndrome at 18 % of surveyed children. At the majority of patients complaints of vegetative character were marked. The increase of local or general sweating was established in all the patients. Patients complaints of undue fatigability, emotional lability, sleeplessness, headaches, dizzy. Disturbance of thermoregulation manifested itself by chilling and fall in temperature of distal parts of extremities. Significant meteotropism was discovered in children with BA (46,4 %), that caused the aggravation of general condition in change of weather. At children with the expressed clinical displays of a pathology biliar systems the tendency to higher parameters of the general IgE (514 ± 303 IU/ml), than at children with asymptomatic current of illness (320 ± 200 IU/ml) was marked. At 15 % of children IgE was within the limits of norm. Correlation analyses between level of general IgE and quantity of basofile showed the positive correlation r=+0.63. One of in children with the level of general IgE in limit norms observed increasing quantity of basofiles peripheral blood. May to suppose that in these children happen IgE-independent degranulation fat cell, connected, possible, with breach of vegetative innervation.

Conclusion: Thus, the lead researches dictate necessity of more profound inspection immunologices parameters at the given contingent of children.

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Prognostic importance of IgE for the evolution of recurrent wheezing into the bronchial asthma in children

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The recurrent wheezing associated with childhood can evaluate in the bronchial asthma in 30–70%.

Aim: The study was performed to determinate the potential forecast of IgE for the development of the recurrent wheezing into the bronchial asthma.

Methods: 37 children ages 2–4 years old with the recurrent wheezing were included in the study. The study has been done as a part of the detailed medical cheek up in the follow-up period 3–5 years. We used the stepwise selection Student (t) criterion of statistical analysis, coefficient of contingent (÷2), U-Fisher (F) criterion for the selection of forecast parameters in the development of asthma in childhood.

Results: The statistical discriminative "Stepwise" analysis identificated the level of IgE a parameter with potential informative prognostic for selection the children with risk of asthma. The prognosis of asthma's risk in children with hiperimmunoglobulinemia E (582,03 ± 83,27 IU/ml) which have specificity 56,16%, sensitivity 62,96% and prognosis index 59,46%. The favorable evolution with complete recovery of children with wheezing is determinated by IgE ($p<0,01$) more increased (391,75 ± 54,47 IU/ml) and potential prognosis 77,78%. Knowledge of such bronchial asthma risk's needs elaboration of a program for effective measuring to prevent asthma in children with recurrent wheezing.

Conclusion: The level of hiperimmunoglobulinemia E in child with recurrent wheezing permits to prognosis the risk of the development of bronchial asthma.

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Thalassotherapy (seaside-treatment) in allergic children

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Background: It is generally held that a comprehensive therapy for allergic children includes thalassotherapy, i.e. seaside treatment. We have attempted to objectify the effect of this therapy in two groups of children: one with the diagnosis of bronchial asthma, the other with atopic eczema.

Methods: The effect of a three weeks' seaside stay was evaluated in a group of 31 asthmatic children, compared to 23 asthmatic children in a control group. The evaluation was based on the following criteria: change of activity score, pulmonary functions, price of an anti-asthmatic pharmacotherapy, number of school absences and parent questionnaires. In the second group, which included 25 children with atopic eczema, the effect was evaluated on the basis of the extent of skin affliction, intensity of the symptoms, SCORAD index and parent questionnaires.

Results: In the group of asthmatic children, the score fell from 3.8 to 1.9 (with no change in the control group). The difficulties became less frequent, dyspnea on exertion was alleviated, drug consumption fell, school absences and the price of anti-asthmatic therapy were reduced. Pulmonary functions remained unchanged. In the group of eczematous children the extent of skin affliction fell from an average of 8.68 % to 1.88 %. The score reflecting the intensity of difficulties fell on average from 2.44 to 0.56 points with each child. The average value of the SCORAD index in the whole group before treatment was 10.26, after treatment 2.32. The changes of all evaluated parameters are of statistical significance.

Conclusion: A three week therapeutic seaside treatment reduces the activity of bronchial asthma and atopic eczema and markedly improves an allergic patient's quality of life.

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Ready-to-use HDM atopy patch test (APT) in the diagnosis of sensitization to HDM in children

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Background: Sensitization to allergens derived from house dust mites (HDM) is strongly associated with asthma, perennial rhinitis, and atopic dermatitis. Its diagnosis is considered crucial in the allergic work up of these conditions. An increasing number of reports demonstrate the accuracy of atopy patch test (APT) in the diagnosis of HDM allergy.

Aim: The aim of this study was to assess the accuracy of a new ready-to-use HDM atopy patch test (APT) (Diallertest®) in the diagnosis of sensitization to HDM in children.

Methods: A prospective study was carried out in 47 children, age 57.4 ± 42 (mean ± SD, ranges 7-176 mo), 18 girls. Patients exhibited isolated or combined atopic dermatitis (AD) (n=28, 59.57%). Children were tested for specific

HDM-IgE [against D. pteronyssinus (DPT) and D. farinae (DF)], and skin testing based on HDM (DPT and DF) skin prick test (SPT) and ATP, using HDM-Diallertest®)

Results: Among the 47 enrolled children, 15 cases (31.9 %) exhibited sensitization with positive specific HMD-IgE titers against both DPT and DF and 16 (34.04) and 17 (36.1 %) positive SPT against both DPT and DF respectively. The HDM-Diallertest® was positive in 16 (34.04 %). Among these 16 positive HDM-Diallertest®), 9 exhibited an eczematous reaction and showed an excellent concordance with DPT and DF-SPT and specific IgE against DPT and DF, respectively 93.3%, 97.77%, 90.47% and 90.47%.

Conclusion: The 3 diagnostic techniques exhibited a comparable level of accuracy for the diagnosis of HDM allergens sensitization. The excellent concordance with other techniques of HDM-Diallertest®) with eczematous reactions strongly supports its use as a reliable non invasive diagnostic tool of HDM sensitization.

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Prevalence of asthma, allergic rinoconjunctivitis and atopic dermatitis in the southwest of Mexico City, ISAAC model study

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Introduction: The allergic diseases are the most frequent chronic entities in the childhood, through the time their prevalence has been increased in the last three decades, that is why it was deasigned a task force called International Study of Asthma and Allergy in Childhood (ISAAC) that promotes the epidemiological researches in asthma and allergic diseases.

Objective: To approach the prevalence and severity of asthma, allergic rhinoconjunctivitis and atopic dermatitis in children between 6 and 7 year and 13 and 14 year old in the southwest of Mexico City.

Methods and Materials: It is an observational, transversal, descriptive, prospective study, it includes students of first and second grade of primary school whit age of 6 and 7 year old and junior high school students from 13 to 14 year old. The study was authorized by the Public Education Institute, school principal and parents. The statistical analysis was made by Epi info version 6 whit a significative value of $p \leq 0.05$.

Results: There were 58 schools enrolled, with 6184 students from wouthwest of Mexico city, 32 primary schools with 3093, 1585 girls (51.24%) and 1508 boys (48.75); junior high schools included 3034 students and 26 schools with 1538 girls (50.69%) and 1496 boys (49.30%). In the scholastic population the antecedent of only one wheezing episode was present in 20.43% (632) and the proportion of patients with severe asthma was 6.7% (207), from this data only 4.3% (134) had asthma diagnosded, in the teenagers only 172 (5.66%) had diagnostic. The severe rhinoconjunctivitis was present in 399 kids (25.99%) and only 103 teenagers (11.26%). The major part of students, kids and adolescents did not give correctly the information, about atopic dermatitis, only 9 (0.29%) joined with pets (cats and dogs).

Discussion: Similar studies made around the world reveals that asthma and allergic diseases are a public health problem, with implications in quality of life, it may lead to temporal o permanent incapacity, if they are not diagnoses and treated in early approach. The variation in the prevalence are expected because various factors, the most important is the diversity of each zone about risk factors, in fact the results can not be compared with a different population, they can be used for the epidemiological knowledge of this zone.

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Bronchial asthma and child physical activity in Lithuania

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The background of the study. Prevalence of child asthma is 2-3 % in Lithuania. Asthma is not only clinical, but social problem also. The aim of this study was to evaluate physical activity and its limitation in asthmatic children. The method used. The study group included children suffering from asthma between the ages of 7 and 17. The severity of asthma was assessed by grading the asthma as mild, moderate or severe. We used standardized and disease-specific PAQL questionnaire (national version). Responses of 186 asthmatic children were analyzed. The results obtaines. Only one third of our respondents were involved in extra activity: sport, dancing and singing. 62.9 % of parents noticed that their children have no extra activity. 17,7% of children with asthma were not attending sports at their school. We asked childrens to name three activities, in which they had been bothered by asthma. 16,6 % of respondents indicated more than three daily activities, restricted by asthma. Asthma for boys was the most disturbing while running (85.2%), playing with pets (23.8%), playing ball (25%) and sleeping (26%). Girls marked different activities, such as running (64%), housekeeping (46.9%) and school sports (25%). Asthma bothered boys on the run and playing ball more often than girls. Girls were bothered by asthma more often than boys when doing chores and singing. Statistically significant differences were found comparing all the three main activities for boys and girls: the first activity - $\times^2 = 44.9$, $df = 15$, $p < 0.001$; the second activity - $\times^2 = 42.8$, $df = 26$, $p = 0.02$; the third activity - $\times^2 = 47.9$, $df = 23$, $p = 0.002$. The conclusion reached. Asthma is a limiting factor in the children's level of physical activity. It prevents children from doing various activities. Due to asthma, children were not able to run, play ball, do their chores, play with their pets. Boys more often complained thet asthma had limited their physical activity (running, playing games), whereas girls indicated that the disease had limited both, their physical activity (such as running) as well as passive activities (such as doing chores, in addition to dancing and singing).

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Analysis of allergen-specific serum IgE in children with asthma or cough variant asthma

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Objective: To investigate the positive rates of food allergen (Fx5), mixed molds (mx1), phadiatop and house dust mites (d1) allergen-specific serum IgE (sIgE) in children aging from 6 months to 16 years diagnosed of asthma or cough variant asthma (CVA). Their age distribution characteristics were also compared and analyzed.

Methods: Blood samples of 597 asthmatic children were applied to fx5, mx1, Phadiatop and d1 tests using fluoroenzyme-immunometricassay, UniCAP100. The positive rate of fx5,mx1,Phadiatop and d1 were compared. Age distribution characteristics of children with positive SIgE were analyzed.

Results: 1, The positive rate of fx5, mx1, Phadiatop and d1 were 34.6%, 30.%, 35.2% and 23.5% respectively; 2, The highest positive rate of Phadiatop and d1 were highest in children of 8-16 age group (73.6% and 49.1%); The positive rate of fx5,mx1 peaked in children of 2 years and 5 years separately(44.1% and 53.6%); 3, Among 181 children with mx1 positivity 90 cases manifested phadiatop negativity. 4, 65% of children with clinically diagnosed asthma or CVA presented positivity in test in fx5,mx1 or phadiatop at least.

Conclusion: Two-thirds asthma and CVA patients existing sIgE; Asthmatic infants mainly manifested food allergen sensitization. The positive of Phadiatop increased with age. Positivity of mx1 increased with age in children ≤ 5 years , and it decreased in older children (≥ 6 years). Mx1 was one of the main allergens of asthmatic children.

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An observational study on the effectiveness of montelukast in children with chronic rhinitis

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Background: Impacts of chronic rhinitis in children especially nasal congestion included learning disorder, impaired school performance and quality of life. Intranasal corticosteroid is the most effective medication for chronic rhinitis treatment. Recently, leukotriene receptor antagonist has been added to the modes of therapy approved by the US Food and Drug Administration of allergic rhinitis.
Objective: To assess the effectiveness of add-on montelukast with intranasal corticosteroid in children with chronic and persistent rhinitis.
Methods: We enrolled children attending the allergy clinic, Department of Pediatrics, Faculty of Medicine, Chulalongkorn University, with poorly controlled rhinitis during the period of April, 2005 to December, 2006. Diagnosis of allergic rhinitis was made by positive prick skin test to at least one aeroallergen. Intranasal corticosteroid was the first-line medication for treatment of persistent symptoms. Montelukast was added in cases that no response to intranasal corticosteroid, mostly due to nasal congestion. Efficacy was assessed by symptom improvement (nasal congestion, rhinorrhea, itching, sneezing) reported by patients and physical examination by allergist.
Results: There were 10 boys and 16 girls (average age 13). The mean duration of symptom before diagnosis was 4.7 years. Twelve children were diagnosed to be allergic rhinitis and 14 children were non-allergic rhinitis by prick skin testing. The mean duration of intranasal steroid use was 4.4 year in children with allergic rhinitis and 2.7 years in children with non-allergic rhinitis. Symptom improvement, especially nasal congestion, was reported in 8 of 12 (66.6%) allergic rhinitis subjects and 11 of 14 (78.5%) non-allergic rhinitis subjects after add-on 10 mg of montelukast daily. Montelukast was overall well tolerated. Mean duration of montelukast therapy was 1.7 years in both groups. Intranasal corticosteroid was able to stop in 8 of 14 (57%) children with non-allergic rhinitis and unable to stop in all children with allergic rhinitis, but all of the patients were satisfied with these treatment.
Conclusion: Montelukast have an advantage in additional treatment of either allergic or non-allergic rhinitis children, but particularly more benefit on children with non-allergic rhinitis.

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Sputum eosinophils in childhood asthma: Correlation with PEFR in controlled asthmatics and with exacerbation

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Background: Sputum eosinophilia is a non-invasive airway inflammatory marker. Airway inflammation which is the characteristic functional abnormality peculiar of asthma, leads to variable airflow limitation. Significant correlation between PEFR and absolute eosinophil counts have been observed among patients with asthma. However, these studies are limited and others show conflicting results.
Objective: To determine the correlation between sputum eosinophilia and PEFR among patients with asthma.
Methodology: Following clinical assessment and peak flow determination, sputum eosinophil count was determined among patients with stable asthma (29) and acute exacerbation (36). Patients were classified as to intermittent and persistent asthmatics. Age, sex, atopic status and asthma duration were obtained. Sputum eosinophil count of = 3% was considered significant.
Results: Patients with sputum eosinophilia had lower PEFR, compared to those without sputum eosinophilia ($p = 0.004$). Patients with sputum eosinophilia were more likely to have abnormal PEFR compared to those with normal PEFR (OR = 6.64, 95% CI = 2.02-21.90). Patients with persistent asthma were more likely to have sputum eosinophilia compared to those with intermittent asthma (OR = 3.06, 95% CI = 1.02–9.18). Similarly, patients in acute exacerbation were more likely to have sputum eosinophilia compared to those with stable asthma (OR = 3.42, 95% CI = 1.07–11.04).
Conclusion: This study demonstrates that there is a direct correlation between sputum eosinophilia and PEFR among patients with acute exacerbation. However, no correlation could be made among patients with stable asthma since all patients had normal PEFR.

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Serum levels of VEGF, TGFβ2, IL-4, IL-2R and ECP in children with acute astma bronchiolitis

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Background: Asthma is leading cause of chronic illness with increased vascularity, vascular permeability, airway and lung tissue remodeling. Although, various growth factors and cytokines have been implicated in modulating in these processes, the role of factors and cytokines are not known very well in asthmatic children. Aim of this study is to determine the serum levels of vascular endothelial growth factor (VEGF), transforming growth factor beta-2 (TGFβ2), interleukin-4 (IL-4), interleukin-2-receptor (IL-2R), and eosinophilic cationic protein (ECP) in acute astmatic children (AAC) and compare with acute bronchiolitis and healthy subjects and to assess the changes with treatment.
Methods: These parameters were measured by ELISA techniques in AAC and bronchiolitis patients before and after inhaled or systemic steroid (SS).
Results: Serum VEGF, TGFβ2, IL-4 and IL-2R levels were increased in patients with AAC and bronchiolitis than in controls before therapy. These parameters were higher in AAC than bronchiolitis except IL-2R (Table-I).

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	Patients with Bronchiolitis	Controls (Bronchiolitis)	Patients with Acute Asthma	Controls (Acute Asthma)	p
VEGF (before treatment)	469,7 ± 117,8	145,9 ± 56,1	870,75 ± 496,5	210,3 ± 50,5	p1, 2, 3 < 0, 0001
IL2R (before treatment)	2663,3 ± 829,6	904,6 ± 178,2	2458,2 ± 851,80254	909,6 ± 196,9	p4, 5 < 0,0001, p6 > 0, 05
IL4 (before treatment)	15,0 ± 3,7	5,3 ± 3,2	28,6 ± 8,9	4,3 ± 3,0	p7, 8, 9 < 0, 0001
TGFB (before treatment)	8661,4 ± 2384,5	3220,7 ± 1556,7	18573,2 ± 8862,7	3434,3 ± 1638,5	p10, 11, 12 < 0, 0001
ECP (before treatment)	16,9 ± 5,2	5, 2043 ± 2, 6	55,6 ± 23,0	4,6 ± 2,2	p12, 13, 14 < 0, 0001
VEGF (after treatment)	257,5 ± 100,7	145,9 ± 56,1	366,1 ± 268,2	210,3 ± 50,5	p15 < 0, 05, p16, 17 < 0, 0001
IL2R (after treatment)	2020,6 ± 790,9	904,6 ± 178,2	1272,9 ± 515,9	909,6 ± 196,9	p19 < 0, 0001, p20 < 0, 05, p21 < 0,0001
IL4 (after treatment)	2, 2± 1,8	5,3 ± 3,2	6,8 ± 4,8	4,3±3,0	p22 < 0, 0001, p23 < 0, 05, p24 < 0,0001
ECP (after treatment)	7,8 ± 4,0	5,2 ± 2,6	23,1 ± 14,9	4,6 ± 2,2	p25 < 0, 05, p26 < 0, 0001, p27 < 0, 0001

Serum VEGF, IL-2R and ECP levels were higher in AAC than in controls after treatment. After steroid therapy, VEGF, IL-4 and ECP levels were higher in AAC than in bronchiolitis but IL-2R levels were higher in patients with bronchiolitis. Only serum IL-4 levels in bronchiolitis were similar with controls after treatment. By the severity of disease, only VEGF levels were increased in modarate AAC and bronchiolitis higher than in mild patients ($p < 0,0001$). Only VEGF levels were lower in AAC who were treated with prophylactic drugs than in AAC without prophylactic treatment ($p < 0,05$). According to modality of steroid treatment, only VEGF levels were lower in AAC who were treated with inhaled steroids compared with SS($p < 0,001$). In AAC; VEGF, IL-4, IL-2R and ECP levels were higher than in controls after treatment with SS ($p < 0,0001$). But in AAC who were treated with inhaled steroids, VEGF and IL-4 levels were similar with controls after treatment ($p > 0,05$).

Conclusion: VEGF, TGFβ2, IL-4 and IL-2R may play role in pathogenesis of asthma and bronchiolitis. These parameters aren't useful for distinguishing asthma and bronchiolitis. VEGF could be useful parameter for distinguishing mild and moderate disease in asthma and bronchiolitis. According to our findings, inhaled and SS could be combined for appropriate treatment in moderate AAC. Also, anti-VEGF therapy may be useful in AAC.

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Effect of self-management education on asthma complication and pulmonary function in Iranian children with asthma

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Background: Despite global efforts to manage asthma, the adverse outcome of this disease is increasing. Effective self-management and treatment compliance is important in achieving good symptom control in asthma. The aim of this study was to determine whether asthma nurse intervention during clinical period could increase knowledge and improve self-management and whether this would influence the frequency and severity of symptoms, and pulmonary function test.

Setting: A university hospital pediatric outpatient clinic.

Methods: Randomized controlled trial of 60 children with intermittent and mild to moderate persistent asthma were assessed to indicate whether the educational self-management intervention would improve lung function and the use of inhaler with spacer score, increase feeling of control over asthma, and decrease the use of health care services. 60 children with asthma, who were categorized based on disease severity, duration of disease, age were randomly assigned to an experimental group (n=30) which received an educational program or to a comparison group (n=30) which received common education. The education program consisted of 4 group sessions which focused on improving the patients and parents' self-management skills. The follow-up continued for 4 months. Results: At follow-up, most indicators of the use of MDI and spacer score and asthma control test had improved significantly ($p < 0.005$) among educated patients. Also cough decreased significantly in the intervention group ($p < 0.005$). The pulmonary function test did not show increase in FEV1/FVC after this period, it was not statistically significant as compared to the control group.

Conclusion: The nursing educational intervention for children with asthma and their parents effectively improved inhaler use skills and decreased cough and ER visits in this group of patients. Regarding the insignificant changes in FEV1/FVC, it should be considered that pulmonary function test is not already seriously affected in the group of intermittent and mild to moderate persistent asthma. Further studies including severe asthma patients and longer periods of follow up are warranted.

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Toxocarosis in the background of chronic cough in childhood: a longitudinal study in Hungary

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Background: Chronic cough lasting 8 weeks or more often seems an intractable problem in childhood. Toxocara infestation is associated with an increased prevalence of airway symptoms and may be the possible etiologic agent of chronic cough.

Methods: Spirometry, bronchial reversibility tests and nonspecific bronchus provocation tests were carried out in patients who could cooperate in these tests. Measurements were made of the peripheral blood eosinophil count, the serum total IgE level, the serum total magnesium (Mg) concentration. The serological examination for *Toxocara canis*, based on the most widely used enzyme-linked immunosorbent assay of IgG (*Toxocara canis* IgG ELISA, Novatec, Germany) and more sensitive Western blotting (Toxocara Western Blot IgG, LDBIO, France) methods, was performed. All patients with positive serology for *T. canis* index of positivity > 9 were treated with albendazole in a dose of 10 mg/kg/day, patients weighing > 40 kg received 400 mg albendazole a day for 7 days. All patients with bronchial asthma and CVA were treated with inhaled corticosteroid (ICS). Further, short-term beta2-mimetics could be administered in cases of dyspnoea "as needed".

Results: One-hundred and thirty-six of 425 children (32%) aged 2–17 years were seropositive for *T. canis* antigens. Ninety-three children were adequately assessed, diagnosed and followed up during 1 year. Bronchial asthma was diagnosed in 40%, cough variant asthma (CVA) in 27% and nonasthmatic eosinophilic bronchitis (NAEB) in 33% of children with chronic cough. The eosinophil cell count, the *Toxocara canis* IgG, the total IgE levels and the total Mg concentrations in the serum are predictors of the improvement or the decline of the patients' condition. We could significantly decrease the dose of ICS in 23 of 37 (62%) children with bronchial asthma. Anthelminthics and avoidance were sufficient for children with NAEB; none needed ICS. ICS therapy could be stopped 2-3 months later in 17 (68%) of 25 patients with CVA. We found that 8 of 25 patients with CVA (32%) presented asthmatic symptoms at the end of the 1-year period.

Conclusion: In Hungary, *T. canis* may be the potential sensitizer for chronic cough in seropositive children. Deworming therapy will then alleviate the airway symptoms without exacerbation in patients with bronchial asthma, and definitively treat children with NAEB and the majority of children with CVA.

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Peak inspiratory flow rate via incheck-dial ® in children with asthma

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Background: The detrimental effect of chlorofluorocarbon on the ozone layer has resulted in the prohibiting of CFC-containing medication usage. Dry powder inhaler (DPI) devices are alternative devices to deliver medication for treatment of asthma. Drug delivery and lung deposition from DPI are dependent on the peak inspiraory flow rate (PIFR).

Methods: Using PIFR meter (Incheck-dial) that mimics the internal resistance of commonly use DPI devices one high-resistance(Turbuhaler) and two low-resistance (Accuhaler and Easyhaler) determined the PIFR generated by asthmatic children aged 5-18 years through each of them. Pulmonary function was measured by using spirometry.

Results: 252 asthmatic children were studied, 169(67%) children were male. The mean age was 10.58 + 2.97 years. The mean FEV_1 (% of predicted) was

80.30 + 10.91. The mean PIFR with the internal resistant of turbuhaler, accuhaler and easyhaler was 85.11 + 14.84 L/min, 103.01+16.01 L/min, and 114.72+11.74 L/min, respectively. PIFR was statistically significant correlations with age, body weight and height (p value <0.001). All children could generated PIFR more than 30 L/min in both accuhaler and easyhaler resistance. 8(3.17%) children were unable to generate PIFR more than 60 L/min in turbuhaler resistance. There were no significant correlations of FEV_1 (%of predicted), FEV_1/FVC% or FEF_{25-75} with PIFR.

Conclusion: Most children aged at least 5 year can generate enough PIFR for using DPI devices especially in low-resistance devices.

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IgE level, CD26, CD30 expression and intracellular interferon-γ production by cord blood mononuclear cells as predictors of atopic dermatitis forming in infants: a one-year prospective birth cohort study

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It is supposed that prenatal Th1/Th2 polarization of immune response is possible, but the markers of such polarization, trustworthy associated with atopy forming are not known.

Aim: To evaluate the connection between IgE level, CD26, CD30 expression and intracellular interferon (IFN)-γ production by cord blood mononuclear cells (CBMC) and the risk of infant atopic dermatitis (AD) forming.

Methods: We defined per cent content of CD26+and CD30+in CBMC by using flow cytometry as well as the concentration of total IgE in 117 full-term newborns. Simultaneously, intracellular IFN-γ expression as well as early activation marker CD69 (separate for total mononuclear cells population and CD4+lymphocytes) were examined (95 newborns). We estimated AD presence in 41 enrolled newborns in the age of one year (medically diagnosed AD was marked in 25 of them). Data are shown as median (25-75% quartiles) of absolute cells count (10(-)6/l). Statistical analysis was performed using the Mann-Whitney test. Relative Risk (RR) was present with 95% CI.

Results: Only for nonactivated CBMC, producing IFN-γ (subtype CD69-/IFN-γ+), the association with AD forming has been found. The number of CD69-/IFN-γ+CBMC in AD infants was 3.3 (0-6.4), as opposed to infants without AD -9.4 (5.3-16.5), p = 0.006. Relative risk for AD forming in newborns with low CD69-/IFN-γ+(< 5.0 - median for our cohort) was 3.4 (1.19-9.69). Significant differences in the number of CD69-/IFN-γ+CBMC was kept after adjusting to such factors as family atopic history, place of living (city/village), birth order (first/second or later), the number of family members living together, bedsharing smoking, pets owing and the newborn gender.We revealed the negative concentration of total IgE in cord blood with the ratio CD26+/CD30+CBMC (r = −0.37, p = 0.0005). We did not find any association of CD26, CD30 expression, the CD26/CD30 ratio, as well as the level of total IgE with AD forming in infants (medians were chosen as cut off points). RRs were 1.5 (0.67-3.33), 0.67 (0.37-1.19), 0.78 (0.42-1.45) and 1.24 (0.59-2.61), respectively.

Conclusion: Thus, the decreasing of intracellular IFN-γ production by nonactivated CBMC, proving the prenatal displacement in Th1/Th2 balance, is the strong predictor of infant AD forming. However, the decreasing of CD26/CD30 ratio as well as the level of total IgE in cord blood are not the predictors of atopic phenotype forming in infants.

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Sputum eosinophil level in children with mild, moderate to severe exacerbation and stable asthma

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Background: Sputum eosinophilia is a non-invasive marker in the evaluation of airway inflammation as well as asthma control. Increased proportion of sputum eosinophils have been observed among subjects with asthma, especially among those with exacerbation. However, there are also studies that show airway eosinophilia among patients with controlled asthma.

Objective: To determine sputum eosinophil level among asthmatic patients in exacerbation and those with stable asthma and determine its association with severity of disease.

Methodology: Following clinical assessment and peak flow determination, sputum eosinophil level was determined among patients with stable asthma (n = 29), mild (n = 18) and moderate to severe (n = 18) asthma exacerbation. Age, sex, atopic status, asthma duration and medications were obtained. Sputum eosinophil count of ≥ 3% was considered significant.

Results: The mean eosinophil count in children with stable asthma, mild exacerbation and moderate to severe exacerbation were 4.65% ± 12.53%, 1.94% ± 4.30%, and 8.83% ± 11.12% respectively. Sputum eosinophils were significantly higher in children with moderate to severe exacerbation than in mild exacerbation and stable asthma (p = 0.0086). The percentage of patients with significant sputum eosinophilia was higher among those with moderate to severe exacerbation (17%) than mild exacerbation (6%) and stable asthma (8%). Statistically significant difference was seen among the three population (p = 0.0234). Logistic regressions showed that wheezing (p = < 0.0001), peak expiratory flow rate (p = 0.0010), sex (p = 0.0110) and asthma duration (0.0232) were significantly associated with sputum eosinophilia (p = 0.432). No statistically significant difference was seen with respect to long term asthma severity classification (p = 0.432).

Conclusion: This study demonstrates that significant sputum eosinophilia is seen among patients with moderate to severe asthma exacerbation compared to those with mild exacerbation and stable asthma. Sputum eosinophilia was also significantly associated with wheezing, lower peak expiratory flow rate, male sex and longer asthma duration. Long-term asthma severity is not a defining factor in the evaluation of sputum eosinophilia among asthmatics but the severity of exacerbation.

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Allergy in children: managing patients at risk of anaphylaxis

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Aim: To report the management of children with allergy in a district general hospital, in particular to review prescribing practices of epinephrine auto injectors (Epipen) and to compare the management of children seen in allergy clinic with those seen in general paediatric clinic.

Methods: The clinical records of all children who were prescribed Epipen or Epipen junior between November 2003 and October 2005 were retrospectively reviewed. 39 patients were included of which 23 were seen in allergy clinic and 16 were seen in general paediatric clinics. There ages ranged between 6 months to 17 years (median of 7 years).

Results: The common allergens were Nuts/seeds (79%) followed by egg (23%), milk (18%) and fish (8%). 15 children had positive skin prick tests and 18 had positive RAST (total 33/39 had positive tests). Information on treatment with Epipen was provided to 22/23(95%) children seen in allergy clinic compared to 10/16 (62%) children seen in general clinics. Practical instruction of Epipen administration was provided to 21/23(91%) compared to

	Allergy Clinic	General paediatric clinic
Allergen avoidance	82.6%	81.2%
Epipen information	95%	62%
Epipen Training	91%	75%
Information on antihistamine use	91%	68%

12/16(75%) children seen in general clinics. Information on treatment with antihistamines was provided to 21/23(91%) children in allergy clinic compared to 11/16(68%) in general clinics. Information on allergen avoidance scored equally in allergy clinic (82.6%) and general clinics (81.2%).
Conclusion: This case series demonstrates that children managed in allergy clinic are better informed about their allergies and management compared to those managed in general paediatric clinics.

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Metabolic responses to treatment with salbutamol and theophylline in asthmatic children

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Drug therapy and its abuse have been incriminated as a main contributive factor in asthma mortality. The effect of treating asthmatic with salbutamol inhalation (0.15 mg/kg body weight up to 5 mg/dose, group A), theophylline (5mg/kg body weight, group C) or both drugs (group B) was investigated in 30 asthmatic children. Ten healthy children of matching age and sex were also included to serve as control. Serum glucose increased significantly in group A and B, but did not change significantly in group C as a compared to pretreatment level. Serum inorganic phosphate decreased significantly in group B and C, with no significant change in group A. serum sodium level was significantly decreased an all asthmatic children before treatment. Group B and c showed significant reduction in serum sodium level as compared to pretreatment values. Serum sodium level did not change significantly in group A. Serum potassium decreased significantly in the three studied groups as compared to pretreatment values. These metabolic changes could possibly help in explaining the observed increase in cases of fatal asthma, when using a combination of inhaled beta-agonist and oral theophylline preparation.

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Multymeasuring phenotyping and automated prevention system of Bronchial asthma in children

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Background: Prevention in early age period significantly decreases morbidity of allergic diseases and improve quality of life. Sometimes doctors are unable to manage well this pathology, because of neglected early diagnostic. According to the most actual and perspective direction of nowadays allergology it is working out new scientific approach of resolution of bronchial asthma (BA) problems in young population. Hereditary predisposition plays the most important role in the developing of disease, but it is not everything recognizable, it may be not demonstrated clinical symptoms in parents and other close relatives of affected individual.
Aim: To elaborate the new automated assessment cost-effective system which would allow early diagnostic and prevention BA by using "formalized cards" by providing multymesure phenotyping in Georgian children population. This system is based on the primary and secondary prognosis of BA.
Methods: Choosing of markers: HLA-markers (Terrasaki micro lymphocytetoxic tests) typing was performed with 2nd class (DR, DQ) and 1st class (A, B, C) antigens locus's; Erythrocyte markers: ABO, Rhesus and MNS systems, Clinical-genealogic method (Falconer D., 1965): study of genealogical tree of investigated families; Genetic- statistic analysis – Informative Criteria of different signs: ontogeny, phenotypic, environmental factors (by Kulback, Goubler E.B, 1990). Method of multi-dimensional phenotyping: agglomerative-hierarchical cluster analyse of chosen systems of signs. Analyse was performed in the interactive order.
Results: Multi-dimensional phenotyping shoves phenotype classes: Classes of basic predisposition, including up to 80% of individuals with manifested asthma; Classes of relative predisposition, including less then 80% of individuals with manifested asthma; Phenotype classes including a few patients; Phenotype classes not predisposed to BA, basically including healthy persons.
Conclusion: Elaborated computing table is enough simple and opportune for comprehensive use. It is proposed, for primary prognostic of BA, to practical doctors working in outpatient clinics and dispensaries, as well for military doctors working with enlistment committee or in military units. This method gives the possibility for prognosis predisposition to bronchial asthma in young population.

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TBXA2R gene polymorphism and its pharmacogenetic effect to leukotriene receptor antagonist in children with asthma

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Background: Thromboxane A2 receptor(TBXA2R) gene polymorphism has been associated with atopy and asthma. However its role in children has not been defined. We investigateed associations between asthma-related phenotypes and TBXA2R polymorphism, and also to analyze whether TBXA2R polymorphism has an effect on the efficacy of the leukotriene receptor antagonist(LTRA), montelukast, in asthmatic children with exercise-induced bronchoconstriction (EIB).
Methods: Asthmatic children(n = 695) and control children(n = 159) were evaluated for asthma-related phenotypes including total IgE, pulmonary function test, and BHR to methacholine or exercise. Genotypes were detected by PCR-RFLP. In the montelukast study, exercise challenge was performed before and after 8-week montelukast treatment.
Results: The TBXA2R polymorphism was not associated with asthma susceptibility and clinical parameters of asthma. However, asthmatic children with combinations of the TBXA2R + 795T > C and + 924T > C risk alleles had significantly higher total IgE levels (P = .01), total eosinophil counts (P < .01) and lower FEV1 (P = .02) and FEF 25–75% (P = .02) than those carrying the common alleles. When compared with individuals with the common alleles, in patients with the TBXA2R + 924T > C TT homozygote and TBXA2R+795T > C hetero-or homozygote (CT or CC) had a 3.67-fold poor response to 8-week montelukast treatment regarding to maximum percent fall in FEV1 after exercise (odd ratio, 3.67; 95% CI, 1.15-11.15).
Conclusion: A combined effect of TBXA2R + 795T > C and + 924T > C risk alleles may be linked to IgE production, eosinophilic inflammation, and severity of asthma. In addition, the combined genotype of TBXA2R may be a predictive marker of clinical response to the LTRA in Korean asthmatic children with EIB.

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A randomized prospective double blind controlled trial on effects of long-term consumption of fermented milk containing Lactobacillus casei in pre-school children with allergic asthma and/or rhinitis

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Background and Aim: Probiotics may have immunomodulatory functions, exerting beneficial effects in allergic disease. We investigated whether prolonged daily consumption of Lactobacillus casei DN-114 001 fermented milk improve health status of children aged 3-6 years allergic to inhalants.

Methods: In a randomised prospective double blind controlled trial patients were randomly assigned to receive for one year 100 ml per day of L. casei DN-114 001 fermented milk (109 cells/ml), or no probiotics (controls). Clinical evaluations occurred every 3 months. Faecal flora composition was assessed every 6 months (30 treated, 15 controls). Outcome measures were time free from and the number of episodes of asthma/rhinitis, number of fever or diarrhoea episodes, IgE-G-A serological levels (12 months vs baseline).

Results: Participants (187; 92 treated) were similar regarding gestational age, breastfeeding, family smokers, pets, siblings, day-care admission. No statistical difference between intervention and control group occurred in asthmatic children. In children with rhinitis, the annual number of rhinitis episodes was lower in the intervention group, mean difference (95%CI), -1.6 (-3.15 to -0.05); the mean duration of an episode of diarrhoea was lower in the intervention group, mean difference -0.81 (-1.52 to -0.10) days. Faecal analysis showed Lactobacillus casei DN-114 001 in the gut flora of > 78% of supplemented children through the study period.

Conclusion: Long-term daily consumption of Lactobacillus Casei DN-114 001 fermented milk may positively influence on the clinical and immunological status of allergic children.

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Prevalence of pediatric allergic diseases in the first three years of life

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Background: We have maintained birth cohort study in Sagamihara-City since 2002 to clarify the prevalence of atopic dermatitis (AD), food allergy (FA), bronchial asthma (BA), and Japanese cedar pollinosis (JCP) from infancy to childhood.

Methods: Using the mass medical examination system at the age of 4 months (mo) old, we first had obtained information of eczema, nutrition method, family history of allergic diseases, and results of allergy examination by questionnaire. We followed up the subjects, whose parents agreed to participate in this study, at the age of 8 mo, 12 mo and 3 years (y) old using questionnaire by mail. Furthermore the risk factors for the subjects to develop BA by the age of 3 y were analyzed from the information obtained in the study.

Results: Informed consent was obtained from 5247 parents of 4 mo infants out of 5932 parents from 1/1/02 to 12/31/02. We could follow up 4214 infants at the age of 8 mo, 4068 infants at the age of 12 mo, and 2888 children at the age of 3 y. The incidence of chronic eczema to suspect AD at 8 mo, 12 mo and 3 y was 18.8%, 13.3% and 14.8%, respectively. The incidence of food elimination due to FA reported by parents at 8 mo, 12 mo and 3 y was 17.0%, 12.8% and 5.4% respectively. Interestingly, the incidence of doctor-diagnosed FA at 8 mo, 12 mo and 3 y was 2.4%, 2.8% and 5.1% respectively. Confusion related to FA and AD during infancy seems to exist between parents and doctors. The prevalence of BA at 12 mo and 3 y was 2.9% and 8.7% respectively. The risk factors for infants to develop BA by 3y were related to the possession of AD or FA at 8 mo, 12 mo and 3 y, family history of allergic diseases and history of indirect cigarette exposure. The incidence of JCP at 3 y was even 3.0%.

Conclusion: Since the population of children in Sagamihara-city represents about 5% of that of Japan, we can now estimate the prevalence of allergic diseases from infancy to childhood by the data obtained in this study. However, it was very difficult to estimate the prevalence of FA especially during infancy due to the under-evaluation by doctors and over-reaction by parents. It is important for both doctors and parents to fix the confusion in our society. The data can make us possible to clarify the prevalence of pediatric allergic diseases and to reveal the transition of the diseases.

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Update of the guideline on allergy prevention

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Background: Prevention is crucial to counteract the rising trend of allergies. In 2004, we published an evidence-based and consented guideline on primary and secondary prevention of asthma, allergic rhinitis and atopic eczema. In order to update this guideline, we repeated a systematic literature search in 2007 and present the results here.

Methods: A literature search was performed in Medline for the years 2003 through April 2007. The used mesh-term groups were as follows. Endpoints: asthma, allergy, allergic, atopic, hay fever, dermatitis, eczema, rhinitis. Interventions: prevention, risk factor, epidemiology.

Study types: randomized controlled trial, clinical trial, control study, systematic review, meta analysis, case control study, cohort study. Terms were combined with "and" within groups and "or" between groups. We included studies on humans published in English or German language and excluded therapeutic and drug trials. A first selection process was performed on the basis of titles and abstracts.

Results: We obtained 1551 hits of which 139 potentially relevant studies were identified. The later consists of 6 reviews, 9 RCTs, 91 cohort-and 33 case-control-studies. After an initial review, the studies could be allocated to the areas breast feeding, hypoallergenic formula, introduction of solid food, diet of the mother and the child, smoking, mould and dampness and vaccination. The recommendations of the guideline to these areas were supported by the actual evidence. There is new evidence which allows a specification of the current recommendations for the topics body mass index and exhaust. There is indication that the recommendations on house dust mite, pet keeping and unspecific immune modulation need to be revised.

Conclusion: By this literature search it is possible to update the current evidence-based recommendations on allergy prevention and to adapt the recommendations according to the current literature.

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A specific mixture of short chain galacto-oligosaccharides and long chain fructo-oligosaccharides induces an anti-allergic immunoglobulin profile in infants at risk for allergy

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Background: In a prospective study in infants with a family history of atopy a specific prebiotic oligosaccharide mixture (90% short chain galactooligosaccharides and 10% long chain fructo-oligosaccharides (GOS/FOS) (IMMUNOFORTIS) reduced the cumulative incidence of atopic dermatitis at six months of age. In a subgroup of these infants it was possible to obtain a blood sample at six months of age to analyse the potential effect of these dietary oligosaccharides on the immunoglobulin profile.

Methods: In this prospective double-blind randomized, placebo controlled, study the infants received a hypoallergenic formula with either 8g/l GOS/FOS or 8 g/l maltodextrine (placebo) for six months. At three months of age, children were vaccinated against diphteria, tetanus ad polio (DTP). At six months of age total plasma levels of IgE, IgG1, IgG2, IgG3, and IgG4 as well as cow's milk protein (CMP) and DTP specific immunoglobulins were measured.
Results: Supplementation of GOS/FOS has lead to a significant reduction in the plasma level of total IgE ($p = 0.007$), IgG1 ($p = 0.0054$), IgG2 ($p = 0.029$) and IgG3 ($p = 0.0343$) immunoglobulins whereas no significant effect on IgG4 was observed. The plasma levels of CMP specific IgG1 was significantly decreased ($p = 0.015$) in the GOS/FOS group. The levels of CMP specific IgE were very low and no effect of GOS/FOS supplementation could be observed. CMP specific IgG4 was not detectable in the samples. No influence of GOS/FOS supplementation was found on any vaccine specific antibody isotype levels.
Conclusion: Evidently GOS/FOS supplementation induced an anti-allergic immunoglobulin profile in infants at high risk for allergic diseases whereas desired specific immune responses were not affected indicating the potential role of oral GOS/FOS exposure for primary prevention of allergies.

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Bacterial DNA in infant faecal samples, as assessed by Real-time PCR, in relation to allergy development in children up to five years of age

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Background: An altered microbial exposure may underlie the increase of allergic disease in affluent societies. Early colonisation with Bifidobacterium (B.) and Lactobacilli (L.) have been postulated to prevent children from developing allergy, while *Clostridium (C.) difficile* colonisation has been associated with allergic disease. Previous studies have mainly been performed with culture dependent techniques. However, lately the sequencing of bacterial genomes has made it possible to study the gut flora with molecular techniques. The Real-time PCR technique uses primers targeting conserved genes of bacteria, leading to efficient qualitative and quantitative analysis of bacterial DNA from faecal samples.
Methods: Presence and amounts of bacterial DNA in infant faecal samples, collected at 1 week, 1 month and 2 months, were measured with Real-time PCR and related to allergy development in Swedish children, followed prospectively at 6, 12, 24 and 60 months of age. Children regarded as allergic (n=14) had developed allergic symptoms and sensitisation to food and/or inhalant allergens during their first five years of life while non-allergic children (n=23) were non-sensitised without symptoms. Primers binding to *C. difficile, B. bifidum, B. longum/infantis, B. adolescentis, B. breve, Bacteroides fragilis,* Lactobacilli group I (*L. rhamnosus, L. paracasei, L. casei*) and Lactobacilli group II (*L. gasseri, L. johnsonii group*) were used.
Results: The most abundant bacteria were *B. longum/infantis* while few children were colonised with *C. difficile*. At one week of age none of the allergic children was colonised with Lactobacilli group I compared to 56% of the children who remained non-allergic (p=0.004). Also, at 1 month of age *B. adolescentis* was more common in non-allergic than allergic children (p=0.008). Furthermore, the persistent colonisation with these bacteria were more common among non-allergic children, p=0.018 and p=0.060 respectively.
Conclusion: A more diverse gut flora might prevent allergy development and the early colonisation might be of major importance.

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Efficacity of educational activity in the children with bronchial asthma

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Aim: To evaluate the efficacity of educational activity from School of Asthma in children with asthma and/or their family.

Methods and Materials: In the school of asthma children the specialists have performed educational activities in 172 children with different forms of asthma and their parents. Educational seminars consisted of 5 sessions which approached the following subjects: triggers, asthma symptoms, prophylaxis and asthma medicines. Each session also included individual interrogation of the trained to estimate their knowledge of asthma before and after the education session according to the theme of the working day. The quality of knowledge of some children with asthma and/or their parents was also tested after 3–12 months and afterwards visits.

Results: It was found that the knowledge- of children and parents before the training at the School of Asthma Children was incomplete and incorrect. Children with severe asthma had wrong knowledge about triggers (47,73%), about prophylaxis (52,76%) comparatively with those who suffer of mild asthma (58,06% and 60,76% respectively) and moderate (51,81% and 55,94% respectively). Knowledge of children with mild asthma referring in triggers has grown to 90,77%, of children with moderate asthma to 83,72% and those with severe asthma to 80%. Also was established a growth in knowledge about prophylaxis in all groups of children: mild asthma-83,82%, moderate asthma - 85,72%, severe asthma - 79,26%. Knowledge of children regarding clinic manifestations constituted: mild asthma - 5,5%, moderate asthma - 74,79%, severe asthma - 78,28%. Knowledge level under the subject "treatment" is the most incomplete in all groups of the questioned persons: mild asthma - 37,93%, moderate asthma - 36,96%, severe asthma - 38,78%. Towards the end of the course they were appreciated as follows: mild asthma - 63,17%, moderate asthma - 72,67%, severe asthma - 73,0%. The average of knowledge in all groups of children after 3–12 month referring triggers was 79,72%, about prophylaxis 79,01%, regarding clinic manifestations - 69,75% and referring treatment - 52,44%. For compare, the average of knowledge before educational activity was: referring triggers - 52,43%, prophylactic measures - 56,32%, clinic manifestations - 45,95% and treatment - 37,89%.

Conclusion: The knowledge of those questioned has little decreased during 3–12 months, but it remained net superior to the level before the seminars.

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The usage of paracetamole \ The subsequent development of allergy and asthma -Study of matched patients- siblings

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Background: Many studies on this field have come to the suggestion that, the intake of paracetamole during pregnancy and during the first Months of life is risky as it increase the risk of childhood asthma. Our aim is to deterring the asocial between paracetamole usage during pregnancy and the first ten moths of life and childhood allergy, allergy asthma and asthma.

Methods and Precautions: A matched patients-sibling study comparing patients with allergic asthma with their healthy siblings without any symptoms of allergic diseases. Allergic in patients and their siblings was determined by skin prick tests. We assumed that children having up to one positive skin prick test were considered to be allergic. Intake of paracetamole was assessed by standardized interviewer –administered, questionnaire.

22 pairs of allergy asthma patients' vs. non-allergic siblings were compared to determine the risk factors for allergic and asthma. While 12 pairs of allergic asthma patients vs. allergic siblings were compared to determine the risk factors for asthma. However, 35 pair of allergic asthma patients vs. non-asthmatic sibling (with and without allergy) were compared to determine the risk factors for asthma, also 15 allergic siblings were compared with 19 non-allergic siblings (with asthma) to determine the risk factors of Allergy.

Results: Intake of paracetamole during pregnancy was associated with allergic asthma ($P = 0,04$). Intake of paracetamole between birth and ten months of age and between four to seven moths of age, was also found to be associated with non-allergic asthma ($p = 0.004$ and $p = 0.03$) respectively. Usage of paracetamole during pregnancy and during early months of life may play a role in the development of allergic and non –allergic in children.

Conclusion: Intake of paracetamole during pregnancy and during first month of life is associated with an increase risk of childhood asthma.

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Cow's milk allergy in premature infants: a ready-to-use cow's milk atopy patch test before starting an amino-acid formula

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Background: Cow's milk (CM) allergy (CMA) is an infantile disease, usually appearing in the first months of life, with very few cases up to now reported in the premature infants. The study was designed to detect CMA in premature infants with digestive symptoms, based on a ready-to-use Atopy Patch Test (APT), Diallertest®.

Methods: During the year 2006, 13 premature infants (31 w±2 weeks, 4 girls), aged 42±18d , receiving formula for prematures with CM and presenting with digestive symptoms (rectal bleeding, 5, vomiting,1, diarrhea, 3, severe reflux, 2, others, 2) were tested for CMA by Diallertest®. Whatever the results, all children received an amino-acid formula (Neocate® and the outcome of symptoms was evaluated one month later.

Results: Among the 13 premature infants tested, the digestive symptoms disappeared under amino-acid formula in 10. Diallertest® was positive in 7 cases. All infants with a positive Diallertest® improved with the amino-acid formula (no false positive) and 1 child with a negative Diallertest® improved with the diet (1 false negative).

Conclusion: CMA is a frequent and mostly underestimated cause of digestive symptoms in premature infants, mostly fed with CM based formula. APT seems an appropriate method to diagnose CMA in this age range and Diallertest® might thus be a useful tool in the neonatal ward.

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Which is more important regarding psychological consequences: level of asthma control or overall disease severity in paediatric asthma?

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Aim: The study's objective was to examine the consequences of overall asthma severity and current asthma control regarding psychological symptoms in paediatric asthma.

Methods: 108 patients, age: 11.75±3.10 (mean±SD) years; (boys 11.6±2.8 years and girls 12.1±3.7 years) completed the Child Depression Inventory (CDI), the State Trait Anxiety Inventory for Children (H.STAIC), the Pediatric Asthma Quality of Life Questionnaire (PAQLQ), and a symptom score. FEV1 was also measured.

Results: Mean FEV1% was 97.4±12.8. Twenty-three patients (21%) had intermittent asthma, 40 patients (37%) had mild persistent, 43 patients (40%) had moderate persistent, 2 patients (2%) had severe persistent asthma. The only factor associated with depression is: FEV1% less than 100. Factors associated with anxiety are: night symptoms, asthma control. Factors associated with quality of life are: the age at the beginning of asthma, asthma duration, current symptoms, night symptoms, day symptoms. Factors that have no association with the above are: disease severity, other allergies, skin test positivity.

Conclusion: Not the overall disease severity, other allergic co-morbidities, but more the current asthma status and level of control alter the psychological status of the asthmatic child. They can distinguish between very small changes in the level of control. Thus professional asthma care should focus on achieving the best possible asthma control at all times.

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Changes of induced sputum cytology by leukotriene antagonists

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Inhaled corticosteroids $B!J (BICS $B!K (B and leukotriene receptor antagonists $B!J (BLTRA $B!K (Bare major controllers for bronchial asthma. In our previous observation, ICS decreased eosionophils, and increased neutrophils in induced sputum. In this study, we examined changes of induced sputum cytology after starting LTRA.

Methods: Subjects were 8 patients (mean age was 11±3 years, 6 boys and 2 girls) who were prescribed LTRA for asthma control. Before and 1 month after starting LTRA (pranlukast for 6 cases, montelukast for 2 cases), we obtained sputa by inhalation of 4.5% saline. Samples were treated for cell counts and differential cells by Eosin stain.

Results: Cell numbers before and after starting LTRA were $31\pm30\times10^4$ cells/ml, and $21\pm17\times10^4$ cells/ml, respectively (n.s.). The percentage of eosinophils were 13±11%, and 3±2% ($p<0.05$), and that of neutrophils were 79±17%, and 76±27% (n.s.).

Conclusion: LTRA decreased sputum eosinophils as well as ICS, however did not increase neutrophils.

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Abnormal lung function and bronchial hyperreactivity in early infancy – a predictor for severe respiratory syncytial virus infection?

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Background: Two-three percent of infants react with severe bronchiolitis to infection with Respiratory Syncytial Virus (RSV). The cause for such exaggerated response to a common infection is unknown, but it has been suggested that it may be associated with pre-existing abnormal pulmonary function.

Aim: To compare lung function and bronchial hyperresponsiveness in newborns who later develop severe RSV bronchiolitis and infants who do not develop such severe infection.

Methods: A prospective birth cohort study (Copenhagen Studies on Asthma in Childhood) was conducted in 411 infants of asthmatic mothers. Lung function was scheduled at 1 month of age by means of the raised volume rapid thoraco-abdominal compression technique during sedation measuring forced expiratory volume at 0.5 s (FEV0.5). Bronchial responsiveness was determined by metacholine challenge using transcutaneous oxygen as end-point (PD15-TcO2). We defined severe RSV bronchiolitis as a child admitted to hospital or requiring medication with high-dose inhaled budesonide (>800mcg) or oral prednisolone for RSV-verified bronchiolitis.

Results: In this prospective cohort study 23 children developed severe RSV infection before age 2 (mean age, 8 months). All completed baseline lung function measurements before subsequent RSV infection. Children with severe RSV infection and controls never presenting severe RSV bronchiolitis did not differ significantly at baseline lung function (FEV0.5) or metacholine challenge (log PD15-TcO2); p-value>0.1 for all comparisons.

Conclusion: Severe RSV infection in infancy was not associated with pre-existing abnormal lung function and bronchial hyperreactivity.

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A double-blind randomized placebo controlled clinical trial on the supplementation of probiotics in the first six months of life in high risk Asian infants- Effect on eczema in the first year of life

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Background: Several studies have explored the role of probiotics in the treatment or prevention of atopic diseases. The findings were however inconclusive.

Objective: To assess the effect of Bifidobacterium longum and Lactobacillus rhamnosus GG supplementation in the first 6 months of life on the incidence of eczema and allergen sensitization in the first year in high risk Asian infants.

Materials and Methods: Infants (n = 253) with a first degree relative with a family history of allergic disease and sensitization to dust mites were randomized in a double blind, placebo-controlled manner to receive a cow's milk based infant formula with or without probiotics (daily dose of $1x10^9$ colony-forming units (CFUs) of Bifidobacterium longum and Lactobacillus rhamnosus GG) for the first 6 months of life. Subjects were assessed at 1, 3, 6 and 12 months for atopic diseases and skin prick test was conducted at the 12 month visit.

Results: Out of 253 subjects, 235 were followed-up to 12 months (drop out rate 7%). The subjects in the two treatment groups were comparable in terms of demographic and birth characteristics, except for gender and birth order. Incidence of eczema in the probiotic (n = 33/122; 27%) group was found to be similar to placebo (n = 33/113; 28%) (OR, 1.07; 95% CI, 0.60 to 1.89). There was no difference in the rate of sensitization to common allergens (probiotic = 25% vs placebo = 21%, OR, 0.78; 95% CI, 0.42 to 1.45). Sensitization to dust mite allergens (Dermatophagoides pteronyssinus, Blomia tropicalis) was the most common (Probiotic = 20% vs Control = 18%), followed by eggs (Probiotic = 6% vs control = 5%). None of the subjects were sensitized to cow's milk or soy. Potential confounding factors including birth order, household size, smoking exposure, presence of pets and breastfeeding did

not affect these outcome measures. Adjustment for imbalance of gender and birth order between treatment groups did not affect the findings significantly.
Conclusion: This study did not show a protective effect of probiotic supplementation for the first 6 months of life on eczema or allergen sensitization in high risk Asian infants at 1 year of age. The prevalence of eczema in our cohort is also lower than in reported studies.

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Detection of fecal Bifidobacterium infantis in the first year of life in infants at risk of atopy supplemented with Lactobacillus rhamnosus GG and Bifidobacterium longum from birth till 6 months old

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Background: Studies have suggested that oral administration of probiotic bacteria in early life may positively modulate the immune system of infants at risk of atopy towards a non-allergic state.
Aim: This study aims to monitor the intestinal transit of a probiotic supplementation (Lactobacillus rhamnosus GG and Bifidobacterium longum BB536) in the first 6 months of life in at risk infants (first degree relative with allergic disease) participating in an ongoing randomized double-blind placebo controlled trial. Effect of this supplementation on Bifidobacterium infantis prevalence was also investigated. This particular bifidobacterial species has been implicated as a potential signature of infants at low prevalence of atopy.
Methods: Newborns at risk of allergies received either probiotic preparation (n = 55) or placebo (n = 48) daily for the first 6 months of life. Analysis of fecal bacteria and clinical examinations were conducted serially at birth, 1, 3, and 12 months. Bacterial DNA extracted from fecal samples was evaluated with nucleic acid amplification approach using specific PCR primers targeting Lactobacillus rhamnosus GG and Bifidobacterium infantis.
Results: All extracted DNA samples were amplified with universal primers targeting a conserved region of 16S rRNA to ensure extraction of intact DNA. Lactobacillus rhamnosus GG was more commonly detected in the probiotic group compared to the placebo group [at 3 days (94.1% vs 6.8%), 1-month (92.7% vs 14.9 %), 3-month (86.5% vs 21.3%, respectively) ($p<0.0001$)] but not significantly at 1-year old (28.8% vs 14.9%, $p>0.05$). Infants supplemented with probiotics were also more likely to harbour Bifidobacterium infantis at 3 months (n = 19/52; 36.5%) compared to placebo (n = 7/47; 14.9%) groups ($p = 0.0267$).
Conclusion: Our data suggests that probiotic supplementation from birth may modulate the intestinal bifidobacterial species composition. Its effect on allergy outcomes remains to be examined.

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Allergen sensitization in paediatric patients requiring hospitalisation for their severe asthma in Martinique from 2002 to 2005

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Background: Asthma is an important health care problem in tropical regions worldwide. Hospitalisations for asthma were followed from 2002 to 2005 in Fort de France, Martinique.
Objective: The main objectives of this study was to establish allergen sensitization in a large proportion of children who required hospitalisation for a severe exacerbation of their asthma. Material and Methods: Patients hospitalised at the Victor Fouche Hospital were monitored from 2002 to 2005. Patients were contacted by phone after leaving the hospital. Afterwards, they were skin tested with 16 common aeroallergens, including 4 mite species, 3 cockroaches, cat and dog, grasses and 4 foods. A detailed questionnaire concerning environmental and social risk factors was answered by the parents.
Results: A total of 632 hospitalisations were counted in this period of time. A total of 492 children (294 boys and 198 girls; mean age 5.93 years) were hospitalised. The mean number of hospitalisations per child was 1.3 (1 to 8 times). The mean days of admission per patient was 2.81 (1 to 27). The number of hospitalisations in 2002 were 156, in 2003, 191; in 2004, 148 and in 2005 137. A total of 269 patients (54.67%) (159 boys and 110 girls) agreed to undergo skin testing. From this group, 11 had been skin tested prior to the entry in the study and 3 had started, and abandoned, immunotherapy. One hundred ninety six (72.86%) children had at least 1 positive skin test; 158 (58.74%) were sensitised to at least 3 allergens. 183 (68.03%) were positive to at least 1 mite species; 99 (36.8%) to at least 1 cockroach species; 63 (23.42%) to at least one food, among these, 61 were sensitised to shrimps; 18 (6.69%) were sensitised to cat and 14 (5.2%) to dog and just 5 (1.86%) to grass pollen.
Conclusion: We have identified a high rate of sensitization to aeroallergens in a large cohort of young paediatric patients with severe asthma that required hospitalisation in Martinique. Mites, cockroaches and shrimps accounted for the largest number of sensitisations. These results are similar to those obtained in a control group of allergic children being evaluated for allergic respiratory diseases during the same period of time.

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Specific IgE levels to ascaris spp. in a cohort of paediatric patients hospitalised for severe asthma in Martinique from 2002 to 2005

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Background: In tropical regions, there is a large proportion of the population which has been in contact with intestinal parasites, including the roundworm Ascaris spp. There are conflicting reports on the protective effect or harmful influence of sensitization to Ascaris spp. in patients with allergic respiratory diseases, such as allergic asthma. Objectives: to investigate specific IgE levels in children hospitalised with severe asthma in the tropical island of Martinique.
Materials and Methods: From 2002 to 2005 we investigated specific IgE levels in the serum of children hospitalised for severe asthma in Fort de France, Martinique. As controls we used a group of 484 children (mean age 7.42) evaluated for allergic respiratory diseases at an outpatient clinic. Specific IgE to Ascaris spp. was measured by the CAP system (Phadia).
Results: A total of 117 children (68 boys and 49 girls; mean age 6.45) were investigated. Mean number of hospitalisations and of days stayed in the hospital were 1.35 and 3.02 days, respectively. A total of 60 children (51.28%) had a positive determination (mean value 6.69 kU/L). The mean number of hospitalisation in the positive group was 1.28 and the mean days stayed at the hospital, 3.05 days. 47 children stayed 3 or more days in the hospital; in this group there 22 Ascaris spp. negative and 25 Ascaris spp. positive children (NS). In the negative group, the mean number of hospitalisation was 1.42 times and the mean days stayed at the hospital, 2.98 days. In the control group, 215 (44.42%) had a positive specific IgE determination to Ascaris spp.
Conclusion: In contrast to what has been proposed for other tropical countries, sensitization to Ascaris spp. seems not to influence the severity of asthma in Martinique. Similar rates of sensitization to Ascaris spp. are

detected in allergic children who do not require hospitalisation for their allergic asthma.

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Demographic of monitoring and treatment of childhood asthma in Surakarta, Central Java, Indonesia

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Background: In Surakarta, one of the districts in Central Java, Indonesia, with a population of around one million, many areas house industrial companies, leading to high pollution, that can trigger childhood asthma. As the most common chronic illness in children, childhood asthma causes school absences and limitations of children's activities, making asthma especially serious for them.

Methods: Standardized questionnaires were distributed randomly to 200 doctors in Surakarta, who are general physicians, general pediatricians and pediatric pulmonologist.

Results: Of the 200 questionnaires, 78 were returned: 74 from general physicians and 3 from pediatricians. Most of the doctors never use score cards to monitor childhood asthma, never used spirometry and never used peak flow meters. For the treatment of acute asthma methylprednisolone was first choice in 83%, while 17 % used dexamethasone. For maintenance treatment of childhood asthma no physician used montelukast because this drug is not available in Surakarta. However, as a first choice maintenance treatment, most physicians used a long-acting beta-agonist (LABA) in combination with an inhaled corticosteroid in infants, preschoolers and older children.

Conclusion: Striking results were the low usage of score cards, lung function testing in monitoring childhood asthma, and the high usage of a LABA in combination with inhaled corticosteroids as first choice maintenance treatment, suggesting that guidelines on asthma monitoring and treatment are not strictly followed in Surakarta.

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Frequency of infections, atopic eczema and asthma outcome in infancy

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Studies of association between frequent early infection and allergic disease outcomes have produced diametrically opposed results. Some have associated reduced frequency and others increased frequency of infection with more allergic disease. We have employed a unique algorithm (Business EffiScience) to the analysis of a large database from early prevention of atopic asthma in children (EPAAC) trial. 510 infants between 1–2 years of age with atopic eczema (SCORAD> 10) entered the study and 434 completed 18 months follow up. As there was no difference between active and placebo intervention all subjects were merged for the purposes of this analysis which were to establish which factors at recruitment either singly or in combination had the greatest impact on the subsequent development of asthma defined as greater than or equal to three episodes of wheezing and/or paroxysmal cough disturbing sleep for at least three consecutive nights. To analyze the impact of early infection on asthma we analysed the effect of the number of infections per year of infants from birth to asthma onset or end of the study when asthma was not observed. A bell shaped curve was identified with 27% infants less often developing asthma if they had 1.5 or less infections per year, 25% more often to have asthma with 1.5 to 4 infections per year and 11% less often to have asthma with more than 4 infections per year. The only other interaction was with proximity to a factory as a surrogate for pollution exposure which only increased the risk of asthma if there was a frequency of infections between two and five per year. Thus the combination of atopic eczema with raised IgE and proximity to a factory was associated with a 77% uplift in frequency of developing asthma if the frequency of infections was between 1.5 and 4 per year. The bell shaped curve of frequency of infection in relation to asthma risk may well explain the discrepant results in various studies. Very low frequency of infection occurs in infants with an effective TH1 response and therefore reduced probability of developing allergic disease while those with a very high frequency of infection have a greater probability of inducing TH1 responsiveness while normal rates of infection in an atopically predisposed individual will not modify the outcome.

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Challenges of 'peanut challenge'

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Background: The diagnosis of peanut allergy has important consequences for patients and their families. An accurate diagnosis of peanut allergy is essential as it is usually life long and can potentially be fatal. The cornerstones of diagnosis are a detailed history, skin prick tests (SPT), IgE estimation with or without an oral peanut challenge.

Objective: 1. To demonstrate the importance of peanut challenges in diagnosing children with peanut allergy 2.To see whether we can predict children who are likely to outgrow peanut allergy 3.To assess the safety of peanut challenges.

Methods: A retrospective study of peanut challenges performed at a tertiary care paediatric hospital allergy clinic between January 1997 and December 2006.

Results: Of the 79 peanut challenges performed, (data available for 77), 14 (18%) were positive. All children had a history suggestive of peanut allergy and 72 had a skin prick test (SPT) or IgE RAST performed prior to the challenge (SPT-63, IgE-6, SPT and IgE-3) The results (72) of SPT±IgE RAST: •23 were negative (SPT-20, IgE-2, SPT+IgE-1) •49 were either positive for RAST or SPT or both (IgE-4, SPT-43, both-2)

SPT (mm) n=66	Positive challenge n=14	Negative challenge n=52
≥10 mm	3	2
6 – 9 mm	7	7
≤5 mm	3	21
negative	1	22

Only 1 child who had a negative SPT developed a positive challenge (4%), whereas 36/49(73%) children with positive SPT or RAST had a negative challenge. Further analysis of these children who had a negative challenge shows that majority had a SPT of ≤ 5 mm (21/36), where as 7 had SPT of 6–9mm, 2 had SPT of ≥ 10mm. 3 had positive IgE RAST and 2 had both SPT and IgE RAST positive. SPTs ≤ 5 mm was strongly associated with a negative peanut challenge; Fishers Exact test p = 0.02 Adverse clinical effects of positive peanut challenges included urticaria, lip swelling, vomiting and in one anaphylaxis; 6 required medical treatment (5- antihistamines, 1- adrenaline).

Conclusion: The majority of children with a diagnosis of peanut allergy, based on history and skin prick / antibody tests had a negative challenge. Peanut challenges are not warranted in patients with a definite history of peanut anaphylaxis with positive SPT. We recommend that where the history is suggestive but SPT is negative or borderline positive, i.e. ≤ 5 mm, a peanut challenge is necessary to confirm the diagnosis. We plan to SPT and RAST

test all patients before they have peanut challenge to see if this improves sensitivity and specificity of our challenges.

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Effect of lysed Enterococcus faecalis FK-23 (LFK) on intestinal microflora in antibiotic treated mice

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Background: Antibiotic use in infancy may be associated with an increased risk of developing allergic diseases. Lysed Enterococcus faecalis FK-23 (LFK) could suppress the allergic responses; however, the mechanism responsible for this phenomenon remains unclear.
Objective: To evaluate the effect of LFK on intestinal microflora in antibiotic treated weaning mice.
Methods: Three-week-old BALB/c mice were sensitized with cedar pollen allergen to establish an experimental model. Orally administered erythromycin, one kind of macrolide antibiotic, was used for the experiments. The intestinal microbiota, and the allergen-induced accumulation of eosinophils and IgE level in sera were determined in the control, antibiotic, LFK and antibiotic-LFK groups (n = 7, respectively).
Results: The total aerobes, total anaerobes and Enterococcus of intestinal microflora were not significantly different among all groups. Lactobacillus was distinctly eliminated in the mice exposed to erythromycin on day 7 and totally recovered in erythromycin-treated mice with LFK intervention on day 28, but could not recovered in the erythromycin-treated mice without LFK intervention.
Conclusion: LFK improved the intestinal ecosystem disturbed by antibiotic use, and may prevent subsequent development of allergy.

AUTOIMMUNITY

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Anti-neutrophil cytoplasmic antibodies in patients with pulmonary tuberculosis

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Background: Mycobacterial tuberculosis is a major cause of mortality and morbidity worldwide.Infection with this bacteria is known to induce the development of autoantibodies and a few of these antibodies are also known to be diagnostic markers for some other diseases. ANCA'S are one of autoantibodies used in clinical setting for diagnosing systemic vasculitic syndrome . More than 20 studies investigating ANCA positivity in diseases other than small vessel vasculitis . This study was undertaken to determine the prevalence of ANCA in pulmonary tuberculosis (TB) which could lead to false diagnosis of wegner's granulomatosis or vice versa.
Materials and Methods: In a case–control study 32 consecutive smear positive pulmonary TB patients and 32 normal individuals were studied . All cases and controls screened for ANCA by indirect immunoflurecent (IIF) and also myeloperoxidase antibodies (anti-MPO) and proteinase 3 antibodies (anti-PR3) were tested by ELISA.
Results: Perinuclear pattern (P-ANCA) was detected in 25% of cases and 6.25% of controls and cytoplasmic pattern(C-ANCA) in 3.1% of both cases and controls by IIF assay. ANCA specificities by ELISA in cases revealed that 75% had anti-myleperoxidase and 12.5% had anti-Proteinase 3. In controls 3.12% had anti-MPO and no person had anti – PR3 . The positive ANCA significantly correlated with tuberculosis (p value<0.018) and also positive anti-MPO significantly correlated with TB (p value<0.01).
Conclusion: Autoantibodies especially ANCA'S may present in both disease, Wegner Granulomatosis and Tuberculosis .The presence of autoantibodies in TB patient could have a multifactorial etiology. The presence of human T lymphocytes reactive to heat stress proteins may be an important target of immune response against certain intracellular autoantigens such as anti-MPO from PMN added to the mechanism of molecular mimicry would explain the association of ANCA and TB.

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The cells in psoriatic skin

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Rationale: The psoriasis is an autoimmune disease with significant role of the immune system.
Methods: We investigated the group of 16 patients with active form of chronic psoriasis vulgaris before and after treatment with cyclosporine A. The results was compared with the control group of the patients without any skin disease. In the samples of the skin tissue we detected the cells with surface markers CD14, CD45, CD3, CD19, CD46+56, CD4, CD8, HLA-DR, CD25 (we have used the monoclonal antibodies Immunotech, Becton-Dickinson).
Results: The lymphocytes represent 7,3% in the samples of the skin in patients in the active state of psoriasis, 2,1% in control group (p<0,0001) and 3,1% in patients after treatment (p<0,001).Cytotoxic T-cells (CD3+CD8+) represent 37,8% in the samples of the skin tissue in psoriatic patients, 32,4% in control group and 28,4% after treatment, The cytotoxic T-cells expressed HLA-DR in 25,6% in psoriatic patients before treatment, in 10,7% after treatment and in 11,4% in the control group. We found 44,3% of the lymphocytes with CD3+CD4-CD8-phenotype in the skin of patients before treatment, 29,6% (p<0,05) after treatment and 10,3% in the control group. The expression of the surface marker CD25 was lower in the active stage in comparison with control group (9,6% vs 13,1%), its expression increased after treatment at 38,4%.
Conclusion: The activated cytotoxic T-cells and T-cells with ãä-TCR-receptors (CD3+CD4-CD8-) play the important role in pathogenesis of psoriasis. The results of our study confirmed possible regulation role of the T-cells with CD4+CD25+phenotype.

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Anti-endotoxin immune responses in systemic lupus erythematosus and rheumatoid arthritis

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Introduction: Rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) are autoimmune diseases with respectively T cell mediated and B cell mediated mechanisms. Endotoxin (ET) from gram negative flora can rapidly induce or reactivate experimental arthritis, probably by stimulation of secretion of Th1 inflammatory cytokines. ET is a potent polyclonal B-lymphocyte activator. This study investigates anti-endotoxin humoral immunity in patients with RA and SLE.
Methods: 27 patients were studied including: Group I-18 patients with RA; and, Group II-9 patients with SLE. The age range was 30–50 years. Disease duration was from 6 months to 10 years. Disease status based on clinical, including joint involvement, and laboratory data were less than or equal to class II degree of activity. Peripheral blood was obtained and serum analyzed.

Quantification of IgA, IgM, and IgG anti-ET antibodies was done by ELISA. ET from Esherichia coli K30 (09:K; O:H12) was employed as the antigen. The control group included 32 healthy donors.

Results: Patients with RA had levels of IgA anti-ET and IgM anti-ET which were 2.3 fold less ($p<0.001$) than the average in healthy controls. The levels of IgG anti-ET antibodies were not different from the healthy control group. Patients with SLE had levels of IgG anti-ET antibodies which were 2.2 fold greater ($p<0.01$) than normal controls. These same SLE patients had levels of IgA anti-ET and IgM anti-ET which were respectively 2.3 ($p<0.001$) and 2.1 ($p<0.01$) fold less than in the healthy control group. Reduced IgA anti-ET levels occurred in 56.7% ($p<0.001$) and reduced IgM anti-ET levels occurred in 53.6% ($p<0.01$) of SLE patients. Reduced IgA anti-ET levels occurred in 56.7% ($p<0.001$) and reduced IgM anti-ET levels occurred in 56% ($p<0.001$) of RA patients.

Conclusion: Anti-ET IgG levels in RA patients demonstrate a normal level of response while in SLE patients greater than normal responses to ET occur Dysfunctional anti-ET immune responses occurred with respect to reduced IgM and IgA anti-ET antibodies in both SLE and RA patients. There may be a role for gut flora and immune responses to ET as factors modulating immune responses in RA and SLE patients.

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Activity of systemic lupus erythematosus in Mexican children. Correlation between medical clinical evaluation and evaluation based on the use of five indicators of activity

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A computer program has been developed to evaluate five of the most important SLE AI: SLEDAI, SLAM, BILAG, LAI, ECLAM. These indicators have a close correlation with each other in the case of adult patients and also, when they are compared with the expert opinion. On the contrary there is sparse information of the usefulness of these indicators in children with SLE.

Objective: To determine whether is there a correlation between 5 different SLE AI in children and to assess the correlation between the clinical evaluation of activity by physicians of the Department of Immunology and that provided by the use of the five AI of SLE.

Materials and Methods: A transverse analytical study was undertaken in patients under 18 years of age diagnosed with SLE at the National Institute of Pediatrics in Mexico City. A questionnaire was used containing every variable included in the five indices of SLE activity. Statistical analysis. The median and interquartil limits were used as descriptive ststistics because the variables did not show a normal distribution with the Kolmogorov-Smirnov test;. To ascertain the degree of association among quantitative variables, correlation test of Spearman was used. Sensibility, specificity and predictive values were estimated at different cut-off levels selecting the best one using Receptor Operator Curves (ROC). Results. We included 30 patients with SLE; 28 females (94%) and two males. Median age was 15 years (liq 13-17). The median time of evolution from the time of diagnosis was 41 months (liq 16-5). According to the expert 14 patients had active SLE (47%); according to the SLEDAI 13 patients had active SLE (43%) ($p = 0.79$).

Conclusion: The indices with the greater correlation with the SLEDAI were the opinion of the expert and the SIS; those that best correlated with the appraisal of the clinician were the SLEDAI and the SIS. Sensibility and specificity for each one of them were 92% and 89% for the expert; 85% and 88% for the SIS and 85% and 89% for the SLEDAI respectively.

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The value of the *corticosteroids(CSP) in countering the residua of the protracted tuberculous infection/inflammation(TI)

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Purpose: (CS) "The double edge sword" in function the favorable therapeutic advantage against disadvantage should be weighed when the indications arise.

Methods: (TI) being a lingering process is associated with an enhanced degree of exudation,caseation fibrogenesis etc.there may be binding ,obstruction.constrictoion,compression of the surrounding inflamed surfaces,also the constitutional symptoms as a consequence of the release of the inflammatory products.Exudative changes in the cerebrospinal fluid pathways were associated with features of space occupying lesions. (TI) as pleural, pericardial and peritoneal; effusions were associated with fibrous bands formation, also exudative changes in and around the inflammed joint spaces may lead to interference with the function of these structures. In all these cases corticosteroids with in the therapeutic dose had been associated with a remarkable degree of improvement without any fibrous residua and prodromal manifestations were relieved favourably. In relatively seriously ill patients also patients with in the terminal stages of illness (at the verge of death) from advanced tuberculosis and inanition,corticosteroids may be indicated purely on humanitarian grounds as these(CS) detoxicate the patients and allow the chemotherapeutic agent to take their principal effects. Dosage regimen for adults 40mg/day in divided dose for the first day then 20mg/day in divided dose for 3–6weeks to be tapered by 5mg at 3–5 days intervals.For children 2mg/kg/day for those less 2 years, 1.5mg/kg/day for those more than 2years but less than 10 years age.

Results: Patients with a reasonably certain degree of diagnosis of (TI), (CS) in concomitant anti-tuberculosis drugs had a remarkable therapeutic response evidenced as fall of temperature ,improvement in appetite and vigor.

Conclusion: With un-certain diagnosis of (TI), (CS) may mask the untowards effect of chemotherapeutic agents,creating a false sense of improvement and may precipitate steroid dependence.

Clinical Implication: The safety of (CS) is only out of question,if these had been indicated for the shortest possible time and then tailed off in a step ladder pattern.*Prednisolon sulphate.

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The prophylactic value of the low dose intravenous immunoglobulin(IVIG) in reducing the incidence of the infection in chronic lymphatic leukemia(CLL) and secondary hypogammaglobulinaemia

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Objective: In the study concerened low dose of IVIG was as effective as high dose in reducing the risk of infection in patients with Chronic Lymphatic Leukemia(CLL) and secondary Hypogammaglobulinaemia without the inclusion of the control group, behind the idea to assess the IVIG superiority over the empirical therapy.

Materials and Methods: 5 Patients with CLL with the background of hypogammaglobulinaemia(IgG<600mg/dl) with a history of 2 infection episodes in almost 6 months were included, all had been randomly allocated to receive regularly an infusion of 300mg/kg IVIG every 4 weeks for 6 months then for financial reasons 4 could not afford the expense while 1 was continued with the ongoing IVIG schedule.

Results: In response to IVIG there were immediate and accumulative increase in the serum IgG levels and an associated decrease in total and serious infections. In the 12 months study only 4 incidence of infection (1 with severe and 3mild) with 1 neutropenia observed. Four patients having completed 6months IVIG had an overall 50% reduction in the incidence of infection, while 1 patient with 12 months IVIG had 65%reduction in the incidence of the infection.

Conclusion: Infection as the main cause of morbidity and mortality consequent upon the hypogammaglobulinaemia with the background of cell mediated immunity was associate with protracted cytotoxic drugs therapy. In the trials concerened, the protective values of IVIG had been time related than dose related (longer the period of IVIG therapy les was the incidence of infection). Bearing in mind the cost, IVIG is only advisable in pateints with life threatening infection and then too with lower dose to reduce the expense incur upon the treatment. khan et al aljh nsr nwfp pk.

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A six year old girl with recurrent varicella infections associated with a mutation in CD16 (FCGR3A, FcyRIIIA)

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Background: Natural killer (NK) cells are a subset of lymphocytes that play a pivotal role in innate immunity. NK cells recognize antibody-coated target cells using CD16 (FcyRIIIA), and kill the target cells by the process called antibody-dependent cell-mediated cytotoxicity. Quantitative and qualitative defects in NK cell function result in susceptibility to intracellular microbes.
Methods: Lymphocyte subpopulations in peripheral blood were determined using flow cytometry. NK cell surface expression of CD56 and CD16 and the intracellular expression levels of perforin and granzyme B were analyzed by flow cytometry. Patient cDNA for CD16 was sequenced to screen for mutations within the coding region.
Results: The patient is a 6 year-old girl who suffered from recurrent upper and lower respiratory tract infections. She had experienced two episodes of varicella despite receiving the vaccine, and, despite this, varicella antibody was negative. She also has numerous warts on her fingers. Immunologic studies revealed low serum immunoglobulins (IgG 413, IgA 38, and IgM 37 mg/dL at 4½ years), but she had exuberant antibody production in response to pneumococcal vaccine. Peripheral blood lymphocyte phenotyping revealed slightly low T cell numbers for age with preservation of the CD4/CD8 ratio, B cells were normal in percentage and number. CD56+ NK cells were 6 % of the lymphocyte gate, but the expression of CD16+ was less than 1 % using Leu11c/B73.1. However, analysis of NK cells using a different antibody to CD16, NKP46/9E2, revealed normal CD16 surface expression. Sequencing of peripheral blood cDNA demonstrated a homozygous TA transversion at base pair 489 in the CD16 mRNA sequence producing a Leu to His substitution at residue 102 in the peptide sequence, a mutation previously described in a patient with recurrent orolabial HSV exacerbations and respiratory infections (Jawahar et al 1996, Clin. Exp. Immunol. 103:408).
Conclusion: Patients with recurrent respiratory infections and panhypogammaglobulin-emia with exuberant polysaccharide antibody responses may have mutations in CD16 and increased susceptibility to viral infections, especially herpes viruses.

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Cicatricial pemphigoid

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Background: Cicatricial pemphigoid (CP) is a rare autoimmune subepithelial blistering disorder, characterized by predominant involvement of the mucous membrane and occasionally the skin, chronic course, and tendency towards scarring of the affected areas. Most common sites of involvement are oral mucosa and conjunctivae. In our hospital, this is the second case of CP, and the first case with skin involvement for the last five years.
Case: A 72 years old male had undergone dysphagia and blistering disorder in the skin for six month. Clinical manifestation were erythematous and erosions in oral mucous, tongue, and crust in lips. Skin lesions were tense hemorrhagic blisters with negative Nikolsky sign, erosions and crust in chest, axilae, abdomen and left lower arm. Few slight atrophic scars were also found. Pharyngoscopy showed erythematous and brittle of the pharynx, soft palate and tongue. The histopathology examination showed subepidermal bullae, epidermal atrophy, sparse perivascular and periappendageal inflammatory infiltrates consisted of mostly lymphocytes with few polymorphonuclear leukocytes and eosinophils and fibrosis in the dermis. The patient favorably responded to oral steroid, prednisone 45 mg daily in combination with dapsone 100 mg daily, and improved after seven weeks.
Discussion: CP is a rare autoimmune subephitelial blistering disorder, characterized by predominant involvement of the mucous membrane. According to the literature, skin involvement in CP occurs in about one-fourth of cases. Diagnosis in this case was suggested by the presence of scarring lesions in pharyngeal mucous membrane with dysphagia, slight atrophic scar in the skin and confirm by histopathology. Prednisone and dapsone are used due to severe disease and to prevent potential severe complication.
Conclusion: CP with skin involvement is uncommon. Diagnosis is based on clinical manifestations and, confirmed by histopathology. Therapy with prednisone and dapsone shows good result in CP.
Keywords: cicatricial pemphigoid, skin involvement, oral steroid, dapsone

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Prevalence of IgA deficiency in individuals with autoimmune thyroid diseases and DM TI in Iceland

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Introduction: IgA deficiency (IgAD) is the most common immunoglobulin deficiency and one of the most frequent primary immunodeficiencies. The prevalence of IgAD in Icelandic blood donors has recently been estimated to be 1:570. Correlations between having IgAD and various autoimmune diseases, including autoimmune thyroid disease (AITD), have been rapported.
Materials and Methods: IgA was measured in serum from 319 patients with thyrotoxicosis and 199 patients with DM TI. In comparison IgA levels from 609 blood donors was evaluated. IgA was measured with Beckman Array 360 System Nephelometry. IgA levels were also studied in relation to titers of TSH Receptor Antibodies (TRAb) and Thyroid Peroxidase Antibodies (anti TPO). TRAb was measured with ELISA and anti TPO was measured using FEIA.
Results: None of the 319 thyrotoxicosis patients had IgAD (IgA in serum < 0,05 g/L). Women with thyrotoxicosis had significantly lower levels of serum IgA compared to men with thyrotoxicosis ([IgA] male = 2,67 ± 1,16 g/L vs. [IgA] female = 2,11 ± 1,04 g/L; P<0,001). However, similar gender difference was also found in the blood donors. IgA levels increased significantly with age in patients with thyrotoxicosis (P<0,01) and a similar correlation was found in the blood donor group. A negative correlation was found between levels of IgA in serum and TRAb (correlation coefficient=−0.322, P = 0,0455) and also between levels of IgA and antiTPO (correlation coefficient=−0,376, P = 0,0204). One individual with DM TI had hypogammaglobulinemia, but none had selective IgAD in that cohort. Interestingly, men with DM TI had lower serum IgA than women.
Conclusion: These results are a strong indication that the prevalence of IgAD is not increased amont patients with thyrotoxicosis or DM TI in Iceland. The

negative correlation between IgA levels and thyroid autoantibodies suggests a possible role of IgA in the pathogenesis of autoimmune thyroid diseases. IgA might have a protective role in the formation of autoantibodies against thyroid structures or a mutual autoimmune mechanism might cause brake-down of IgA and the formation of thyroid autoantibodies.

POSTER GROUP 2–TUESDAY

ALLERGIC & IMMUNOLOGIC MECHANISMS

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Airway inflammations induced by allergen with LPS are partially dependent on IL-12, but completely dependent on STAT4 signalling

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Background: Asthma is a chronic inflammatory disorder of the airways associated with reversible airway obstruction and airway hyperresponsiveness. It has been thought that Th2 response play a critical role in the pathogenesis of asthma. However, recently, there have been reported that Th1 response also play a important role in pathogenesis of asthma. In Th1 immune response, IL-12 and STAT4 are key molecules. IL-12 play a important role in Th1 development, and promotion of Th1 responses and the production of interferon-gamma. And STAT4 are believed that it also play a important role in Th1 immune rsponse. But it still remains unclear whether these molecules play key roles in asthma development *in vivo.*

Methods: To evaluate the roles of IL-12 and STAT-4 in murine model of asthma, 6-week-old IL-12 receptor β2 (−/−), STAT4 (−/−) null mutant, and their littermate wild type control mice were used, respectively. The mice were sensitized four times with 75μg of OVA at days 0, 1, 2, and 7 in the presence or absence of 10μg of LPS, and then challenged 4 times intranasally with 50μg of OVA at days 14, 15, 21, and 22. STAT4 (−/−) and wild type control mice were sensitized and the challenged in the same manner. After challenging, we tested the airway hyper-responsiveness (AHR) against methacholine challenge. The mice were sacrificed 48 hours after last allergen challeng, and various immunological parameters were evaluated such as BAL fluids cellularity.

Results: Total cell number of BAL fluids was increased in the mice sensitized with LPS and OVA compared with mice sensitized with OVA only. BAL fluids flammatory cells were similar between IL-12 receptor ¥β2 knock-out mice and wild type mice, but significantly decreased in STAT4 null mutant mice. Interestingly, IP-10 level in BAL fluids was partially decreased in IL-12 (−/−) mice, but markedly decreased in STAT4 (−/−) mice.

Conclusion: In Th1 experimental asthma, airway inflammatory responses are partially dependent on IL-12, but dependent on STAT4 signaling pathway.

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The role of the sensitized alveolar macrophage in T cell proliferation

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Background: It is well known that alveolar macrophage (AM), which composes more than 90% of the bronchial alveolar cells, is the first barrier protecting lung from the harmful antigens. However, few different studies suggest that sensitized alveolar macrophage could exacerbate the asthma phenotype such as airwayhyperresponsiveness (AHR). The exact roles of alveolar macrophages in asthma have been unknown. Thus we undertook to determine whether sensitized alveolar macrophage could induce T cell proliferation in asthma animal model.

Methods: BALB/C mice were intraperitoneally sensitized by ovalbumin with alum on day1 and 7, followed by challenge with 1% ovalbumin on day 21, 22, and 23. The lung cells were isolated and analysed by FACS analysis. T cells isolated from the bronchial lymph node and bronchial alveolar lavage (BAL) cells were cocultured with or without ovalbumin in vitro in order to determine ability of sensitized alveolar macrophages to induce T cell proliferation.

Results: After challenge, the percent of CD11chigh/CD11blow lung cells (alveolar macrophage) were not changed, but the MHC II expression of CD11chigh/CD11blow lung cells increased compared to PBS group. The proliferation of T cells increased 3 times in IP group than other groups without stimulation of ovalbumin. Furthermore, the BAL cells of IP group with stimulation of ovalbumin induced 8 times increase of T cell proliferation compared to that of other groups.

Conclusion: It is suggested that the sensitized alveolar macrophages in BAL could facilitate the proliferation of T cells in asthma animal model.

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Toll-like receptor 2, 4, and 6 expression and function in peripheral blood mononuclear cells from asthma patients

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Background: Asthma is a chronic inflammatory lung disease that is caused by impairment of adaptive immune system like many immunologic diseases. However, the pathogenesis of this unwanted response has not well determined. Toll-like receptors have been shown to play a pivotal role in both innate and adaptive immune responses. Recently, many research groups have hypothesized that these TLRs are likely to play important roles in asthma pathogenesis, but, most studies are focused on the effect of TLR polymorphism. Therefore, we investigated whether the expressions of several TLRs are different in asthma patients compared to normal subjects and if so, whether the functionality of these receptors could be related to this change.

Methods: The expressions of TLR2, 4, and TLR6 on peripheral blood mononuclear cells (PBMCs) from asthma patients and normal subjects were analyzed by flow cytometry. To study the functional responses of these receptors, PBMCs were stimulated with PGN and Pam3Cys as TLR2 ligands, LPS as a TLR4 ligand or FSL-1 as a TLR2/6 ligand for 24 hours and the amounts of TNF-α and IL-1β were determined by ELISA.

Results: The expression of TLR2 was up-regulated on PBMCs from asthma patient as compared to normal subject. Upon stimulation with PGN or Pam3Cys, TNF-α and IL-1β production significantly increased in asthma patients. In contrast, the response to LPS (TLR4 ligand) stimulation on PBMCs was higher in normal subject than asthma patients, although the expression of TLR4 was not significantly different between asthma patients and normal subject. In case of TLR6, surface expression was significantly reduced in asthma patients. However, no difference was observed in the amounts of TNF-α and IL-1βsecreted from PBMCs treated with FSL-1.

Conclusion: Our data suggest that the difference of the phonotypical expression and the functional responsiveness of TLRs might be related with pathogenesis of asthma.

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Influences of anti-IgE antibody omalizumab on allergen-induced airway inflammation and bronchial hyperresponsiveness in murine models of asthma

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Background: The aim of this study was to examine the effects of treatment with omalizumab, an anti-immunoglobulin E antibody, on allergic-airway responses in mice after inhalation of the naturally occurring aeroallergen Aspergillus fumigatus (Af) and to examine the effects of omalizumab on specific immune responses to a defined protein antigen with the use of an ovalbumin (OVA) model of asthma.
Methods: Mice were subjected either to repeated, intranasal application of Af extract or to intraperitoneal immunization with OVA, followed by inhalation challenge. Omalizumab or a control fluid was given daily throughout the sensitization process. Immunoglobulin E (IgE) levels, bronchoalveolar lavage-fluid cytokines and cytology, lung histology, and physiologic responses to methacholine were assessed in the allergen-treated mice. Anti-OVA IgE responses and OVA-driven T-cell cytokine production were examined.
Results: Treatment with omalizumab did inhibit bronchial inflammation and bronchial hyperresponsiveness in both Af- and OVA-treated mice. This inhibition required that omalizumab be administered concurrently with allergen sensitization, indicating that the attenuation of bronchial hyperresponsiveness and inflammation was not caused by anticholinergic receptor effects. OVA-responsive T cells from omalizumab-treated mice exhibited depressed production of IL-4, IL-5, and IL-13 and normal amounts of interferon-gamma. The amounts of IL-5 and IL-13 were also diminished in the bronchoalveolar lavage fluid.
Conclusion: Omalizumab, given at the time of exposure to the allergen, inhibits the induction of allergic pulmonary inflammation, and bronchial hyperresponsiveness. These results suggest that omalizumab or similar agents given during times of antigen exposure might alter disease progression in patients with respiratory allergy.

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The non-proteolytic major house dust mite allergen Der p 2 induce proasthmatic responses in bronchial epithelial cells partly through NF-κB and MAPK pathways

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Background: House dust mites (HDM) belong to the most common sources of airborne allergens worldwide and sensitization to them is strongly associated with development of allergic airway diseases. Many proteolytic HDM allergens can activate respiratory epithelial cells to produce pro-inflammatory mediators. In contrast there is limited knowledge regarding potential similar effects of non-proteolytic allergens, although they include many major allergens. One of them is Der p 2 of Dermatophagoides pteronyssinus. To investigate whether Der p 2 activate respiratory epithelial cells to produce mediators involved in pathogenesis of allergic asthma and to elucidate the mechanism of such activation, we exposed the human bronchial epithelial cell line BEAS-2B to this allergen.
Methods: Bronchial BEAS-2B cells were exposed to recombinant Der p 2 (1-80 μg/ml). After 2-24 hours the mRNA levels and the secreted amount of soluble mediators as well as the expression of cell adhesion receptors involved in recruitment, survival and binding of inflammatory cells, for instance GM-CSF, IL-6, IL-8, MCP-1, MIP-3α and ICAM-1, were analyzed. After exposure the adhesion of leukocyte cells U937 to the epithelial cells was also studied. In order to study if the activation was dependent on signalling through NF-κB and the MAP kinases ERK1/2, p38 or JNK specific inhibitors were used.
Results: Der p 2 induced dose-dependent up-regulation in gene expression and protein secretion of granulocyte-macrophage colony-stimulating factor GM-CSF, IL-6, IL-8, MCP-1 and MIP-3α. Expression of ICAM-1 was also up-regulated, which was associated with a subsequent increased adhesion of leukocytes to the epithelial cells. These responses were partly dependent on NF-κB and MAPK activation, since the specific inhibitors reduced the activation.
Conclusion: Taken together these results imply that Der p 2 may potentiate asthmatic responses in airways by direct activation of lung epithelial cells in a protease-independent manner.

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House dust mite (Dermatophagoides pteronyssinus) increases expression of pro-inflammatory cytokines in the human eosinophil cell line, EoL-cells

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Human eosinophils play important roles in the pathogenesis of allergic diseases, specifically asthma. Interleukin 6, IL-8 (CXCL8), and Monocyte chemotactic protein 1 (MCP-1/CCL2) play a pivotal role in mediating the infiltration and activation of immune cells into pathogenic lesions, including the lung and skin. The aim of this study was to examine whether the house dust mite Dermatophagoides pteronissinus extract (DpE) affects the mRNA and protein expression of IL-6, IL-8 and MCP-1 in the human eosinophil cell line, EoL-1 by performing RT-PCR and ELISA. DpE increased the mRNA and protein expression of IL-6, IL-8 and MCP-1 in EoL-1 cells. The increased expression of MCP-1 and IL-8 was inhibited by PP2, an inhibitor of Src, rottlerin, an inhibitor of protein kinase C δ (PKCδ), and PD98059, an upstream inhibitor of extracellular-signal-regulated kinase (ERK). It indicates that DpE increases MCP-1 and IL-8 expression through Src, PKC δ and ERK. We also found that IL-6 expression due to DpE was related to Src, PKC δ, and p38 mitogen-activated protein kinase (MAPK). In early signal pathway, the expressions of IL-6, IL-8 and MCP-1 are regulated by Src family tyrosine kinase and PKC δ pathway activated by DpE. This finding may contribute to the elucidation of the pathogenic mechanism triggered by DpE.

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Perturbations of NK cell regulatory functions in respiratory allergic diseases

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Background: Allergic diseases are characterized by abnormal responses to allergens favored by an inappropriate regulation of the Th1-Th2 polarization. NK cells give rise to a complex NK/DC cross-talk that would help Th1 response. By analyzing Peripheral Blood NK cells from 12 patients suffering from either allergic rhinitis or rhinitis and intermittent asthma, we evaluated whether, these cells were impaired in their ability to interact with DC.
Methods: Different circulating NK cell subsets were analyzed by flow cytofluorimetry. Mixed NK/DC cultures were performed to assess the reciprocal functional interactions. NK cells were analyzed for their ability to induce DC maturation and cytokine production, and to kill immature DC. In addition, DC were assessed for their ability to induce cytokine production by NK cells.

Results: We first analyzed the CD56+CD16+/−cells: a subset of circulating NK cells that is able to respond to DC by proliferating and producing IFN-γ. Our analysis revealed that this NK cell subpopulation was sharply reduced in most patients. This was reflected by reduced NK cell-mediated IFN-γ production in response to DC. Also the capability of promoting DC maturation and/or to kill immature DC, a function sustained by CD56+ CD16+NK cells, was reduced in most patients.

Conclusion: We suggest that allergic diseases are accompanied by a partial impairment of the NK cell capability of promoting and maintaining appropriate Th1 responses.

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Humoral and cellular immune responses to blomia tropicalis and its concanavalin a-binding fractions in atopic patients

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Background: *Blomia tropicalis* (Bt), *Dermatophagoides pteronyssinus* (Dp) and *D. farinae* (Df) are the prevalent house dust mites (HDMs). Concanavalin A-binding components derived from *B. tropicalis* (Bt-ConA extract) have shown to be highly immunogenic in allergic diseases. This study aimed to evaluate the humoral and cellular immune responses to *B. tropicalis* in sensitized patients.

Methods: A total of 137 patients with allergic rhinitis with/without asthma and 109 non-atopic subjects were selected and analyzed for skin reactivity (SPT), total serum IgE and specific IgE levels to both Bt-total and Bt-ConA extracts, proliferative response and cytokine (IFN-γ and IL-5) production by peripheral blood mononuclear cells stimulated with both extracts.

Results: SPT showed that 70% of patients were sensitized to Bt (Bt+) and no patient was monosensitized to Bt. Similar levels of specific IgE to Bt-total and Bt-ConA extracts were found in Bt+patients, while higher levels of total serum IgE were found in atopic than non-atopic subjects. Significant PBMC proliferation was observed in response to Bt-total extract in Bt-sensitized, but not in Bt-non-sensitized patients and non-atopic subjects, while Bt-ConA extract was able to induce increased proliferative responses in all patient groups. Significant IFN-γ production was observed only after Bt-ConA stimulation in Bt+patients, while Bt-total extract showed no changes. IL-5 production was consistently seen in Bt+patients after allergen-specific stimulation or even with no stimulus.

Conclusion: We can conclude that Bt-ConA extract may contain relevant antigens that are involved in both humoral and cellular immune responses, with potential use in diagnostic procedures.

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Inhibitory action of epinastine hydrochloride on eosinophil survival in vitro

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Background and Purpose: Eosinophils are well accepted to be the most important cells in the development and maintenance of the clinical conditions of allergic diseases such as allergic rhinitis and asthma. It is also accepted that nasal mucosal epithelial cells secrete several types of cytokines and chemokines, which affect eosinophil accumulation into the site of the disease and cell survival. However, the influence of second-generation antihistamines, which are used for the treatment and prevention of allergic diseases are not well understood. The present study, therefore, was undertaken to evaluate the influence of epinastine hydrochloride (EP), the most famous second-generation of antihistamine in Japan, on cytokine secretion and eosinophil survival.

Materials and Methods: Epithelial cells from nasal polyps were stimulated with 25 ng/ml TNF-α with or without EP for 24 h. Cytokine levels in culture supernatants were examined by ELISA. Human peripheral blood eosinophils were cultured with epithelial cell conditioned medium (CM) and EP. Eosinophil survival was assessed by Trypan blue dye exclusion test. Results are expressed as the mean SE of cytokine concentrations (pg/ml) and eosinophil survival index (%).

Results: Addition of EP into cell cultures dose-dependently inhibited the ability of cells to produce granulocyte-macrophage colony stimulating factor (GM-CSF), vascular endothelial growth factor (VEGF) and IL-8, which are increased by TNF-α stimulation. EP also could inhibit eosinophil survival induced by CM and the minimum concentration of EP that caused significant suppression of the survival was 20.0 ng/ml.

Conclusion: The inhibitory effect of EP on inflammatory cytokine production from epithelial cells and on eosinophil survival contributed to its therapeutic effect on allergic airway diseases, including allergic rhinitis.

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Inhibitory action of epinastine hydrochrolide on the production of anti-angiogenesis factors from mouse peritoneal mast cells in vitro

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Background: H1 receptor antagonists, so called antihistamine, are accepted to have anti-angiogenesis properties in addition to being H1 antagonists. Epinastine hydrochloride (EP) is the most famous H1 receptor antagonists in Japan and used for treatment of allergic upper airway inflammatory diseases such as pollinosis with remarkably success. However, the influence of EP on pro-angiogenesis factor production is not well understood. In the present study, therefore, we investigate the influence of EP on the production of angiogenesis factors, vascular endothelial growth factor (VEGF), keratinocyte-derived chemokine (KC) from murine mast cells *in vitro*.

Materials and Methods: Murine mast cells were stimulated with ovalbumin in the presence of various doses of EP for 24 h. The levels of vascular endothelial growth factor (VEGF), keratinocyte-derived chemokine (KC) and tumor necrosis factor (TNF)-α levels in culture supernatants were examined by ELISA. mRNA expressions of these factors were also examined by semiquantitative RT-PCR in 4 h-cultured mast cells.

Results: EP could suppress the production of VEGF, KC and TNF-α induced by an IgE dependent-mechanisms in dose-dependent manner. The minimum concentration of the agent that caused significant suppression was 45ng/ml. EP also suppressed mRNA expression examined when the agent was added to cell cultures at a dose of 45 ng/ml.

Conclusion: These findings strongly suggest that anti-angiogenesis activity of EP may confer the attenuating effect of the agent on allergic diseases, including allergic pollinosis.

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Reduced levels of total serum IgE and FcRI expression in releaser and non-releaser basophils

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FcRI mediated signal pathway in basophils and mast cells leads to release of histamine and other mediators in-vitro and in-vivo systems. Interestingly, basophils from 18-20% of the population do not release histamine and other mediators on activation of the IgE signal transduction pathway and this has been attributed to the absence of tyrosine kinases Lyn and Syk. The present work deals with the histamine releasibility in Indian subjects to assess relationship between releasers and non releaser basophils with expression of FcRI and total serum IgE levels.Basophils from peripheral blood of healthy adults was purified by density gradient centrifugation and negative immuno- selection technique. Histamine release assay was performed flourometrically. Assessment of IgE receptor expression was carried out by flowcytometry and total serum IgE was estimated by ELISA method.Histamine release after ConA challenged varied from 0–100% in Indian subjects. Eighteen percent subjects showed less than 5% histamine release and were considered non-releasers while those with more than 20 % were considered as releasers. Flowcytometric analysis revealed a significantly reduced expression of FcRI in non releaser basophils ($p < 0.05$). Interestingly, total serum IgE levels were also significantly ($p < 0.05$) reduced in non-releasers suggesting a common regulator of the phenotype. An in depth evaluation could lead to identification of a potential target for the development of therapeutics for allergic patients.

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An Investigation into the interaction of IgE with truncated recombinant CD23 (FcåRII) fragments

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CD23, low-affinity receptor for IgE, has been widely implicated in the synthesis of IgE as well as in IgE-mediated immune and inflammatory functions. There are two forms of CD23 in humans-CD23a and CD23b, their cell expression and functional activities are different. Structurally, CD23 presents a single membrane-spanning domain followed by an extracellular domain that consists of three regions: the á-helical coiled coil stalk region, which mediates the formation of trimers, followed by the lectin head, which binds IgE, and at the C terminus, a short tail containing an inverse RGD (Arg-Gly-Asp) sequence.

Aims of the project is to express defined truncated fragments of CD23 and to assess the physical and biological properties of these truncated fragments with regard to: IgE independent mast cell /basophile secretayogue activity. After obtaining all the necessary information concerning the different generation of CD23 fragments as well as the vector PET-14 and pIRES-EGFP, the appropriate primers were designed. The primers are essential for PCR amplification of the desired sequences and for subsequent cloning into the PET-14 and pIRES vectors.

CD23 a and b –RGD sequence and also the whole things of CD23 a and b have been cloned into pIRES vector for transfection and subsequent evaluation of IgE in functional assays. Overlap PCR was used to produce the different truncated fragments of CD23 molecule, consisting of CD23 a and b minus the RGD sequence (adhesion part).

In the meantime, the plasmid vectors PET-14 and pIRES were introduced to XL1-E.coli cells in order to be obtained using the Mini and Midi preps protocol (gene transfection). Their DNA and protein was purified and obtained the characterisation by electrophoresis.

CD23 a and b- RGD sequence and their whole things was transfected into the J558L (mouse myeloma cells) in order to study of expression of EGFP and biological activity by FACS (Fluorescence activated cell sorting) and FACS analysis for investigating the interaction of the IgE with different expression of CD23. Binding of IgE-Fc fragments to cell surface and expressed human CD23 were assessed using flow cytometry to detect the binding of IgE to cell surface receptors, using a biotinylated anti-IgE, followed by a streptavidin phycoerythrin conjugate.

The results show that there is no expression of CD23 a- RGD sequence on J558L cells and these cells can express CD23 b-RGD sequence but it is not stable.

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Allergenic and hypoallergenic isoforms of the major birch pollen allergen Bet v 1 are differentially uptaken by DCs of allergic and healthy individuals

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Background: The major birch pollen allergen Bet v 1 is present in structurally slightly different isoforms that vary in their allergenic properties. Whereas Bet v 1a represents a potent allergen, the naturally occurring isoforms Bet v 1d and the engineered Bet v 1mut display strongly reduced allergenic activity. Although the biology of T cells in allergy is well understood, little is known about the mechanisms that control the initial T cell polarization by DCs in response to allergens.

Objective: The present study investigated the different properties of Bet v 1 isoforms to be uptaken by human monocyte-derived DCs (MoDCs) of birch pollen allergic and healthy individuals.

Methods: Eight birch pollen allergic and 7 healthy donors were included in this study. MoDCs generated from peripheral blood and taken at day 7 of differentiation were incubated with various concentrations of fluorescein isothiocyanate (FITC)-labeled recombinant Bet v 1a, Bet v 1d, and Bet v 1mut for 10 to 120 minutes at 37°C and studied by means of flow cytometry. Mechanisms of antigen internalization were investigated by the same experimental setup in the presence of inhibitors of pinocytosis, endocytosis, and receptor internalization.

Results: FITC-labeled proteins were taken up by MoDCs in a dose-and time dependent manner. The percentage of FITC positive cells increased from 1 to 5 µg/ml of labeled proteins whereas the cells internalized most proteins between 10 and 30 minutes. At the lowest concentration of 1 µg/ml, the uptake of Bet v 1a was lower compared to the uptakes of Bet v 1d and Bet v 1mut in DCs of both study groups. This fact did not change for healthy individuals when higher concentrations of proteins were used whereas in allergic individuals Bet v 1a uptake at higher concentrations was similar to that of the two other isoforms. Inhibition of the uptake was generally lower using DCs from allergic compared to those of healthy individuals.

Conclusion: All different agents showed inhibitions indicating that the uptake of Bet v 1 isoforms is not only mediated via one pathway but an interplay of mechanisms like receptor-mediated internalization, endocytic processing and macropinocytosis. Our observations implicate that different mechanisms are involved in Bet v 1a uptake in allergic individuals possibly leading to the induction of signals for Th2 polarization. This study was supported by the SFB F1802 of the Austria Science Fund and the Austrian Academy of Sciences.

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HLA-DRB1 polymorphism in atopic dermatitis with egg white allergy in korean children

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Purpose: It is known that polymorphism of the human leukocyte antigen (HLA) class II can restrict specific IgE responses. We investigated whether particular HLA-DRB1 polymorphisms contribute to the development of AD and EW sensitization in Korean children with AD.

Methods: A total of 185 patients with AD and 109 normal controls (NC) with no personal and family history of allergy were included. HLA-DRB1 typing was done using PCR-SSO (sequence specific oligonucleotide) and PCR-SSCP (single strand conformation polymorphism) methods. Phenotype frequencies of the HLA-DRB1 alleles of AD patients were compared with those of NC. AD patients with allergy to EW was defined as group A (96 patients) whose EW specific IgE was over 2 kU/L in less than 2 year old age and over 7 kU/L in greater than 2 year old age. Group B (89 patients) was defined that EW specific IgE was negative among AD patients. Phenotype frequencies of the HLA-DRB1 alleles in group A and group B were compared with those of NC. HLA-DRB1 alleles were classified into functional groups (A, De, Dr, E, Q, R, a) and frequencies of HLA-DRB1 functional groups in group A and group B were compared with those of NC.

Results: HLA-DRB1*1101 was present at significantly higher frequency in AD patients compared with NC (12.4% vs 1.8%, P=0.002, OR=7.796, CI 1.775–32.883) and was regarded as a factor associated with AD susceptibility. The result was significant after Bonferroni correction (Pc=0.048). The frequency of HLA-DRB1*0803 (10.8% vs 19.3%, P=0.043) was decreased in AD compared with NC, showing a weak protective effect against the development of AD. HLA-DRB1*0802 was decreased in group A compared with group B (2.1% vs 10.1%, P=0.021) and was regarded as a weak protective factor against the development of egg allergy in AD. HLA-DRB1*1501 was increased in group A compared with group B (22.9% vs 11.2%, P=0.036) and was regarded as a weak susceptibility factor associated with the development of egg allergy in AD. HLA-DRB1 functional group "a", in which DRB1*1501 is included, was also weakly associated with the development of egg allergy in AD. However, none of theses results remained significant after Bonferroni correction.

Conclusion: There was a significant association between HLA-DRB1*1101 and AD. Weak association between HLA-DRB1*1501 with susceptibility to and HLA-DRB1*0802 with protection against the development of EW allergy in AD were observed.

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Participation of indicators of sensitized T lymphocytes in diagnosis IgE independent and IgE dependent cow's milk allergy in children

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Background: Food allergy is an abnormal response of immunological system, especially of mucosa immunological system on antigens supplied per os. There are very complicated and still unexplained to the end immunological mechanisms, which lead to hypersensitivity reaction. Most often food hypersensitivity is identified as the effect of atopy, which is connected with humoral response (specific IgE antibody). On the contrary cell immunological response are less investigated, however they can be very important, especially as a significant factor to initiate pathological allergic processes.

Aim: The study concern the usefulness of flow cytometry to estimate specific sensitization of subpopulation of lymphocytes to food allergens in the allergy diagnosis.

Methods: The investigations were performed on 60 children since 6 month to 5 years old: 20 children with CMA IgE dependent, 20 with CMA IgE independent and 20 healthy children. IgE total, sIgE, IgG, IgA, IgM, basic immunological panel, CD 23, CD25, CD26, CD30, CD69, PCNA were measured.

Results: We noticed decrease of expression of CD4+CD30+between I and II examine (p=0,029), between I and III (p=0,009); decrease of expression of CD8+CD26+between I and III test (p=0,038); decrease of expression of CD19+CD23+between I and II examine (p=0,012) in I type of hypersensitivity. We observed decrease of expression of CD4+CD25+between I and III examine (p=0,026) and decrease of expression of CD4+CD26+between I and III examine (p=0,036) in IV type of hypersensitivity. Expression of CD69 was decreased after diet in allergy IgE dependent. Values of expression of PCNA are similar in I and IV type of hypersensitivity in children with CMA. Decrease of expression of PCNA in II examine was observed in both cases. Allergen reintroduced caused increase of expression of PCNA in both types of allergy (p=0,048 and p=0,041).

Conclusion: Our recent research confirm changes of the expression of T lymphocytes activation markers. It is connected with in vivo stimulation to allergen or with allergen elimination. The study of expression of activation markers using flow cytometry in food allergy in children can be helpful in observation of the dynamic progress process, but it cannot be used as a single diagnosis test.

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Probiotic components can induce the Th1 polarization through keratinocytes

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Background: Probiotics are defined as live non-pathogenic microorganisms that beneficially affect the host. It has been suggested that administration of probiotics may have therapeutic and/or preventive benefits by improving the Th1 responses in atopic dermatitis (AD).

Aim: We investigated the possibilities that skin can be used as a target for the prevention of AD and *Lactobacillus rhamnosus*, one of the most commonly used in the therapeutic intestinal probiotics, can induce the Th1 responses through keratinocytes.

Methods: Keratinocytes were treated by *L. rhamnosus* for 2 hours and then cultured with new media for 24 hours. The supernatant (KCM) of *L. rhamnosus*-treated keratinocytes were collected and used for the maturation of immature dendritic cells from 5 normal and 5 AD individuals. CD14+ cells and naive T cells were isolated from peripheral blood of normal and AD individuals using isolation kits. CD14+cells were differentiated to immature dendritic cells (iDCs) with GM-CSF (200 ng/ml) and IL-4 (20 ng/ml) for 6–7 day. The iDCs were activated by KCM or LPS lipopolysaccharide (LPS) (positive control) for 48 hours and cocultured with naive T cells for 7 days.

Results: We could confirm the maturation of iDCs by KCM and LPS through morphological changes and the expression of mature DC specific markers (CD80, CD83, and CD86) in normal and AD individuals. ELISA analysis showed that the mature DCs activated by KCM could induce the Th1 reponses. So, the IFN-γ was significantly increased in the co-cultured supernatants of mature DC and naive T cells, but IL-4 level was almost based. Furthemore, we found that IL-8 and HBD-3 were highly increased in keratinocytes after *L. rhamnosus* treatment.

Conclusion: Probiotics, *L. rhamnosus*, can improve the Th1 responses through keratinocytes. The effects of probiotics will be mediated by IL-8 and/

or HBD-2 secreted by keratinocytes. However, there is no difference for the effects of probiotics between normal and AD individuals.

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Interaction of aeroallergens with the respiratory interphase: degradation, processing and transmission via the epithelial barrier

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Background: Antigen presenting cells are considered as the most important immune cells giving rise to allergy or tolerance development. But APCs are already embedded in a cytokine milieu, which in part predicts the immune response. Epithelial cells form a barrier to the environment and they are activated upon contact with molecules e.g. allergens. Therefore, they might play a pivotal role in the sensitization process. We studied the fate of the major grass pollen allergen Phl p 1 and its molecular modifications after contact with the respiratory interface.

Methods: The influence of mucosal secretions (nasal secretion, bronchial lavage fluid) was investigated by immunoblotting and zymography and the uptake and transmission of the allergens by epithelial cells (cell lines and nasal and lung biopsies) were studied by FACS analysis, determination of cytokine release and immunohistology.

Results: Nasal secretions of individuals suffering from bronchial inflammations or allergy to aeroallergens showed proteolytic activity. Experiments using supernatants from mast cells or neutrophils, similar to acute inflammation, resulted in a partial cleavage of the allergens. To investigate the allergen uptake two epithelial cell lines were used mimicing different sections of the lung, A549 (derived from alveolar pneumocytes) and Calu-3 (from the upper respiratory epithelium). Both cell lines were activated by Phl p 1 as demonstrated by the release of IL-8 and IL-6. Calu-3 cells in contrast to A549 cells expressed MHCII, a prerequisite for antigen processing and presentation. A549 cells, on the other hand, take up allergens by macropinocytosis and probably perform transcytosis. First results of human lung slices incubated with Phl p 1 showed only a faint allergen uptake, while macrophages in the alveolar space showed a considerable uptake. Whether macrophages after allergen uptake enter the interstitium again and/or whether the epithelial cells transfer allergens or fragments to professional APCs in the interstitium is still under investigation.

Conclusion: The secretions of the respiratory tract cause a more or less incomplete fragmentation of allergens which may facilitate the allergen uptake by epithelial cells. Our results reveal differences in the uptake and transmission of allergens in the upper and lower airways. While the epithelial cells of the upper part probably degrade and process the allergens, cells of the lower part seem to transmit the unprocessed allergens.

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The level of IL-6 and its soluble receptor at children with atopic march

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The estimation of influence of various programs of therapy on system and local level IL-6 and SR-IL-6 was made at development atopic march of teenager patients. The research allowed to reveal, that therapy with the help of topical steroids practically did not influence SR-IL-6 level in serum of blood (22,72±0,25ng/ml), but led to decrease in IL-6 level at patients with BA (0,17±0,001 pg/ml) and combination BÀ and ÀR (3,59±0,37 pg/ml). At combination of BÀ and ÀR the simultaneous decrease of IL-6 and SR-IL-6 was established while using topical steroids. On the contrary, having clinical BÀ and AD evidence steroid therapy led to one-stage increase cytokine (6,45±0,41pg/ml), and level of receptor to it (31,76±0,15ng/ml). Thus, research of nasal lavage can be offered as nonivasive method for the control of efficiency of therapy of patients having combination BÀ and ÀR while using topical steroids. Therapy with topical steroids of patients with BÀ and with combination BÀ with ÀR and ÀR with AD provided decrease of IL-6 level and did not lead to change of SR-IL-6 level in serum of blood. Therapy with topical steroids did not lead to decrease in content of IL-6 both in serum and in nasal secret only in group of patients with combination BÀ and AD.

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Systems biology of type-I birch pollen allergy in human patients

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Previous work in type-I pollen allergies has focused on lymphocytes and immune responses. Here we begin to analyse with a systems biology top-down view the differences in nasal epithelium obtained from healthy and allergic subjects. Light and immunoelectron microscopic analysis showed that birch pollen Bet v 1 allergen bound to epithelial cell surfaces within minutes even during non-symptomatic winter seasons only in allergic, but not healthy individuals. Bet v 1 also travelled through epithelium together with lipid rafts/caveolae and reached mast cells only in allergic, but not healthy individuals within minutes. A putative viral entry protein E3 ubiquitin-protein ligase and three enzymes involved in lipid rafts/caveolae metabolism were among the few Bet v 1 binding proteins found in allergic subjects with affinity chromatography and LC-MS2. Nasal epithelial cell transcriptomics during non-symptomatic winter season identified a list of putative receptors by which Bet v 1 might be recognized. It also suggested that defence mechanisms (such as expression of histatins) could be impaired in allergic patients. Comparing nasal epithelial transcriptomics taken in winter and during symptomatic summer seasons provided hints to the cellular perturbations enabling the Bet v1 traffic through nasal epithelial cells and tissues. Thus application of discovery and hypothesis driven methodologies on human nasal epithelial tissue could provide new hypothesis worth further analysis of the underlying molecular mechanisms.

The most significant aspect of this and other top-down explorative studies using whole genome or other large-scale analysis is that they can provide truly new hypothesis. Before this work few would have argued that the nasal epithelium is so markedly different in healthy compared to allergic subjects already during non-symptomatic winter session. Now based on these validated results the mechanisms of Bet v 1 pollen allergen binding to and traffic through the epithelium can be further explored and analysed.

Furthermore it must be noted that no single data domain, whether is would haven microscopy, proteomics, transcriptomics etc. would have alone pointed out the above-described phenomenon. Thus systems level understanding of complex pathophysiological phenomena will need all possible wet

lab techniques combined with computer sciences as the biology will be transformed into computer-readable format.

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Maternal tolerance achieved before pregnancy is transferred to the offspring and prevents asthma development in the next generation

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In this study we hypothesized that immunologic tolerance acquired before conception can be transferred from the dam to the pup. In a murine model, we induced tolerance before conception by oral application of antigen. We than immunized the offspring of tolerized dams with the same antigen. While the offspring of naïve dams developed an asthma-like phenotype with airway hyperreactivity, inflammation, Th2-cytokine production after immunization, the offspring of tolerized dams was protected, even when immunized as late as 8 months after birth. Critically involved in tolerance transfer is allergen-specific IgG that was increased during pregnancy in the tolerized mouse, fetus and newborn. $FcRn^{-/-}$mice, that cannot transport IgG via the placenta, transferred tolerance to the offspring only when the missing diaplacental IgG transfer was compensated by IgG transfer via breast milk from tolerant dams but not when the offspring was crossfostered by naïve mice. Inhibition of IFN-γ, produced by memory B cells in the offspring, abrogated the protective effect of maternal tolerance demonstrating its crucial memory role in materno-fetal tolerance transfer. Our data show that maternal immunologic memory has significant and persistent impact on the immune response of the offspring indicating that e.g. allergy prevention strategies might be effective for more than one generation.

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Meditation as a suggestion for allergies and autoimmune diseases

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Objective: The aim of the present study was to analyze the effects of a Meditation program on Complete Blood Cell count(CBC), IgG and C3 as a marker of specific and nonspecific humoral immunity.

Methods: 78 subjects, aged 18–56 years, of whom 62 were male and 20 female undergoing a period of one month Meditation, were excluded. They were all experiencing meditation for the first time. Before starting the meditation training, all of these volunteers have been visited by a physician and only healthy subjects(78 subjects), not taking any type of drug and with regular life habits were chosen to be at this clinical trial. Blood samples were taken from all subjects, the day before the study commenced, and again one month later, at the end of the study. The blood parameters investigated included the number of Leukocytes(total Leukocytes, Monocytes, Neutrophils, Eosinophils, Basophils, Lymphocytes),Red cell count(RBC), Hemoglobin(Hb), Hematocrit(Hct), Mean cell volume (MCV), Mean cell hemoglobin (MCH), Mean cell hemoglobin concentration(MCHC), Red cell distribution width (RDW),Platelet count, as well as the concentrations of immunoglobulins(IgG) and complement(C3). 4 of these volunteers has been excluded from research plan because of their sickness during the period.

Results: Statistically significant differences were found between first and second blood samples of volunteers showing lower numbers of total Leukocytes(P=0.02) and Eosinophils(P=0.01),number and percentage of Monocytes(P=0.01),as well as complement C3 concentration(P=0.004).

Conclusion: These findings demonstrate that one month of practicing Meditation, can decrease some immunological parameters. According to these results Meditation can be suggested for Hypersensitivities(Allergies and Autoimmune Diseases) although further studies seem to be needed.

Key words: Meditation, Complete Blood Cell count(CBC), IgG, C3.

ASTHMA TREATMENT

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A randomized, double-blind, placebo controlled trial on the effect of zinc supplementation on bronchial asthma as measured by sputum eosinophil count and asthma control test (ACT) in children

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Backround: Zinc being a major dietary anti-oxidant has a protective role in the airway epithelium. It may therefore have important implications for asthma where the physical barrier is vulnerable and compromised.

Objective: We investigated the effects of zinc supplementation on bronchial asthma as measured by sputum eosinophil levels and asthma control test (ACT) in children.

Methodology: There were sixty-six asthmatic subjects age range 7 to 18 years old were randomized to receive zinc 20mg/day(n=29) or placebo (n=37). Sputum eosinophil count were checked before supplementation and 12 weeks later. Likewise asthma control test (ACT) scores were obtained before supplementations and at 4 weeks, 8 weeks and 12 weeks thereafter.

Results: After 12 weeks of supplementation, the sputum eosinophil count decreased in both groups but the zinc group has the more significant change in eosinophils as compared to placebo at p=0.029 and p=0.059 respectively. However, ACT score from week 1 to week 12 showed no difference between the zinc and placebo group. (p=0.069).

Conclusion: A dietary zinc supplementation significantly reduced sputum eosinophil count as compared to placebo. However, the decline in sputum eosinophil count was not associated with improvement in asthma control. Zinc should be considered in decreasing airway inflammation but not asthma control.

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Documentation of the relative efficacy and safety profile of the salbutamol delivery by slow infusion(SI) VS inhalation(IN) to patients with acute severe bronchial asthma

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Purpose: Patients with acute severe bronchial asthma are in a moribund position. The art of devising an effective delivery system which may be followed by an optimal relief in the degree of bronchospam is the goal of resolving the imminent urgency of an acute severe bronchial Asthma.

Methods: Patient's both sex age16–50 years, with history of acute severe bronchial asthma have been included in the study. They had been Group A, medicated with Prednisolon 15–20 mg immediately followed by 10 mg 6hourly for 2 days along with Salbutamol 4mcg/kg body weight in slow infusion. Group B, having being medicated with Prednisolon as in Group A, along with inhalational delivery of Salbutamol in dilution of 1.5ml+2.5ml and 1.25ml+2ml (for adult and childhood bronchial asthma respectively). Therapeutic response as observed had been as under.

Results: Group B, had been followed with optimal bronchodilation in about 5–10 minute while with group A, it was 15–20 minutes.

Conclusion: Group B had therapeutic advantages vs A (e.g.) optimal bronchodilation in a relatively shorter period, no hazard of parental administration and a simple technique of delivery that can be managed even by any one.

Clinical Implication: Both techniques of delivery of Salbutamol are effective with in the limits of availability of services of healthy care providers and appropriate selection of cases.

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Role of body weight reduction by diet control and physical exercise in controlling asthmatics (children and adults) with obesity

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The prevalence of asthma has increased worldwide since the 1960s. The incidence of asthma is high among children; however, a relative high annual incidence is also estimated in adults. Studies for risk factors have mainly analyzed cross-sectional design data based on prevalent cases of asthma. Such studies may determine either cause, consequence or both. Incidence studies have mostly focused on occupational asthma or smoking habits. Since the late 1990s, obesity has been reported to be associated with asthma, and an increase in the prevalence of obesity has been reported along with a parallel increase in asthma prevalence. However, a convincing relationship between asthma and obesity has not been established. Accordingly, this study was carried out aiming at demonstrating the effect of weight reduction on clinical, functional and serological parameters in obese asthmatics. In this study, obese asthmatic patients showed marked improvement clinically, functionally, with marked decrease in number of exacerbations and medications used.

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Feasibility and applicability of secondary prevention of asthma in allergy practice

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Background: The possibility for secondary prevention of asthma was highlighted by the prevention of asthma study, which demonstrated that immunotherapy in patients with allergic rhinitis, could prevent asthma in some patients. The feasibility of secondary prevention of asthma and the proportion of patients likely to be benefited need to be demonstrated as immunotherapy requires time to exert its beneficial effects.

Methodology: All the patients who underwent a detailed evaluation including allergy testing during the period 2003 to 2006 were included in the study. A structured questionnaire was applied to collect data on demography, a detailed clinical history, the duration of asthma and allergic rhinitis and the proportion of patients whose rhinitis preceded asthma and the time interval between the development of rhinitis and asthma.

Results: A total of 934 cases were included in the study. 701 (75%) patients had allergic rhinitis for varying intervals before they developed asthma. 143(15.5%) patients continued to have rhinitis for more than 5 years without developing asthma. 57(6%) patients had only asthma without rhinitis. 33(3.5%) patients developed asthma before they developed rhinitis. 327 (46.6%) developed asthma within 1 year of developing rhinitis. Of these, 164 developed asthma within 6 months and 169 between 6–12 months. 374 patients developed asthma more than a year after the onset of rhinitis and the frequency gradually decreased with increase in duration of rhinitis.

Conclusion: Majority of patients had rhinitis before they developed asthma. Nearly 50% of these patients with preceding rhinitis developed asthma within one year, who may not be amenable for secondary prevention with immunotherapy due to the delays in referral to the allergist and the time required for obtaining the necessary benefit from immunotherapy. Patients who develop asthma alone or develop rhinitis after the onset of asthma are not candidates for secondary prevention. Of the total cases studied, less than 40% can undergo interventions targeted towards secondary prevention of asthma. Intense education of the Family physicians and ENT specialists is needed to ensure prompt referral to the allergist to help these patients.

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Asthma control assessd by the asthma control test and markers of airway inflammation

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Aim: to assess the relationship between ACT and different markers of airway inflammation in asthmatic subjects

Methods: In a crossover design, we studied 106 patient suffering from mild or moderate asthma (median age 31years IQR 16–45)). Fifty five percent were only treated with short beta agonist on demand. On the same visit patients fullfiled the ACT questionnaire, exhaled nitric oxide (eNO) (NiOx Mini) was measured and bronchial challenge with hypertonic saline and sputum induction were performed.

Results: On the whole sample ACT was modestly correlated with eNO ($r=-0,40$, $p< 0,001$), eosinophils in induced sputum ($r=-0,30$, $p< 0,05$) and the slope of hypertonic saline bronchial challenge ($r= 0,43$, $p< 0,001$). Patients with very good control (ACT 24 0r 25) had significantly lower levels of eNO (41 vs 72 ppb, $p<0,001$) and eosinophils (10 vs 6%, $p< 0,05$). Correlations of all these variables improved, when only the 55 patients treated with beta agonist on demand were included: eNO ($r=-0,55$, $p< 0,0001$), eosinophils in induced sputum, ($r=-0,50$, $p= 0,002$), and slope of hypertonic saline bronchial challenge ($r= 0,52$, $p< 0,0001$. ACT was not correlated to FEV1 of FEV1/FVC.

Conclusion: asthma control assessed by the Asthma Control Test was correlated to direct and indirect markers of airway inflammation. ACT and pulmonary function variables were not correlated.

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Salbutamol metered-dose inhalation is as effective as nebulisation in managing acute asthma exacerbations in hospitalized children in accordance with an asthma pathway

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Background: Acute asthma exacerbations have traditionally been managed with nebulised bronchodilators in the emergency rooms as well as for inpatients. In 2003, Singapore was hit by SARS and the fear that the use of nebulisation could cause the spread of the virus prompted the development of an asthma pathway using MDI.

Aim: To compare the length of stay in children admitted for asthma exacerbations before and after the introduction of an asthma pathway for managing acute exacerbations with salbutamol metered-dose inhalers(MDI) instead of nebulisation.
Methods: Children presenting to the Children's Emergency and admitted to the wards for asthma exacerbations were managed according to an asthma pathway.
Results: There was no increase in admissions from the emergency department or to high dependency or to the intensive care after implementation of the asthma pathway with the use of Salbutamol MDI instead of nebulisation. The average length of stay decreased from a high of 2.78 days pre-implementation of pathway to a low of 2.19 days post-implementation. The average cost per patient decreased from $1,136.85 in 2001 to between $974.00 –$1,011.05 post pathway implementation.
Conclusion: The asthma pathway has shown that the use of salbutamol MDI is as effective as nebulisers in the management of acute asthma exacerbations and has led to a decrease in the average length of hospitalisation as well as decreased cost per patient.

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A troublesome diagnosis of Churg Strauss Syndrome

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Case Report: A 41- year-old asiatic man was admitted to the emergency room of our hospital with fever, wheezing, dyspnea, arthralgias.

His past history was remarkable for allergis rhinitis and recurrent episodes of asthma since his arrival in Italy at the age of 34.

The patient was treated with a course of systemic corticosteroids (80mg daily prednisolone), theophylline, inhaled budesonide/ formoterol 160/ 4.5 mcg (twice a day), montelukast 10 mg . His chest radiograph and full blood count including eosinophil count were normal. His spirometric test showed FEV1 1.57 (42%), FVC 2.52 (57%), PEF 240 l/min.

One month later he was seen in the outpatients department and noted to be generally unwell, very wheezy and 2 days before the visit he has noticed the comparison of hemoptysis.

His chest radiograph and full blood count, including eosinophil count, were normal. Antinuclear antibody and antineuthrophil cytoplasmic antibody (ANCA) were both negative. Sputum culture and urine microscopy were unremarkable. Aspergillus precipitins and radioallergosorbent test (RAST) against Aspergillus were negative. Cultures for mycobacteria were negative. The electrocardiogram and echocardiogram were both normal.

Computed tomography of the paranasal sinuses dimostrated a near-total opacity with the material of the soft parts of the normal sinus and right sphenoidal sinus, as well as of ethmoidal cell, the right maxillary sinus and nasal cavities. Thorax computed tomografy revealed generalized enlargement of the mediastinic lymph nodes.

Treatment with montelukast was stopped and high dose oral prednisolone was commenced.

The patient elected to discontinue his inhaled treatment .Three months later his symptoms of asthma returned. Chest- X-ray showed patchy bilateral infiltrates in the lungs. He had marked hypereosinophilia (15%), elevated serum IgE levels (465 IU/ml), and antineuthrophil cytoplasmic antibody (ANCA) were positive. His spirometric test showed FEV1 1.60 (45 %), FVC 2.35 (55%), PEF 240 l/min. He required a further short course of oral coricosteroids.

His symptoms remained troublesome and his treatment was changed to cyclosporine 3 mg/ kg and his condition showed marked clinical as well as radiological improvement.
Discussion: We must carefully diagnose and treat patients with middle-age onset of severe asthma, because the symptom may be a lung manifestation of CSS, in which various organs are involved as a result of systemic necrotizing vasculitis.

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Educational courses improve asthma management and treatment

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Background: Education of the patients is one of the first goals of asthma management in all the international treatment protocols. We made this study in order to estimate the need and the effect of an educational program prepared by the Macedonian Asthma Center.
Methods: We studied 300 asthmatics. We prepared a pilot educational course, covering the most important and "hot" subjects concerning asthma, following the guidelines from the Global strategy for asthma management and prevention. (May 1996), and a questionnaire with 10 questions treating the problems concerning the treatment problems in asthma management. The questions were grouped in 2 parts : I - general knowledge of asthma medications and the necessity of adequate and continuous anti-inflammatory treatment; and II - accent on the emergency interventions and actions during acute exacerbations. The questionnaire was completed by all of the patients, before and after attending the educational course.
Results: Before visiting the course, only 46.3% answered correctly the questionnaire (39.9% correct answers on part I and 52.8% on part II). These results imply that more than 50%of the tested asthmatic population do not realize the need for preventive medication or would in fact take the wrong medication during worsening of their disease. After visiting the educational course the results are as follows: 54.8% overall correct answers (nonsignificant-NS). The results from part I showed that only 52.4% (NS) of the patients gave correct answers and 73.6% (significant) answered the II part correctly.
Conclusion: We conclude that patients more willingly accept advice on emergency self management of acute asthma exacerbations, but it is the most difficult to overcome the "fear of preventive treatment" and to change the dogmatic opinion on the therapeutic issues of asthma management. The study shows the necessity of organizing further and continuous asthma educational programs on all levels of the health care.

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Clinical evaluation of severe asthma attack requiring tracheal intubation and respirator management

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Background: Recently, the number of patients requiring hospitalization because of an asthma attack has decreased; however, there are still patients who require hospitalization for tracheal intubation and respirator management for a severe asthma attack. Therefore, we evaluated the background features of 20 asthmatic patients who required tracheal intubation and respirator management in our hospital.
Methods: We evaluated 20 asthmatic patients who visited our hospital from January 2001 to December 2005 and required tracheal intubation and respirator management. All the patients had severely exacerbated asthma, as determined on the basis of the guideline of the Global Initiative for Asthma (GINA) 2006, and they required tracheal intubation and respirator management. We evaluated their history of smoking, the days from asthma attack

onset to their visit to our hospital, the level of asthma control as determined from the GINA 2006 guideline, treatments taken before the patients visited our hospital, their frequency of visiting a hospital, the reason for tracheal intubation and respirator management based on the Asthma Prevention and Management Japanese Guideline 2006, and their prognosis.
Results: The patients who required tracheal intubation and respirator management were the following: 1. smokers, 2. patients not taking or irregularly taking treatment, 3. patients who used inhaled short-acting β2 agonist only at the time of an asthma attack, and 4. patients not using inhaled glucocorticosteroids. The reasons for tracheal intubation and respirator management were as follows: PaO2 of less than 50mmHg despite maximum oxygen administration in 30% of the patients; sudden increase in PaCO2 leading to unconsciousness in 20%; both PaO2 of less than 50mmHg despite maximum oxygen administration and sudden increase in PaCO2 leading to unconsciousness in 25%; and severe ventilatory or cardiorespiratory disturbance in 25%.
Conclusion: We observed that a thorough education of patients and treatment mainly using inhaled glucocorticosteroids are important for preventing a severe asthma attack that requires tracheal intubation and respirator management.

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Assessment of quality of life in caregivers of asthmatic children

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Background: Interest in the impact of illness on day to day function is leading investigators to include both disease specific and generic health related quality of life (HR QOL) questionnaires in a broad range of clinical studies and to gain a full picture of the impact of asthma on the lives of caregivers of asthmatic children, it is necessary to make direct measurement of health related quality of life.
Methods: In response to this need, we used Juniper's Pediatric Asthma Caregiver's Quality of Life Questionnaire (PACQLQ) that has been developed based on guidelines for construction of over a dozen validated disease specific quality of life instruments.The PACQLQ that contains 13 items in two domains of emotional and activities disturbances. The study design consisted of an 18 month single cohort study. Patients participating in the study were 113 children, 7–17 years of age, with a wide range of asthma severity and their caregivers. For each caregiver a PACQLQ was completed. One week before visit patients recorded morning peak flow rates, medication use and symptoms in a diary. After complete physical examination, for determining of asthma severity, spirometry was performed.
Results: The questionnaires after statically analysis showed good levels of both longitudinal and cross sectional correlations with the conventional asthma indices and with general quality of life.
Conclusion: We found a good relevancy between severity of asthma and QOL scores and more disturbances of QOL in caregivers of male asthmatic patients than caregivers of female asthmatic patients. Bronchial asthma as a chronic and devastating disease can affect not only life style of the patients but also their caregivers. Increment of our knowledge about these disturbances can help the physicians for better understanding of burdens of their patients.

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Unified treatment in school children with allergic rhinitis and asthma

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Aim: To verify the efficacy of beclomethasone dipropionate (BDP), administered through nasal inhalation, in the simultaneous treatment of asthma and allergic rhinitis (AR).
Methods: 30 patients with asthma and AR, aged 7–17 years were randomly allocated. During eight weeks, 20 of them received BDP-CFC (at least 500 mcg/day) inhaled exclusively by the nose (mouth closed) using a large-volume (650 ml) spacer attached to a facemask. Control group (10 subjects) received conventional treatment, i.e., BDP by dual administration (aqueous intranasal and oral inhalation through the mouthpiece of the same spacer device). Every two weeks a clinical score for AR and peak expiratory flow (PEF) was assessed by independent observers. Spirometry was performed at admission and at the end of the follow up. A minimum decrease in 66% in clinical scoring was considered as therapeutic success.
Results: Therapeutic success rate was of 50.0% for the experimental group and of 70.0% for the control group ($p=0.11$). PEF and FEV1weren't statistically different in the two groups neither at admission nor by the end of the treatment ($p > 0.10$).
Conclusion: Results suggest that this alternative treatment could be recommended for the simultaneous treatment of AR and asthma, specially in developing world. Other advantages are higher compliance, lower costs and fewer side effects.

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Influence of topical intranasal steroids on improvement of ventilating capability of lungs in patients with bronchial asthma

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Background: Treatment of a bronchial asthma (BA) is often conducted without an allowance of a concomitant allergic rhinitis (AR), which can be a reason of unsuccessful results in an achievement of a maximized therapeutic effect. Research about an interaction between BA and AR is conducted in the republican allergic center of scientific research institute of cardiology and internal diseases under Ministry of Public Health of the Republic of Kazakhstan.
Aim: To determine the degree of influence of allergic inflammation of the upper respiratory tracts on ventilating capability of lungs in patients with bronchial asthma.
Materials and Methods: The thirty patients with severe BA and concomitant the AR were prescribed to intranasal mometazon spray in a dose of 100 microgram two times a day for a one month. This treatment followed after two weeks course of standard hospital therapy that includes steroids (intravenous, inhalation), b2-àgonists, etc. The age of the patients was ranged from 25 to 50 years old. Therapy effectiveness was controlled both by clinical and by laboratorial methods: spirography and daily pickflowmetry, determination of the resistance of a bronchial tree, rhinomanometry and identification of ECP.
Results: All patients at the end of the first month of treatment mentioned the stable improvement of nasal breathing and sleep recovery. Also, a valid reduction of the resistance of nasal apertures was discovered ($p<0.005$). The majority of the patients (93.3%) have the improvement of clinical picture of the BA, which confined in less amount of asthma attacks and also improvement of the functional and laboratorial measures: PEF growth, decrease of its variability ($p<0.05$), reduction in the resistance of the bronhial tree ($p<0.05$) and normalization of ECP.
Conclusion: Conducted research proved a valid increase in the effectiveness of supporting therapy of the BA with the combination of active treatment of the AR, which becomes apparent besides clinical improvement in an additional growth of measures of ventilating capability of lungs, level of the resistance of the bronchial tree and nasal apertures and normalization of ECP content.

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An all Wales audit of secondary care of acute asthma during February 2006

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We audited the secondary care offered to acute asthmatic patients admitted to all 17 hospitals in Wales, UK during February 2006. The audit analysed referral source, care delivered by the ambulance service, emergency units, medical admission units and inpatient units in all cases of asthma aged 18 years or greater. All data was extracted by qualified medical personnel. The results show that 248 patients were admitted during that month (36 patients were excluded as not having asthma). 44 were acute severe/severe, 91 moderate, 75 mild and in 38 severity was not documented. Ambulance transfer was used in 99 cases, 63 patients calling the ambulance directly. Overall, ambulance staff measured vital signs well but peak flow (PFR) was measured only 8 cases. Most patients received nebulised bronchodilators/high flow oxygen in the transfer process but steroids were used rarely and observations were repeated in only 27 cases. Emergency units were the primary site of acute care. PFR was measured in only 67% of cases compared to 90% having oxygen saturations measured. Only 162 of 248 cases had a record of steroid administration and only 67% were reassessed. Of those admitted (99) only 51% were under specialist care and only 32% were treated in a specialist respiratory ward. Overall discharge planning both from emergency and inpatient units was disappointing with only 13% of inpatients having written management plans and only 4.5% of patients discharged directly from emergency units offered further review.

Secondary care of acute asthma in Wales does not confirm to national guidelines for acceptable levels of care.

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A survey in physicians in China on monitoring and treatment of childhood asthma

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Objective: To investigate the approach (monitoring and treatment) of pediatricians and general physicians to childhood asthma in China.

Methods: A standardized questionnaire survey, which was construced by APAPARI on monitoring and treating childhood asthma was send to physicians in 25 provinces and cities of China.

Results: 1863 questionnaires were received, of which 912 were completed (48.95%) 57% of the doctors were general pediatricians, 20% were pediatric pulmonologists or pediatric allergists, 9% were adult pulmonologists or allergists who also treatment children, and 14% were others (non-specified). Working places were: 71% in a tertiary hospital, 23% in a secondary hospital, and 4% work in a first grade hospital.Most of the doctors (66%) used peak flow meter or spirometry to monitor childhood asthma, while 31% of them used diary cards. For treating acute asthma exacerbations, the first choice treatment was nebulized salbutamol/terbutaline every 20 mins (29%), the second choice was salbutamol inhalation with a spacer (22%) and the third choice was systemic corticosteroids (12%). Pediatric pulmonologists and pulmonologists tended to use ICS for acute asthma. The most frequently used systemic corticosteroids for acute asthma treatment were dexamethasone (86%), methylprednisolone (83%) and hydrocortisone (81%). The criteria applied by doctors to start a maintenance therapy in asthmatic children were the frequency of symptoms (36%), severity of symptoms (32%) and whether or not there was a severe disturbance in lung function testing (27%). For the selection of drugs, doctors tended to choose ICS. Generally, the first choice of drugs used in maintenance treatment of asthma in young children (0–3y) were ICS (85%), the second was montelukast, and the third was ICS+ oral LABA(slow release sulbutamol) (67%), while in preschool and school children (4–16y) the first choice was a LABA + ICS (84–87%), the second was ICS (81–83%) and the third montelukast (73%). For moderate to severe asthma , the first choice was ICS + LABA.

Conclusion: The awareness and recognition of childhood asthma, the use of peak flow meter, the implementation of GINA guidelines and the use of ICS has improved among the doctors of the cities in China. Nevertheless, dissemination of the GINA program among non-specialists and doctors in countries and communities must be continued and re-enforced.

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Impact of patient education on knowledge, attitude, practice and self-efficacy in patients with asthma in India

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Background: Patient related factors like poor knowledge, individual beliefs and attitudes of patients influence control of asthma leading to nonadherence. This study was conducted to develop a validated KAPSE questionnaire for the Indian population and to assess the knowledge, attitude, practices and self-efficacy (KAPSE) of patients in Mysore regarding asthma and the impact of patient education on KAPSE in these patients.

Methodology: A total of 225 adult patients participated in the KAPSE study and were part of the study that compared the efficacy and quality of life in patients receiving beclomethasone, budesonide or fluticasone. A questionnaire with 22 items was selected from Knowledge, Attitude and Self- Efficacy (KASE), a validated instrument for asthma and suitably modified to suit the cultural and educational level of the population. Two medical experts performed content validation and only those items with content validation index of above 0.75 were retained. Cognitive debriefing in 5 patients confirmed the suitability and acceptability of the questionnaire. Patient counseling individually and in groups and information leaflets about the pathology of asthma, drugs and delivery systems were conducted at every visit on 7 occasions, until 6 months. KAPSE was assessed at baseline before education and at the end of the study.

Results: Patient education significantly improved most of the questions assessing Knowledge, Attitude, Practice and Self-efficacy compared to the baseline ($p<0.05$). Subgroup analysis revealed that KAPSE at baseline were not influenced by Age, Gender and Duration of disease ($p>0.05$), but was related to educational levels and area of residence ($p<0.05$). Following patient education KAPSE of all the patients improved significantly ($p <0.05$) and there was no difference between patients of different areas of residence.

Conclusion: This study demonstrated that regular patient education significantly improves different aspects of Knowledge, Attitude Practice and Self-efficacy of patients regardless of their age, gender, education or place of residence, which could positively influence patient medication adherence.

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Asthma - a curable disease

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Background: In normal individuals, the Serum Histamine Binding Capacity (SHBC) is 20 to 30% whereas it is only 0 to 5% in allergic patients. The SHBC can be raised with Histaglobulin (HG) if it is given after preparing the patient properly and drugs like mast cell stabilisers, bronchodilators, antihistamines and anti-inflammatory drugs are given along with HG therapy. In allergic patients serum IgE level is high which comes down after HG therapy and medicines. Patient becomes symptom free and cured.

Methods: It was an observational study for 14 years. 161 patients were registered at different periods from out-patients consisting of all age-group, both sexes and from urban and rural areas. Immunoglobulin E (IgE) levels were estimated and other routine tests were done before starting treatment.

Patients were prepared before HG therapy; HG was given in two phases: Phase I – 6 primary doses, Phase II –3 booster doses. Along with HG therapy, drugs like mast-cell stabilisers, and H1 receptor-specific Histamine antagonists (Loratadine, Rupatadine, Cromolyn etc.) bronchodilators (Theophylline), antihistamines (Cetirizine, Levocetirizine) and anti-inflammatory drugs (Monteleukast) were given. 2 to 3 months after 3rd booster dose IgE levels were estimated for 61 patients. Patients were examined periodically to assess clinical improvement.

Results: Out of 161 patients, 152 patients (94.6%) are free from asthma and allergic rhinitis symptoms. Among 152, 96 patients are free for more than 4 years. 9 patients did not respond satisfactorily. Out of 61 patients, 55 patients (90.2%, $p<0.0001$) showed reduction in IgE level and are clinically free from asthma. 6 patients showed raised IgE level and correspondingly no clinical improvement. It is also found out that allergic manifestations are lowest up to 2 years of age; between 2–5 yrs, it increased and more males are affected; the occurrence rate came down in the age group of 6–15 yrs in both sexes. Above 15yrs, again occurrence rate raised and more females were affected.

Conclusion: Thus in our study comprising of 161 patients having the complaints of allergic asthma with or without allergic rhinitis, the HG therapy given after preparing the patient and continuing the above-mentioned drugs, during HG therapy was found to be effective in curing the allergic asthma. HG is effective in raising the SHBC thereby improving the immunity of the patient and also in reducing the serum IgE level.

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Flavonoids and related compounds as anti-allergic substances

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The prevalence of allergic diseases has increased all over the world during the last two decades. Dietary change is considered to be one of environmental factors that cause this increase and worsen allergic symptoms. If this is the case, an appropriate intake of foods or beverages with anti-allergic activities is expected to prevent the onset of allergic diseases and ameliorate allergic symptoms. Flavonoids, ubiquitously present in vegetables, fruits or teas possess anti-allergic activities. Flavonoids inhibit histamine release, synthesis of IL-4 and IL-13 and CD40 ligand expression by basophils. Analyses of structure-activity relationships of 45 flavones, flavonols and their related compounds showed that luteolin, ayanin, apigenin and fisetin were the strongest inhibitors of IL-4 production with an IC50 value of 2-5 mM and determined a fundamental structure for the inhibitory activity. The inhibitory activity of flavonoids on IL-4 and CD40 ligand expression was possibly mediated through their inhibitory action on activation of nuclear factor of activated T cells and AP-1. Administration of flavonoids into atopic dermatitis-prone mice showed a preventative and ameliorative effect. Recent epidemiological studies reported that a low incidence of asthma was significantly observed in a population with a high intake of flavonoids. Thus, this evidence will be helpful for the development of low molecular compounds for allergic diseases and it is expected that a dietary menu including an appropriate intake of flavonoids may provide a form of complementary and alternative medicine and a preventative strategy for allergic diseases. Clinical studies to verify these points are now in progress and we will present the result in the congress.

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Sublingual progesterone dilutions as bronchodilator in asthmatic females

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Background: Asthma is associated with hormones. Increases in asthma symptoms have been associated with menstrual variances. Pre-menstrual asthma has been noted in the literature and studies from as early as 1921 have suggested an allergic reaction to hormones. Bronchial asthma in female patients could be due to allergy to self-hormones particularly progesterone. Interestingly, progesterone is also a potent respiratory stimulant. The present study was aimed to evaluate efficacy of sublingual progesterone dilutions as bronchodilator.

Methods: This study was approved by the ethics committee of Austin, Texas. Sixteen females who had a previous diagnosis of severe asthma and who were nebulization dependent were selected for the study. After obtaining written informed consent from each patient spirometric studies were performed on these subjects Using sublingual progesterone as a bronchodilator. We analyzed changes over time of the forced expiratory volume in one second (FEV_1), the forced vital capacity (FVC), and the peak expiratory flow (PEF). For each patient we measured lung function three times: (1) before treatment, (2) after sublingual normal saline treatment (3) after sublingual progesterone treatment.

Results: After treatment with sublingual progesterone, twelve of the sixteen patients (75%) experienced a significant bronchodilator effect (greater than 12% increase) in either FEV_1 or FVC. Eight (50%) experienced significant increase in both FEV_1 and FVC. Eight (50%), had an increase of 27% or greater in PEF.

Conclusion: Progesterone dilutions are candidate drugs for the treatment of bronchial asthma. The possibility that dilutions of progesterone can act as a progesterone antagonist and that bronchial smooth muscle may be sensitive to such dilutions of progesterone due to activation of the L-arginine-nitric oxide (NO) pathway is interesting because of the close relationships between the immune and neuroendocrine systems. The study might have important implications for the development of a novel safe non-invasive treatment strategy against bronchial asthma due to hormone allergy.

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A randomized, double-blind, placebo controlled trial on the effect of zinc supplementation on bronchial asthma as measured by sputum eosinophil levels and asthma control test (ACT) in children ages 12–18 y/o

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Zinc may have its role in bronchial asthma by regulating airway inflammation and subsequently preventing asthma exacerbation.

Objective: To determine the effects of zinc supplementation on bronchial asthma as measured by sputum eosinophil levels and asthma control test (ACT) in children ages 12–18 y/o.

Methods: After consent was obtained, 30 children at a local community ages 12–18 years old diagnosed with intermittent, mild-moderate persistent asthma were randomly allocated to receive zinc supplementation 20 mg/day or placebo for thirty days. A change in the sputum eosinophil level and ACT score was obtained before and after intervention.

Results: Fifteen subjects were included in the zinc group and 15 subjects in the placebo group. There were no differences in the demographic profile between the two groups in terms of age, severity of asthma, atopy, use of maintenance medications and infections during the study period. Intragroup statistics showed a statistical significance in the zinc group for sputum eosinophil ($p=0.045$) and ACT score ($p=0.004$) while the placebo group showed no statistical significance for sputum eosinophil ($p=0.954$) but was statistically significant for ACT score ($p=0.045$). The mean difference in sputum eosinophils between zinc and placebo showed no significant difference between the two groups. ($p=0.270$). The mean change in the ACT score of zinc and placebo group showed a significant difference between the 2 groups ($p=0.067$).

Conclusion: Supplementation with zinc sulfate improves bronchial asthma in terms of ACT score but not in sputum eosinophil count in children ages 12–18 years old.

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The application of the small doses of the human recombinant alpha-2 interferon oromucosal for the patients with seasonal atopy (bronchial asthma and allergic rhinoconjunctivitis)

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Purpose: Of the research is to determine an influence of the small dose of the human recombinant alpha-2 interferon (HRá2-IFN) on the symptoms of allergy, on the level of ECP of the patients with seasonal atopic allergy.

Materials and Methods: 20 patients with seasonal atopic allergy (bronchial asthma and allergic rhinoconjunctivitis, sensitizing to the allergens of the pollen of trees). All of these patients had been taking for 30 days oromucosal HRá2-IFN in the dose of 2000 ME everyday till the begining of the season of the blossoming of trees. The group of control - 11 patients.

All of the patients filled out an individual form where fundamental clinical symptoms of pollinosis.

The immune–enzyme method in commercial test system Immuno CAP 100, Pharmacy, Sweden was used to determine ECP.

Results: The analisis of the journals of the patients showed that in comparison with three previous years the intensity of the stuffiness in nose, rhinorrea owing to the preventive therapy HRá2-IFN were authentically lower in comparison with the group of control and were 0.85 ± 0.75 and 1.00 ± 0.97; in the group of control −1.64 ± 1.03 and 1.75 ± 0.79. The necessity in B2-agonists was authentically two times lower in the analyzing group of patients than in the group of control so that it was in the analyzing group 3.25 ± 4.12 days while in the control group it was 7.82 ± 7.78 days.

The level of ECP before the medical treatment was higher in the analyzing group 252.89 ± 141.15 mkg/l (the norm is till 11.5 mkg/l), than in the control group 61.36 ± 55.47 mkg/l. After the medical treatment the level of ECP authentically became lower in the analyzing group 100.57 ± 124.68 mkg/l, however after the season of the blossoming of trees the unauthentic increase of it was registrated till 161.16 ± 143.32 mkg/l. The level of ECP in the control group unauthentically increased after the medical treatment till 84.81 ± 111.60 mkg/l, however the level of ECP in this group authentically increased after the season of the blossoming of trees till 151.02 ± 183.98 mkg/l.

Conclusion: we can assert that the prevention usage of small doses of HRá2-IFN guarantees positive clinical effect on the pollinosis (bronchial asthma and allergic rhinoconjunctivitis), provides for the lowering of the levels of ECP and therefore the decrease of inflammation process in the windpipe. So it can be recommended as the modern medicine for the prevention theraphy for the patients with the seasonal atopy.

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Polyprenol as a possible supplement for treatment of steroid resistance in patients asthma

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Background: Plant Polyprenol (PP) is approved as a substitute of Dolichyl Phosphate (DP) which decreases P-glycoprotein, enhances IL-10 synthesis and alpha GP isoforms expression in vitro. In a proof of our previous study, the effect of oral administration of PP was investigated in steroid resistant asthma (SRA) patients.

Methods: The samples obtained from 58 patients with asthma: 27 patients with SRA, 31 patients with steroid sensitive astma SSA and 20 donors. The patient's forced expiratory volume in one second (FEV1) and forced vital capacity (FVC) was measured with a microspirometer, following recommended procedures for standardization of pulmonary function measurements. Patients with forced expiratory volume in 1 s (FEV1) < 70% predicted were studied. SSA patients were with FEV1 increased > 30% after a 1-wk course of oral prednisone 20 mg twice daily and SRA if they failed to increase > 15%. Dolichol (Dol) in blood and urine were assayed by HPLC method. Dolichyl phosphate, alpha- and beta-GR isoforms expression were measured in CD4+ T-cells.

Results: Blood Dol concentration in persons SSA was 255.6 + 25.9 ng/mL and urinary Dol concentration was 18.9 + 4.5 mg/mmol. Blood Dol in patients with SRA was increased up to six times making up 689.2 + 47.9 ng/mL. Urinary Dol concentration was increased up to 590.9%, making up to 48.8 + 9.7 mg/mmol. The normal levels of Dol in donors are 8,0 + 2,9 mg/mmol in urine and 110,4 + 12,6 ng/mL in blood. The synthesis of DolP was 8.8–10.5-fold decreased in T-lymphocytes in patients with SRA. 10 days course of PP 5 mg supplementation in SRA patients returned DolP concentration in blood, urine and T- cells to the normal level. 8 of the 10 SRA patients demonstrated a significantly increase FEV up to 30% after 2-wk course of oral prednisone 20 mg twice daily and 5 mg of PP.

Conclusion: Dolichyl Posphate Cycle (DPC) disorder is a rate limiting mechanism of steroid resistance in asthma and associated with a marked defect of glucocorticoid receptors (GR) N- glycosylation in CD4+ T-cells. Presented findings provide evidence that PP supplementation in patients with SRA enhanced the expression of alpha GP isoforms, restore the possibility to induce IL-10 synthesis and made CD4+ T-cells more responsive to steroids. Plant Polyprenol and prednisone increase FEV and FVC in SRA patients. These preliminary results may have important implications for the design of alternative treatment approaches for steroid resistant asthma.

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Uncontrolled severe asthmatic patients clearly improve under anti-IgE (Omalizumab) treatment. 14 cases reported

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Background: Anti-IgE Monoclonal antibodies (Omalizumab) had been recently introduced in the treatment of bronchial asthma. Last October 2005, the EMEA authorized this drug in the European Union for allergic severe asthmatic patients as an add-on therapy. Experience with a 14 patient-treatment is reported.

Methods: 14 patients, 11 females and 3 males, were studied, age average 57 years, all of them suffering from severe allergic asthma (step V present GINA guideline) from the last 5–37 yr, average 19 yr. A positive skin prick test and/or specific IgE antibodies to a variety of common aeroallergens was observed in all patients. IgE sera levels were between a range of 40–1000 IU/ml (overage 316 IU/ml). FEV1 were under 80% predicted in all patients, overage (63% predicted). All patients were under treatment with high doses of inhaled corticosteroids, long acting inhaled beta-agonists and leukotriene modifiers and eleven of them also were on treatment with oral corticosteroids, prednisone, in a range from 5–20 mg. Omalizumab 150 to 600 mg per month were subcutaneously administrated to each patient once or twice monthly depending of IgE levels and body weight. Every month after onset of treatment the following parameters were evaluated: Asthma quality of life questionnaire (AQLQ, E. Juniper) Asthma control questionnaire (ACQ, E Juniper), FEV1, physician overall assessment and possibility of side effects. Patients were following up for two to ten months.

Results: A marked improvement of AQLQ and ACQ could be observed in 11 out of 14 patients. Physician overall assessment also improved in the same patients. This improvement was moderate in the remaining 3 patients. Response to treatment was appreciated into the first two months in all patients. FEV1 also improved in all patients in a range of 8 to 35% predicted. One patient developed marked edema in both legs after 12 wk and another acute otalgia and erythema nodosum in legs after 20 wk of treatment. Both patients withdrew omalizumab treatment. Oral corticosteroids were withdrawn in two patients and lowed in other four.

Conclusion: Omalizumab treatment has proved to be effective in patients with uncontrolled severe allergic asthma. A total control was shown in more than half of patients and an enough control was got in the remaining patients. No exacerbations were observed in any patient all over the omalizumab treatment. Adverse reactions possibly but not sure related to omalizumab were shown in two patients.

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Efficacy of budesonide/formoterol in the treatment of children with moderate-severe asthma

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Background: Our previous survey indicated that Chinese pediatricians tended to choose iv corticosteroids when treating moderate-severe childhood asthma,while GINA recommends the combination of inhaled corticosteroids (ICS) and long-acting β_2 agonists (LABA) for the treatment of persistent asthma. In order to evaluate one combination product budesonide/formoterol in the treatment of moderate-severe childhood asthma, we investigated its efficacy and safety.

Methods: In a 12-week study, 34 children with moderate-severe asthma (28 males, 6 females,mean age 9.7±2.5years; receiving no systemic corticosteroids 4 weeks prior to study and having no LABA or SABA 1 week before study) were given nebulized budesonide suspension and salbutamol plus ipratropium bromide to relieve acute exacerbations at the clinic, then were given budesonide/formoterol (160ug/4.5ug) one inhalation in the morning and two in the evening for 2 weeks and one inhalation twice daily afterwards till the end of 12 weeks. Oral prednisone for 1–3 days and slow release salbutamol for 3 days were prescribed to severe asthmatic children. Lung function, symptom improvements and adverse effects were mornitored after inhalation therapy.

Results: Lung function variables such as FEV1, FEV1%, FVC, FVC%, PEF, PEF%,MMEF and MMEF% were statistically improved 2 weeks and 1 month post treatment ($P<0.01$).The mean time for relief were 3–4 days after inhalation therapy. 91.2% of the children gained complete asthma control in 2 weeks and all children gained complete control at 4 weeks, 8 weeks and 12 weeks post treatment. No child had emergency treatment and short-acting β_2 agonist inhalation during the period. Such side effects as hoarseness and pharyngeal discomfort were observed.

Conclusion: Budesonide/formoterol is an effective and safe treatment option for children with moderate-severe asthma, such treatment regimen may reduce intravenous infusion, simplify therapy and improve patients' adherence to treatment.

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Therapy of chronic airway diseases with nucleic acids

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Background: RNA interference (RNAi) holds considerable promise as a therapeutic approach to silence genes involved in inflammatory pathways. It has been reported that intranasal administration of small interfering RNA (siRNA) is able to down-regulate protein expression. Aim of the project is the development of a nucleic acid based therapy for asthma and other chronic airway diseases.

Methods: Different knock-down strategies were evaluated *in vitro*. Various cell lines were transfected with siRNA or short hairpin RNA (shRNA) using different transfection reagents. Furthermore, the optimal route of application *in vivo* was investigated. The intranasal administration and the intratracheal administration of RNAi agents were investigated. For these studies constitutively GFP-expressing transgenic mice were used, which allowed the monitoring of cell specific knock-down effects. Finally genes relevant for chronic airway diseases will be knocked down *in vivo*.

Results: In comparison to other transfection reagents, a cationic lipid showed highest transfection efficiency along with lowest toxicity *in vitro* as well as *in vivo*. Using this reagent *in vivo* an uptake of shRNA in 30% of lung cells could be detected. In comparison down-regulation of GFP in cultured lung cells from transgenic GFP-mice was analysed. siRNA directed against GFP silenced GFP expression by about 40%.

Conclusion: The cationic lipid transfection reagent is suitable for RNAi *in vitro* and *in vivo*. Lung cells are amenable for RNAi agents as shown by intranasal application of a shRNA vector.

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Effect of immunotherapy with ISS-ODN and allergen in animal model of mugwort allergy

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Background: Despite a number of effective pharmacological options for the prevention and treatment of the pathophysiologic responses that occur in sensitized patients on allergen exposure, the termination of allergic hypersensitivities remains an elusive therapeutic goal. By specific immunotherapy (SIT) with allergen extracts, allergy can be cured, however it has a limited scope of efficacy. Immunostimulatory sequence oligodeoxynucleotide (ISS-ODN) was known to provide potent allergen-independent immune redirection from Th2 into Th1.

Objective: We investigated that with the animal model of mugwort allergy, the allergic inflammation is controlled by ISS-ODN and what is the mechanism of the immune redirection. Also we investigated that ISS-ODN with allergen as a model of potentiated SIT could have better anti-allergy effects on allergic inflammation induced by specific allergen over ISS-OND alone.

Methods: Experiments were performed in four groups of mice which designed to develop mugwort allergy with sensitization and aerosol challenge with mugwort extract. Group 1 received 100 mg of ISS-ODN with 110 mg of mugwort allergen as a therapeutic group, Group 2, ISS-ODN with bovine serum albumin, Group 3, C-ODN (control ODN) with mugwort allergen, Group 4, ISS-ODN alone. Cytokine profiles of IL-4, IL-5, IL-10, IL-13, and IFN-γ, degree of inflammation in lung histology, and differential counts of bronchoalveolar lavage cells were measured.

Results: Significant decreases of allergic inflammation in lung were observed in ISS-ODN with allergn group and ISS-ODN alone group comparing to negative control groups. IFN-γ was significantly increased in ISS-ODN with allergen group and ISS-ODN alone group comparing to control group.

Conclusion: Allergic inflammation is controlled by ISS-ODN with allergen, which could be a new treatment of human allergic diseases.

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Estrogen exerts protective effects in a murine model of asthma

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Background: Sex hormones have been suggested to play a role in the pathophysiology of airway diseases. Recent studies indicated an important role for estrogen in the development and severity of asthma. However, the exact effect of estrogen on lung physiology and asthma status remains poorly defined.

Objective: We investigated the effect of estradiol (E2) on airway hyperresponsiveness (AHR) in vivo and airway contraction in vitro.

Methods: A mouse model of asthma was established by sensitizing and challenging female BALB/c mice with ovalbumin (OVA). In addition, some mice were given intranasal (IN) IL-13, which induces an asthma-like disease, including airway inflammation and airway hyperresponsiveness. For in vivo studies, the effect of IN E2 treatments on methacholine (MCh)-induced bronchoconstriction was analyzed by using a Buxco system, and airway responsiveness was recorded as PenH values. An increase in bronchoconstriction is reflected by an increase in PenH values. PenH was expressed as the mean ± SEM. For in vitro studies, lungs of OVA-sensitized female BALB/c mice were sectioned and mounted in a perfusion chamber, and observed under a microscope. Images of selected airways were captured pre- and post-treatment with acetylcholine (ACh) or E2.

Results: In vivo, the MCh-induced PenH of OVA-sensitized mice before treatment with E2 was 8.30±0.58. When this group of mice was treated IN with 1 nM E2 4-hr prior to MCh challenge, their PenH was lower (5.54±0.84). OVA-sensitized mice pre-treated with IN PBS, on the other hand, did not display any change in PenH. In preliminary studies, E2 also suppressed airway responsiveness in IL-13-treated mice (PenH of 5.35±2.39 versus 7.34±1.95 for E2 and control mice, respectively). In vitro, ACh-induced airway contraction (lumen area measured 99.7 μm^2 after 1 μM ACh compared to 116.8 μm^2 at baseline) was reversed when treated with 1 μM E2 (109.1 μm^2). Pre-treatment of the airways with 1μM E2 pre-treatment blocked the ACh-induced contraction (lumen area measured 125.1 μm^2 after 1μM ACh compared to 126.8 μm^2 at baseline and 125.8 μm^2 after E2).

Conclusion: These data suggest that estradiol can inhibit and prevent AHR associated with antigen and IL-13 in a murine model of asthma. Thus, estrogen analogues might play a therapeutic role in the management of asthma.

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Engineering combination vaccines for allergic and infectious asthma

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Allergens and rhinovirus infections are among the most common elicitors of asthma, a severe disabling disease affecting more than 300 million people worldwide. We report the construction of a recombinant combination vaccine for allergen- and rhinovirus-induced asthma. Using a peptide from one of the most frequent respiratory allergens, the major timothy grass pollen allergen Phl p 1, and the human rhinovirus-derived coat protein VP1 required for infection of respiratory cells a recombinant fusion protein was produced. Immunization with this fusion protein induced in mice and rabbits protective IgG antibodies which recognized the allergen and neutralized the infection of cells expressing the receptor for human rhinoviruses. The vaccine exhibited neither IgE nor T cell-mediated allergenic activities. The described principle may be used for the combined vaccination against allergic and infectious asthma.

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Adherence to treatment by patients with asthma or COPD: Comparison between inhaled drugs and transdermal patch

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An Internet-based questionnaire study was conducted of patients with asthma and chronic obstructive pulmonary disease (COPD) and parents of children with asthma to evaluate adherence to regimens of treatment and convenience of use of inhaled and transdermal preparations. Valid responses were obtained from 1,470 patients. Among asthmatic patients, the percentage of those who selected "taking as prescribed" was 52.7% for inhalant users and 83.2% for transdermal users. Among patients with COPD, the corresponding values were 54.7% and 86.6%. There was a significant difference ($p<0.01$) in percentage between inhaled and transdermal preparations for both airway diseases. The most common reason for poor adherence was "frequency of administration", and a high proportion of patients, 83.2%, preferred once-daily administration. In addition, patients who had used both preparations preferred transdermal to inhalant drugs, to a significant extent. In conclusion, health care professionals should further educate their patients about the importance of treatment with inhalants, since poor adherence to treatment with inhaled agents significantly hinders achievement of optimal efficacy. In addition, transdermal tulobuterol patch, which is administered once daily as a long-acting beta2-agonist, appears to be useful for long-term control of both asthma and COPD.

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Aerosol characteristics of admixture of budesonide inhalation solution with beta2-agonist, procaterol

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Background: Nebulizer solutions of asthma medications are often mixed together in order to simplify inhalation regimens, although not recommended. We therefore evaluated the effect of admixture on aerosol characteristics.

Methods: An 8-stage cascade impactor was used to measure the particle size distribution of admixture of Pulmicort® Respules® (budesonide, 0.5mg/2mL) with Meptin® Inhalation Solution Unit (procaterol hydrochloride, 30μg/0.3mL) from a jet nebulizer, PARI LC Plus®. Concentrations of each drug were assayed by HPLC. Physico-chemical compatibility was also assessed up to 48 hours after mixing.

Results: Impaction analysis revealed no differences in aerodynamic size and output of each drug between admixture and single-drug solutions. The mass median aerodynamic diameter (MMAD) of budesonide from the admixture with procaterol was 2.92 ± 0.03 μm, and 2.99 ± 0.14 from single-drug solution. The respirable mass of budesonide from the admixture was comparable with that from single solution (146.8 ± 2.9, 147.6 ± 8.2 μg, respectively). There was no significant change in pH or visual identification of a precipitate in the admixture. Recovery rates of each drug kept more than 96% of the initial values during the observation period.

Conclusion: Our study demonstrated compatibility of co-administration of budesonide with procaterol in the aspect of aerodynamic characteristics and physico-chemical stability. *In vivo* data will be needed for the clinical implications of our findings.

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Efficacy of sublingual immunotherapy in asthma control

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Background: It has been of great interest to evaluate the clinical efficacy of sublingual immunotherapy (SLIT) in achieving asthma control. This interest is justified as the most recent GINA guidelines put an emphasis on control of asthma as a baseline for asthma management. Furthermore, this is the first study on SLIT in Bulgaria since this rout of administration of immunotherapy has been available for two years in the country.

The aim of the study was to investigate the potential effect of one year SLIT with dust-mites on asthma control.

Methods: 50 patients with asthma and allergy to dust mites (Dermatophagoides pternyssinus and/or Dermatophagoides farinae or both) were included in the study. 21males and 29 females, aged from 12 to 35 years (mean 23,5). SLIT has been prescribed in 25 of them (12 men) for one year along with other antiasthma therapy. A control group of 25 patients (10 men) had been maintained on medical therapy alone. Asthma Control Test /ACT/ was performed to asses the level of control for the last four weeks. On the basis of number score, asthma control was estimated as: out of control, partially controlled and completely controlled. Assessments of FVC and FEO1 were performed too.

Results: From the group of patients, subjected to SLIT + medical treatment, 12 (48%) were completely controlled, 9 (36%) – partially controlled and 4 (16%) out of control for the last four weeks of the study. From those on medical treatment alone, 5 (20%) were completely controlled, 10 (40%) partially controlled and 10 (40%) were out of control for the past four weeks. FVC and FEO1 did not differ in both groups. Patients on SLIT were better controlled ($p<0.05$) after the first year of treatment.

Conclusion: The results of the study suggest that SLIT affords additional benefit to medical treatment in patients with asthma and allergy to dust mites. The results suggest that SLIT, added to medical treatment is of advantage in achieving asthma control.

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Evolution of quality of life in patients treated with therapeutic vaccine containing depigmented and polymerized allergen extracts of dermatophagoides pteronyssinus and D. farinae in allergic asthmatic patients: results of a double-blind placebo controlled study

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Background: Depigmented and polymerised allergen vaccines have shown to be efficacious using objective and subjective outcomes. It is now recognized that treatments under evaluation should enable patients to feel better in their day-to-day life activities. The objective was to evaluate the impact in the quality of life of a group of 64 asthmatic patients sensitised to house dust mites.

Materials and Methods: The study was double-blind and placebo-controlled. Sixty-four patients were randomly allocated to either receive active treatment (n=32) or placebo (n=32). The active treatment was a therapeutic vaccine containing a mixture of depigmented and glutaraldehyde polymerized mixture of allergen extracts of D. pteronyssinus and D. farinae. The Asthma Quality of Life Questionnaire developed by Marks was used. It was conducted at baseline and after each administration of allergen vaccine (19 times). This is a 20-item self administered questionnaire, developed to measure quality of life in adult subjects with asthma by measuring a total scale score together with subscale scores for breathlessness, mood disturbance, social disruption and concerns for health. The result was expressed as the mean of all 19 measurements, Mann-Whitney's test was used to evaluate the statistical differences between both groups, and Hodges-Lehmann for the effect size of these differences.

Results: The AQLQ overall score was the sum of all individual scores. The values of the mean for these scores were 7.44 (95% CI: 5.78–9.11) for the active group and 11.44 (95% CI: 9.67–13.22) for the placebo. The differences between both groups were significant ($p=0.043$, Mann-Whitney's test) and relevant (Hodges-Lehmann test: −4.36, 95% CL: −8.69,−0.10). The active group showed an improvement of 34.98% over placebo.

Conclusion: The results of this clinical trial show that the immunotherapy-treated patients, in contrast with untreated, experience a significant improvement in the overall quality of life related to asthma. The statistical significance as well as the improvement on the active group compared with the placebo occurred also in the domains of breathlessness, social disruption and concerns for health, with the exception for mood disturbance.

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The leukotriene receptor antagonist pranlukast improves quality of life in poorly controlled asthmatic patients

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Background: Asthma is a chronic inflammatory disease, and inhaled corticosteroids have been shown to be very effective in controlling symptoms. However, it is important to minimize the dosage of corticosteroids used because of local and systemic adverse effects. The importance of leukotrienes in the pathogenesis of asthma is established and many studies have shown that leukotriene receptor antagonists can improve asthma control in patients with poorly controlled asthma despite the use of inhaled corticosteroids.

Objective: The aim of this study was to assess the effects of 8 weeks' pranlukast treatment on quality of life and pulmonary function in patients with inadequately controlled asthma.

Methods: This study was conducted at 312 hospitals and clinics in Japan from October 2004 to March 2005. We recruited 1546 outpatients (mean age 51.4±18.5 years) with mild-to-severe asthma (as defined by the Japanese Society of Allergology) who were not well controlled with daily antiasthmatic treatment. During the 8-week treatment period, patients received pranlukast (225mg, twice a day) in addition to their previous treatment. Patients were required to complete symptom diaries and record peak expiratory flow (PEF) on a daily basis. Quality of life (QOL) was assessed using the Asthma Health Questionnaire (AHQ).

Results: A total of 839 asthmatic patients were enrolled. Significant improvements were observed in the AHQ total score and all subscale scores. When the patients were classified according to baseline disease severity (mild, moderate and severe), the improvement from baseline remained significant for

all AHQ subscale scores other than the economics subscale in all patient groups.

Patients also demonstrated significant improvements from baseline in PEF (+34.9 L/min; $p < 0.001$), forced vital capacity (+0.17L) and forced expiratory volume in 1 second (+0.14L; both $p < 0.005$).

Conclusion: The leukotriene receptor antagonist pranlukast improves QOL and pulmonary function in asthmatic patients inadequately controlled with antiasthmatic therapy.

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Effectiveness of short-term oral corticosteroid for preventing relapse following the emergency treatment of acute asthma

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Background: Acute asthma is responsible for many emergency department (ED) visits. It has been shown that high-dose intravenous methylprednisolone in addition to standard emergency treatment for acute asthma induces the early termination of the asthmatic attacks and reduces the number of hospitalizations. In addition, it has been reported that about 15 to 30% of the asthmatic patients treated in the ED will relapse to require additional interventions within 2 weeks of ED discharge. However, little is known about the efficacy of oral corticosteroid in reducing the relapse in the asthmatic patients discharged from the ED after treatment of acute asthma. The purpose of this study was to determine the effectiveness of oral corticosteroid in reducing relapse following the emergency treatment of acute asthma.

Methods: Patients with acute asthma were treated with nebulized salbutamol and intravenous 125 mg methylprednisolone and 200 mg theophylline and were then discharged from the ED. Upon discharge, some patients were given oral 20 to 30 mg/day prednisolone for 5 days.

Results: The asthmatic patients who received oral prednisolone had a significant decrease in the need for repeated emergency care (5.4%, 4 out of 74 patients, $p<0.0001$) within 2 weeks as compared with those who did not (33.3%, 73 out of 219 patients). The mean dose of prednisolone was 24.4 mg/day and the mean duration was for 4.4 days.

Conclusion: It is concluded that a 5-day course of medium-dose oral prednisolone (0.5 mg/kg/day) in outpatients prevents the relapse following the emergency treatment of acute asthma.

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Management of childhood asthma in east-java indonesia

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Background: Treatment guidelines for childhood asthma is available, but it is important to know whether this guideline is being used in East Java.

Methods: Using a standardized questionnaire on monitoring and treatment of childhood asthma, distributed to doctors treating childhood asthma in East Java.

Results: Only 70 (20%) out of 350 doctors participated in this questionnaire study. General practitioners were 75.7%, 5.7% paediatricians and 2.9% pulmonologists. 88.6% worked in policlinic or private hospitals, and only 5.7% worked in university hospitals. Symptom score, peak flow meter or spirometry never or seldom used to monitor childhood asthma (94%). Nebulised salbutamol/terbutaline was the first drug of choice (63.8%) for treating acute asthma attack, followed by iv or im corticosteroids (23.7%), and iv aminophylline (30.6%). The three main criteria for admitting acute attacks were based on personal clinical judgement (80.0%), no improvement after 2 or 3 doses of salbutamol inhaler (44.4%) and other factors such as home distance and travel time (37.0%). IV aminophylline was seldom used (60.0%). Systemic (57.1%) and inhaled corticosteroids (50.7%) were seldom used in outpatient setting. Corticosteroids were used in all admitted patients. The corticosteroid of choice were dexamethasone (50.0%), methylprednisolone(45.7%), and were used same the dose for 5 days. Oxygen therapy, based on pulse oximetry reading were used only for severe asthma (45.6%). Antibiotics were used only in case of pneumonia, otitis media or sinusitis (40.0%). Drug of choice in the maintenance treatment of asthma in all ages were the same: LABA was the first choice, followed by LABA + inhaled corticosteroid and inhaled corticosteroids. Maintenance treatment was seldom used in all ages. The main criterium to start a maintenance treatment was the severity of the symptoms (47.1%). Specific immunotherapy never been used (72.1%).

Conclusion: Although treatment guidelines for asthma in children is available, in daily practice we still find many modifications, including the usage of a LABA as first choice maintenance treatment. More efforts in socialization of the guidelines and providing tools for monitoring and treatment of asthma in children are needed.

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The use of yoga as an adjuvant therapy in the management of bronchial asthma: based on a randomized controlled trail

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Background: Complementary and alternative medicine (CAM) therapies such as herbal therapy, acupuncture, yoga, chiropractic, relaxation techniques, nutrition and dietary supplements, continue to gain popularity as modalities for the management of asthma. However there is a dearth of qualitative research with strong methodology to understand plausible mechanism of action of these therapies on asthma. The present randomized controlled trial (RCT) examines the use of yoga as an adjuvant therapy in the management of bronchial asthma.

Methods: The study was conducted on 57 adult patients having mild or moderate bronchial asthma who were allocated randomly to either the yoga group ($n = 29$) or the wait-listed control group ($n = 28$). The control group received conventional treatment whereas the yoga group received, in addition to conventional treatment, also an intervention based on certain yogic practices. The outcome measures were assessed in both groups at 0 wk (baseline), 2 wk, 4 wk and 8 wk.

Results: In the yoga group, there was a steady and progressive improvement in pulmonary function. There was also a trend towards a reduction in exercise-induced fall in timed vital capacity (FEV1) in the exercise-sensitive (exercise induced asthma) subgroup of subjects in the yoga group. However, there was no corresponding reduction in the excretion of urinary prostaglandin D2 metabolite (11β-PGF2α) in response to the exercise challenge, which is an indicator of mast cell activation. There was also no significant chan there was a significant reduction in serum soluble interleukin-2 receptor (sIL-2R) levels after 2 wk in yoga group alone. NNT for quality of life indicators worked out to be 2.41 in QOL symptoms, 1.66 in QOL activity limitation, 1.91 in QOL emotional function, 1.70 in QOL environmental stimuli and 1.82 in total quality of life.

Conclusion: The results indicate that of yoga in addition to conventional treatment as an adjuvant therapy in asthmatic patients results in improvement in pulmonary function and quality of life; reduction of T-cell activation and response to exercise stimuli. These findings provide yet another substantial evidence towards the use of yoga as an adjuvant therapy in the management of bronchial asthma.

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The effect of inhaled steroids and long-acting beta2 -agonists on oxidative stress in stable asthmatics

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Background: The effect of inhaled steroids on oxidative stress in asthmatics is unclear. In this study the levels of lipid peroxides in the serum of asthmatic patients, whose symptoms were controlled with inhaled corticosteroids and long-acting beta(2)-agonists, were measured.

Methods: Twenty-four asthmatic patients and 15 matched, healthy controls were recruited. Oxidative stress levels were quantified by measuring thiobarbituric acid reactive substances.

Results: After 3 months of treatment, the mean lipid peroxide concentrations were significantly higher in asthmatic patients than in the healthy controls (4.3 +/− 0.15 micromol/mL vs. 3.4 +/− 0.03 micromol/mL, respectively).

Conclusion: The parameters of oxidative stress was higher in patients with asthma than in healthy controls, even when the asthma is well controlled after 3 months of treatment.

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Effect of thalidomide in murine model of asthma

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Background: Tumor necrosis factor-alpha (TNF-alpha) has been identified as a proinflammatory cytokine that might be important in airway hyperresponsiveness (AHR). Thalidomide exhibits diverse actions of anti-inflammation and immunomodulation including attenuation of TNF-alpha production.

Objective: To evaluate the role of thalidomide in murine model of asthma.

Methods: Six-week-old female C57BL/6 mice were sensitized with LPS 0.1 mg/ml and ovalbumin (OVA) via intranasal route and were exposed to OVA via intranasal route for 3 days. Thalidomide 50mg/kg was given via gavage twice a day from a day before the challenge. Airway responsiveness, inflammatory cells, cytokines in bronchoalveolar lavage fluids (BALF), serum OVA-specific IgE, IgG1, IgG2a, and histological examination were carried out.

Results: Airway hyperresponsiveness decreased significantly in mice treated with thalidomide. There were no differences in IL-4, IL-5, IL-12, IFN-gamma, and eotaxin levels in BALF.

Conclusion: Thalidomide decreases AHR in murine model of asthma.

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Therapeutic comparison between low-dose sustained-release theophylline dry syrup and capsule in children with mild persistent asthma

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Theophylline has been recently reported to have concurrent anti-inflammatory effects at low therapeutic plasma concentration which is below the dose at which significant clinically useful bronchodilatation is evident. Sustained-release formulation as capsule and dry syrup forms was developed to reduce its adverse effects and improve its clinical effects. We compared between therapeutic effects of theophylline dry syrup and capsules in children with mild asthma.

Methods: Ninety children with mild asthma were randomized to receive either theophylline dry syrup (n = 44) or theophylline capsules (n = 46), 4 mg per kilogram of body weight twice a day for 12 weeks. Baseline and serial measurements of daytime and nighttime asthma symptom score were performed. Compliance score, drug swallowing score, and drug usability score were measured every 4 weeks. Each scoring rated on a scale of 0–4. Serum theophylline concentration were mesured at 4 and 12 weeks. To examine anti-inflammatory effect of theophylline on asthma, Serum eosinophilic cationic protein as a marker of airway inflammation caused by eosinophil was measured 12 weeks pre- and post-administration.

Results: Daytime and nighttime asthma symptom score of two groups after 4 weeks significantly improved than baseline score. Daytime and nighttime asthma symptom score in dry syrup group were statistically lower at all time points except for nighttime symptom score at 4 weeks. Compliance score, drug swallowing score, and drug usability score in dry syrup group were significantly higher at end time point. Only in dry syrup group, serum ECP at end time point was statistically lower than baseline.

Conclusion: Low-dose sustained-release theophylline may be safe and effective in bronchial asthma and this effect may be mediated by its anti-inflammatory action mechanisms. Especially, when used in children with asthma, dry syrup formulation is recommended because of its higher compliance than that of capsule formulation.

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Steroid-dependent bronchial asthma: how to reduce the dose of oral glucocorticosteroids and improve asthma control?

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Background: Patients with chronic severe asthma are often dependent on the long term prescription of oral glucocorticosteroids. The use of steroids is associated with serious side effects. Physicians treating such patients continue to search for alternative therapies that reduce the need for chronic dosing oral steroids.

Aim: To assess efficacy and safety of 24- months treatment with Salmeterol/ Fluticasone propionate (100/ 1000 mcg daily) and Fluticasone propionate (1000 mcg daily) in 105 steroid-dependent bronchial asthma patients aged 18 to 60 years, who had received oral glucocorticosteroids (predisolone equivalent) less than 10 mg daily (75,2%) and more than 10 mg daily (23,8%) for at least 6 months but not more than 10 years.

Methods: This was comparative, randomized, multicentre, open study in parallel groups (15 visits at 2 weeks intervals). Oral glucocorticosteroids dose reduction phase started at 3-rd visit.

Results: Inspite of continual treatment with oral glucocorticosteroids patients did not reach asthma control. After 6-months treatment maintenance dose of prednisolone was 6 times lower in comparison with baseline. Moreover, oral glucocorticosteroids were abolished in each second patient receving Salmeterol/ fluticasone propionate and each third patient receiving Fluticasone propionate. At the same time asthma control and quality of life significantly increased. Administration high doses of fluticasone propionate and decreasing dose of oral glucocorticosteroids lead to reliable increase the morning level of serum cortisol by the sixth month of treatment in both groups of patients.

Conclusion: The treatment with Fluticasone propionate and Salmeterol/ fluticasone propionate allows reduce the dose of oral glucocorticosteroids, moreover in the most causes patients can abolish oral glucocorticosteroids maintaining good asthma control.

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Cycloferon treatment of atopic bronchial asthma exacerbations with respiratory infections

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Respiratory infections (viral and bacterial) may be trigger of atopic bronchial asthma (ABA) exacerbations. Was revealed II-III degree Interferon (IFN)-a and IFN-g production deficiency of leukocytes by all 19 patients with moderate and mild ABA in unstable remission phase and also acute mixed (viral and bacterial) respiratory infections. The purpose of this study was to use low molecular IFN inducer cycloferon for to improve IFN system indices and to reduce ABA exacerbations rate. It is known, that low molecular IFN inducer cycloferon have both antiviral and immunomodulating activities, can be successfully used in acute respiratory viral diseases. Cycloferon have affinity to alveolar macrophage receptors and induce of IFN in lungs. After the administration of cycloferon IFN-a and IFN-g producing capacity of leukocytes was 2-fold higher in comparison before treatment ($p<0,05$). 82% of patients (n=9) in group with cycloferon and basic therapy had positive clinical effect versus 50% of patients (n=4) with only basic therapy. Effective schemes of therapy viral/bacterial infections with use antiviral drugs, IFN inducers and IFN preparations, having both immunomodulatory properties and antiviral/antibacterial effects, have to be worked out for improvement of treatment efficacy of patients with BA exacerbations.

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The effect of inhaled beclomethason dipropionate (BDP) versus budesonide (B) in asthmatic patients-comparative, randomized, double blind clinical study

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Aim: The aim of the study was to compare the two different topical corticosteroids in the treatment of asthmatic patients.

Material and Methods: 100 naive asthmatic patients with the baseline values of FEV1 from 60-80% were divided in two groups. The first group received BDP and second received B aerosol in total of 800 μg/day, for 4 weeks. Short acting β2 agonist (salbutamol) was used as needed. Clinical symptoms, FEV1, FVC, PERF and the usage of β2 agonist were recorded. Local adverse effects were also documented.

Results: Indices of dynamics show an increase of the values of FEV1 and FVC during the four weeks' therapy with B and BDP, were in average of 22% and 15,6%, respectively. In patients who took B, FEV1 was increased in average 25%, and in patients who took BDP 19%; the difference in proportions was not statistically significant, it was accidental in our sample ($p<0,5$). In patients who took B, FVC was increased in average 16,3%, and in patients, who took BDP 15%, the difference in proportions was not statistically significant, it was accidental in our sample ($p<0,88$). With B, minimal increase of FEV1 was 9,3%, and maximal was up to 51% of the baseline value. Minimal increase of FVC was 3,7%, and maximal increase of the value was 36% after therapy with B. With BDP minimal increase of FEV1 was 5,8% and maximal was up to 40% of the baseline value. Minimal increase of FVC was of 5%, and maximal increase of the value was 31% after therapy with BDP. The difference between morning and evening values of PEFR under the influence of B became reduced in 86% and under the influence of BDP in 92%, the difference in proportions was not statistically significant ($p<0,08$). 25 patients complied on sore throat 19 on hoarseness and in 6 Candida albicans was isolated.

Conclusion: The therapy with topical corticosteroids improved the clinical symptoms and values of FVC, FEV1 and PERF in both study groups, but that difference between them was not statistically significant.

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Fluticasone propionate/salmeterol combination compared to Fluticasone propionate alone in asthma (our experience)

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Background: The rates of morbidity and mortality associated with asthma are increasing.Asthma is chronic inflammatory disease characterized by recurrent attacks of dyspnea associated with wheezing. Inhaled corticosteroids are the effective controller medications currently available.ICS may not provide optimal control of asthma when taken alone and additional therapy is necessary.

Objective: The objective of the research work is to identify the efficiency of Fluticasone/salmeterol (discus inhaler) a 250 μg tvice dauly compared to Fluticasone a 250μg twice dauly in patients with asthma. We have analyzed a group of 35 asthmatics (aged 21–32) being sensitive to allergen dermatophagoides pt and grass pollen. We split them up into two groups:group I consisted of patients who have undergone FP/Sal. (no=19); group II were the patients who have been treated by Fluticasone alone (no=21). All of them have been given a skin sensitivity test to Dermatophagoides pt. from 2+ to 3+,symptom score (0–30,FEV1,morning and evening expiratory flow.

Results: The results of the nvestigation showed statistically significant improvement of thr score symptoms with the first group:the reduction of the number of asthmatic approaches,the increase of FEV1 from (78,1±3,2) to (95,3±2,1) and a cough prevention ($p<0,01$).Following a 12 weeks tratmwnt,93% of the patients from the first group and 73% of the patients from the second group have not shown any side effects during the treatment period.We have notice significantly greater improvements in morning and evening peak expiratory flow of the patients from the first group.Patients from the second group needed additional therapy of short acting beta2agonist.

Conclusion: Patients on FP/Sal. had significantly greater improvement in asthma control than patients on Fluticasone allone.

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Study of inhalation therapy with special reference to inhalation technique and patient's perception

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Drug delivery by aerosol rout is often regarded as a twentieth century innovation,but in fact records of inhalation therapy can be found in the writtings of ancient cultures, notably those in China, India, Greece, Rome and the Middle East. The modern era of inhalation dates back from 70 years ago. One of the major problem in the management of Bronchial Asthma and COPD lies not in the airways but on the prescription. Technique of inhalation is crucial for the management of such diseases. With this background the present study was designed to find out, How the patients are using their inhalation devices? How does patient education affect their inhalation technique? What are their views and apprehensions regarding inhalation therapy and problems they face with this mode of therapy. The study was undertaken in 284 patients, 196 using Dry Powder Inhalation and 88 using MDI. The results were evaluated and we found that about half of the patients were not using their devices correctly. Among incorrect users majority were those with MDI. Among correct users majority of patients were educated the technique by their

physicians or paramedics. One fourth of patients were wrong while preparing for inhalation, more than 80% incorrect users were not breathing out before inhalation, half of the patients were inhaling drug with inadequate efforts and more than 85% were not holding breath after inhalation. Cough, sudden spasm and dysphonia were the pr5oblems faced by 20%, and fear of addiction was the most common apprehension of using inhalation. Details of the study will be presented.

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A psychosocial perspective: Asthma in the third world, a portrait of a society malady

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Asthma in Latin America is a pressing and growing health problem affecting the young and mostly impoverished urban populations across the continent. Venezuela, an oil rich country with 80 % or more of the population living under variable conditions of poverty, has an acute asthma morbidity that ranks ahead of diarrheas and seconds the "viral syndrome" and thus representing more than one million visits per year at the ambulatory health services of the Ministry of Health (caring for the majority of the population), a tendency constantly on the rise for roughly 27 million inhabitants.

Efforts for over 25 years by an existing National Asthma Program and the dissemination of GINA and similar guidelines have all been unable to reverse this trend; the prevailing asthma approach centers around a perverse paradigm of vicious cycles consisting of repeated acute asthma care; this boldly denotes a morbidly persistent lack of awareness in the minds of physicians (Health System), patients and lay public at large.

A unique trait, valid alike for the upper rich as well as Venezuelas' poor and marginal classes, is acknowledged from many years of psychotherapeutic practice: it is a tendency of sorts to live in a "state of emergency", a lacking in future provisions of any kind and an attitude of strong accent on the immediacy of life along with no risk prevention. Poverty, understood as a "precarious condition of daily life", lies beneath these set of highly complex issues. Our asthma abounds in these attributes and have been in part considered a psychosomatic expression of this malady i.e.: "if....when a next asthma attack or crisis comes, we'll then see about it". The above represents an overwhelming historical challenge that heavily pounds on individual minds and society, undermining a much needed reflection and making the surge of a proper response no easy task.

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Allergy & asthma training program in India

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Background: India with a population of a billion and estimated number of allergy & asthma patients running into the millions, had no formal training program in Allergy & Asthma. In spite of significant advances in pharmacological infrastructure, there has been a total lack of formal physician educational programs in this growing important field of medicine.

Methods: International Asthma Services (IAS) a charitable organization started by P.K.Vedanthan MD in the late 80s in Colorado,USA dedicated to patient and physician education in allergy and asthma in several parts of the globe, has partnered with Christian Medical College & hospital (CMC) at Vellore,India to initiate a formal training program in the field of allergy & asthma for the first time in India.

Diploma in allergy & asthma (DAA) is a year long distance learning course, with Personal Contact sessions(PCS) every quarter, with final oral and written exams at the end of the course. The course is limited to 20 post graduate doctors specialized either in medicine,Pediatrics, respiratory medicine will be selected. International and national faculty teach in this course.

Results: The first batch of 18 physicians hailing from different parts of India will be awarded the Diploma this July 2007. The 2nd batch of 20 physicians will be starting the course in the later part of July 2007.

Conclusion: This unique partnership between IAS & CMC will hopefully meet the present challenges of unmet physician manpower needs in the field of allergy & asthma in the Indian subcontinent over the coming years. Allergy-Asthma network of India is also is in the process of formation to facilitate free flow of educational,research, charitable activities throughout the country.

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The implementation process of asthma guidelines in primary health care

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Background and Aim: To improve the clinical practices is known to be a challenging process. Here we describe the implementation of asthma guidelines and the development of the house rule and patient self management protocol. The setting is Leppävaara health care centre with six GP-nurse teams each providing primary care for 10 000 inhabitants.

National Asthma Programme 1994–2004 delegated the responsibility in preventing and treating asthma to the primary health care. The evidence based guidelines published 2000, updated 2006, gives the general outline in asthma treatment.

Materials and Methods: Participatory observation and document analysis using qualitative methods

Results: Two GPs and two nurses were nominated to asthma co-ordinators to act as internal consultants. They organized many workshops during 2000–2007 to introduce the guidelines and to develop asthma management. The governmental Centre for Pharmacotherapy Development joined the implementation activities by providing training of trainers, and material and tutorial support. Therefore, the workshops after the year 2003 were problem-based and multi-professional, and used EBM, feed-back of prescription practises, and case reports as learning methods.

The Leppävaara health centre team produced a written agreement of inter-professional collaboration and asthma management process, the Leppävaara house rule of asthma care, in a workshop in 2004. They arranged a new workshop in 2005 to assess the changes in asthma care. A multi-professional group developed a written self management protocol for the patients in 2006. The team renewed the house rule in a workshop in 2007 according to updated national asthma guidelines.

Discussion: The evidence based asthma guideline was a valuable "golden reference". To implement it the interactive multi-professional workshops were used to develop the house rule. This method seemed to support the professionals to commit to true team work and to jointly agreed practices.

The implementation has been a necessary process, which has to be continued by supporting, giving feedback and evaluating.

We will evaluate the professionals' attitudes towards the usefulness of the house rule, the rate of admissions to the secondary care, and changes in prescription patterns.

Producing guidelines is not enough: The improvement of clinical practices demand the interesting and useful implementation process to every day work.

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Clinical implications of immunological and inflammatory markers in exercise induced bronchospasm

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Exercise-induced bronchospasm (EIB) is widely prevalent in asthmatic individuals particularly in children varying from 40% to 90% in various reports. There are, however, few studies addressing the effects of asthma severity on airway responsiveness to exercise from India.

Aim: In examining the mechanisms of exercise-induced bronchospasm (EIB), it is important to determine which factors mainly affect the severity of EIB. We report such factors in patients with asthma by stepwise multiple-regression analysis.

Methods: Sixty five asthmatic children (age range 10–14 years), 200 adults between 20 to 44 years old were selected randomly as representative of persons living in the city of Mumbai. All subjects filled out a questionnaire on respiratory symptoms. Bronchodilator therapy was withheld three days before the study. The exposure to house dust mite allergens was estimated from dust samples obtained in the subjects' homes. Eosinophil count was done from peripheral blood. Serum Il-4 was estimated in adult asthmatics by ELISA method. Atopic status was labeled on the basis of Total serum IgE and Modified Skin Prick Test and subjected to exercise challenge. assessments of atopy. An exercise challenge test was done (8 min) bout of cycle-ergometer exercise): decrease in PEFR of 15% was considered positive for EIB.

Results: Atopic adults showed significantly higher sensitization to house dust mite compared to children ($p < 0.05$). Significant rise in incidence of EIB was observed in adults (62.6%) against 40% in children ($p < 0.05$) showing direct relationship of house dust mite sensitization in both the groups (adults correlation coefficients: $r = 0.955$ $p < 0.001$, children $r = 03135$ $p \pm 0.05$). Relative risk of developing EIB associated with HDM sensitivity was higher in adults (2.85 at 95% CI). Regression analysis showed direct relationship between the severity of EIB and serum IL-4 levels ($r2 = 0.03$ $p < 0.0001$). Raised eosinophils were well correlated with serum IL-4 levels.

Conclusion: The findings suggest that though EIB is common in children, adults were more significantly affected. Allergen specific IgE to house dust mite, serum IL-4 and eosinophilia were the major associated contributing factors in the development of EIB in asthmatic patients.

Clinical Implications: Patients with exercise induced bronchospasm may benefit from novel tharapies specially designed to target the specfic mechanisms underlying airway inflammation.

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Asthma control and Quality of Life in asthmatic patients

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Background: As indicated in Asthma guidelines, treatments' goal should be the total control of asthma and consequently a minimal impact of asthma on patient's life.

Aim: The aim of our study was to investigate the relationship between FEV1, control of asthma and quality of life in real life.

Methods: In a 1 month period, asthmatic patients were enrolled consecutively during their routine control visit. A spirometry was performed for every patient. Patients were asked to complete 2 different questionnaires: the Asthma Control Test (ACT), a brief instrument developed to assess asthma control of patients in a clinical setting, and the Rhinasthma, a 30 items tool aimed at evaluating the impact of respiratory allergy on quality of life. It provides 4 different scores: Upper Airways (UA), Lower Airways (LA), Respiratory Allergy Impact (RAI) and Global Summary (GS).

Results: Seventy-four asthmatic outpatients (30 men and 44 women; mean age 44.11 + 14.83) were enrolled. 53 had concomitant rhinitis. For 39 patients (52.7%) the control of asthma was achieved; 35 (47.3%) didn't obtain asthma control. FEV1 value (mean 88.45 + 16.13) correlates with ACT and with

Rhinasthma, LA and RAI. ACT significantly correlated with the four Rhinasthma scores. Subdividing patients according to the score (optimal and good control vs non control) and comparing the level of control measured by means of ACT we obtained a significant correlation between ACT and LA (p. 0.0001), RAI (p. 0.01) and GS (0.0001) and no correlation with Rhinasthma UA score (p 0.053). The analysis of the patients' population showed that a well or total control was achieved in 53% of patients with asthma and 51% of patients with both asthma and rhintis. The subanalysis of 53 patients with concomitant rhinitis showed worse Rhinasthma scores for all the domains compared with patients with asthma alone (p. < 0.05).

Discussion: Our results show that asthma control is related to an improvement in quality of life; nevertheless, asthmatic patients often refer also symptoms due to rhinitis that have an important impact on their Quality of Life. So, in order to improve Quality of Life of asthmatic patients, the treatment of patients with rhinitis and asthma includes both lower and upper airways treatment.

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Relationship between quality of life related to chronic cough and asthma level of control; a pilot study

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Background: The antiasthmatic treatments nowadays available can induce a good or total disease control. Recent evidences have also shown that the level of control is strongly related to health related quality of life (HRQL).

Aim: The aim of our real life observational study was to evaluate the relationship between HRQL related to chronic cough and asthma level of control defined according to Asthma Control Test score (ACT).

Methods: In a 1-month period, asthmatic patients were consecutively enrolled during their scheduled visits. Patients were asked to complete 2 different questionnaires: the ACT, a brief instrument developed to assess asthma control of patients in a clinical setting, and the Chronic Cough Impact Questionnaire (CCIQ), a 16 items validated questionnaire divided into 4 areas (sleep/concentration, social relationship, mood and daily life impact).

Results: Seventy-four asthmatic outpatients (30 men and 44 women; mean age 44.11 SD: +/-14.83) were enrolled. 53 had concomitant rhinitis. 39 patients (52.7%) had total or well controlled asthma (ACT score > 20); 35 (47.3%) had non controlled asthma (ACT score < 20) .

ACT didn't correlate significantly with any of the CCIQ domains: sleep/concentration (p. 0.689), social relationship (p. 0.741), mood (p. 0.704), and daily life impact (p. 0.458).

Taking into consideration that concomitant rhinitis could induce a post nasal drip related cough, the subanalysis of patients with both asthma and rhinitis didn't show any correlation between CCIQ and ACT.

Discussion: Previous studies showed a significant correlation between asthma level of control and HRQL. Despite these evidences, analysing the HRQL related to a specific symptom (cough) occurring in asthma, the correlation is lacking. Our pilot observations seem to demonstrate that ACT is an useful tool, but a specific instrument should ameliorate patients' evaluation. Moreover, the presence of other causes of cough should be investigated in well controlled asthmatic patients with persistent cough.

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Clinical effect of clean air administered directly to the breathing zone (Airsonett Airshower®) on perennial allergic asthma

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Background: Studies previously made on air cleaning has shown little or no effect on patients with perennial allergic asthma. We examined a novel treatment (Airsonett Airshower®) using a laminar airflow directed to the breathing zone of a patient during the night, on teenagers and young adults with moderately severe allergic asthma. We hypothesised that this directed treatment would have effect on the bronchial inflammation and consequently, the quality of life.

Methods: 22 patients 12–33 year of age (mean 18.8 yr) was randomized double-blind to 10 weeks active treatment respective 10 weeks placebo treatment. All patients received both active and placebo treatment (cross-over) with a 2 week wash-out period in between treatments. Maintenance treatment with inhaled corticosteroids was unaltered (400 µg budesonid/day or equivalent) during the trial period. Health related quality of life, miniAQLQ, was the primary effectiveness measure. Exhaled nitric oxide (FENO) and lung function was also investigated.

Results: Active treatment resulted in an improved quality of life compared to placebo (mean score 0.54, p < 0.05, n = 20). Also an effect on the bronchial inflammation was detected with significantly lower FENO values during the active treatment period (mean -6.95 ppb, p < 0.05, n = 22).

Conclusion: The result of clean air, administered directly to the breathing zone during sleep, can have effect on the bronchial inflammation and consequently, the quality of life on patients with perennial allergic asthma.

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Current status of Korean traditional medicine for Korean allergic patients

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Background and Aim: Korean traditional medicine (KTM) is popular in Korea. We evaluated the prevalence and compliance of these unproved practices in Korean allergic patients.

Methods: 649 allergic patients were enrolled from 10 general hospitals. Their mean age was 42.5±15.3 years old. Proportions of asthma, allergic rhinitis, atopic dermatitis, and urticaria were 50.7%, 41.3%, 26%, and 11.3%, respectively. The Questionnaire which consisted of 12 items regards the prescription rates, reasons for referring to these remedies, and their opinions for the efficacy of KTM.

Results: 259 (39.9%) patients had experienced KTM (herbal decoction ?34.2%, herbal pills ?6.0%, acupuncture ?12.3%, moxa cautery ?4.1%), and 33.2% of the patients with the history of KTM experienced 2 or more KTM practices. Patients?income or final academic career did not affect the prescription rates of KTM. 34.6% of the KTM experienced patients were satisfied to the practices (herbal decoction ?35.6%, herbal pills ?22.2%, acupuncture ?34.2%, and moxa cautery ?44.4%), and 40.6% of the KTM experienced patients were inclined to further KTM practices. The patients with respiratory allergic diseases were more satisfactory to KTM than those with allergic skin diseases (p = 0.006). The most frequent reasons for choosing the KTM were safety (15.6%), worries about adverse reactions of the proved drugs (20.2%), and boostering effects on immunity or physical predisposition by KTM (36.3%). However, 18.9% of the patients with KTM had experienced the adverse reactions to the practices; skin rashes ?13.5%, gastrointestinal discomforts ?2.3%, and hepatitis ?0.7%. The prescribed patients expensed 915 US dollars/year on average.

Conclusion: Many Korean allergic patients are compliant to KTM and experienced significant adverse reactions. Educations for the proved managements are necessary in Korea.

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Anxiety symptoms in allergic patients: identification and risk factors

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Objective: Multiple relationships between anxiety, allergic symptoms, and treatment difficulties have been observed. The aim of the present study was to estimate the prevalence of anxiety disorders in outpatients with various allergic diseases, to identify diagnostic cues or possible risk factors, and to test the usefulness of self-administered questionnaire screening at the allergy clinic.

Methods: Six hundred forty-six (646) consecutive patients with rhinoconjunctivitis (59.3%), asthma (26.8%), or "other" allergy (13.9%), aged 16 to 65 years, completed self-administered questionnaires in six outpatient allergy clinics; 60 of the respondents also participated in structured psychiatric interviews. Anxiety was measured with the Spielberger State-Trait Anxiety.

Results: According to the interviews, STAI-T > 52 predicted with 86% accuracy a current psychiatric diagnosis, without differentiating between anxiety and depression. Using this threshold, the rate of anxiety and/or depressive disorders is estimated as 19% (95% CI: 15.9–22.1) in our unselected allergic outpatient sample; 46% of these patients never received any psychopharmacological treatment, indicating that anxiety related disorders are underdiagnosed and undertreated. Risk indicators were female gender; asthma; perennial symptoms; sleep problems; nonspecific allergy triggers like strong emotions; stressful situations; and considerable limitation in everyday activities attributed to the allergic symptoms.

Conclusion: Our findings confirm a high rate of anxiety and/or depressive disorders in patients visiting the allergy clinic. Self-administered questionnaires such as STAI-T provide reliable help for the identification of these frequent psychiatric problems.

Key Words: anxiety, allergy, rhinitis, asthma, Spielberger State-Trait Anxiety Inventory, depression.

Abbreviations: DSM-IV = Diagnostic and Statistical Manual of Mental Disorders fourth edition; STAI-T = trait anxiety and STAI-S: state anxiety subscales of the Spielberger State-Trait Anxiety Inventory.

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Healthcare assessment methodology in developing country

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Issues: Health care assessment system in Nepal is still take long time and high cost. Collection of data about prioritization of community health problems by the community is essential for planning and monitoring of programs and interventions for improving community health status. Scientific Rapid Community Health Assessment Methodology needs to be validated in rural community developing country like Nepal which is economic and less time consuming.

Objective: To validate the Rapid Community Health Assessment Methodology (RCHA) for prioritization of community health problems in a rural community.

Study Area: 14 Villages of rural areas. STUDY POPULATION: 34 Primary School Teachers from 10 randomly selected Primary Schools in above villages. (One Teacher each from Class I to V, in each School). VALIDATION: Heads of households from 500 Households (50 per village, systematically sampled). DATA Collection technique: Self-Administered Questionnaires for Teachers, Interview Schedules for Heads of Households. DATA ANALYSIS: with the help of EPI info program.

Results: There was significant correlation between the responses of the school teachers and heads of households on community health problems viz., (a) the prioritization of ten village problems($r = +0.77$, $p < 0.02$), (b) prioritization of utilization of services of various health functionaries for treatment of and advice for children's illnesses ($r = + 0.75$, $p < 0.05$), and (c) prioritization of households using water from different sources($r = + 0.975$, $p < 0.02$). The method was also found to be more rapid (3.3 times) and less costly (6.3 times) compared to the traditional household survey method.

Conclusion: Rapid Community Health Assessment Methodology (RCHA) for prioritization of community health problems in a rural community is validated. The information thus obtained can be utilized for purposes of health policy and program planning, monitoring and evaluation. This is especially relevant for micro planning of child health services in developing countries. Repeated use of questionnaires for monitoring disease control programs must be carefully considered. Further studies to confirm and reconfirm the results of this study may be done before wider application of above methodology.

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Longitudinal assessments of habitual use of medicines in a nested case-control study of italian adults

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Aim: To assess the temporal changes of medicines consumption in relation to risk factors for respiratory symptoms/diseases in a subsample of a general population of Italian adults.

Methods: Within the Po Delta and Pisa epidemiological surveys, subjects who habitually used medicines at baseline were selected as cases. Controls were non-drug consuming subjects randomly drawn and matched for gender and age groups. 1663 subjects (89% of those invited) participated in a telephone interview (follow-up = 11 years) with questions on current drug consumption, respiratory health status, life habits, and comorbidity, beside socio-demographic characteristics. Main characteristics were: 37.2% males, mean age 63yrs, cases = 46.2%. We used logistic regression models accounting for smoking habit, gender, age, comorbidities, respiratory symptoms, occupational/passive smoking/environmental exposure.

Results: There was a non significant increase between baseline and follow-up for prevalence of asthma (5.8 vs 7.1%) or rhinitis (20.6 vs 23%). Prevalence rate of habitual use of any medicine at follow-up was 70.1 vs 46.2% at baseline ($p < 0.001$). Among the users (N=1166), we found higher prevalences of broncho-pulmonary (7.5 vs 4.8%) (significantly) and antiallergic drugs (2.4 vs 1.3%) (non significantly) at follow-up. New regular use of any medicines at follow-up was 60.4%, while persistence and cessation were 37.6 and 18.6%, respectively. Only asthma resulted significantly related to the use of medicines at baseline (OR 1.69, 95%CI 1.04–2.73), while at follow-up the relation was significant for both asthma (2.92, 1.51–5.66) and rhinitis (1.55, 1.03–2.33).). The association of the use of specific medicines (brochopulmonary/antiallergic/cardiovascular/diuretic vs other medicine/no use) was significant with asthma at baseline (3.36, 1.97–5.75) and at follow-up (3.94, 2.14–7.30). Non significant associations were found for positive skin prick tests with use of specific medicines both at baseline (1.19, 0.74–1.90) and follow-up (1.25, 0.78–2.00), and for elevated values of IgE with use of any medicines (1.36, 0.94–1.96, at baseline) and of specific drugs (1.16, 0.79–1.70 at baseline; 1.14, 0.78–1.66, at follow-up).

Conclusion: In this epidemiological study, ageing, asthma and rhinitis were determinant factors of medicines consumption.

URTICARIA & ANGIOEDEMA

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Effects of treatment of Helico bacter Pylori (HP) infection in idiopathic chronic urticaria

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Background: Urticaria is one of the most important cause of skin itching. The aim of this study was to examine effects of treatment of Helico bacter Pylori(HP) infection on urticaria in regard to the possible role of HP infection as a cause for idiopathic chronic urticaria(ICU).

Methods: 56 patients suffering from ICU (Male = 16 and Female = 40) with negative response to skin prick test for common aeroallergens were chosen. For diagnosis of HP serologic tests of antibodies against HP (IgG and IgA) through ELISA method and Ureas Breath Test (UBT) were done. In the basis of the test results, patients were put into three groups: case group (M = 8, F = 15) treated with antiHP drugs(Amoxicillin, Metronidazole, Omeperazole and Bismuth for two weeks), positive control group (M = 3, F = 12) without treated and negative control group (M = 5, F = 13) without HP infection. After one month UBT was done for control of the response to treatment.

Results: The results were analyzed through Chi-Square testing. In the case group 17 patients(74%) were significantly cured (23% partially and 52% compeletly) (P = 0.003). In the positive control group 6 patients (40%) partially treated and in the negative control group 40% of patients were cured.

Conclusion: Anti-HP treatment is significantly effective in treatment of ICU.

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Helico bacter Pylori (HP) infection and chronic urticaria

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Conclusion: Anti-HP treatment is significantly effective in treatment of ICU.

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Chronic idiopathic urticaria and brucellosis

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Introduction: Chronic urticaria/angio-oedema has traditionally been defined as daily or almost daily symptoms lasting for more than 6 weeks. Chronic urticaria affects 0.5–1% of individuals (life-time prevalence) and significantly reduces quality of life.

Optimal management of chronic and acute intermittent urticaria depends on a thorough understanding of clinical presentation, etiologies, triggers and aggravating factors.

An underlying extraneous cause for chronic urticaria cannot be identified in most patients, but infections may play a causative role in a few cases, and when present, chronic infections such as dental sepsis, sinusitis, urinary tract infections and cutaneous fungal infections should be treated. However exhaustive investigations searching for underlying infections are not indicated. Infection with Helicobacter pylori (HP) has been proposed as a possible cause, but the association is unlikely to be causal (particularly in otherwise asymptomatic children where the background prevalence of HP infection is high). Also complement activation can mediate or augment histamine release from mast cells via the anaphylatoxin C5a. This inflammatory pathway is triggered by antibody and antigen interacting to form immune complex-associated urticaria, e.g. in hepatitis C and hepatitis B, EBV, other viral and possibly parasitic infections. However common triggers for episodes of chronic urticaria are intercurrent viral infections and possibly stress.

Human brucellosis is an infectious disease produced by Brucella species: small, coccoid or rod-like, aerobic, Gram-negative bacteria. Every organ and system of the human body can be affected in brucellosis a fact that underscores the importance of including brucellosis in the differential diagnosis in areas of endemic disease, even if clinical features are not entirely compatible. The infection can also affect the skin.

We present a case of chronic urticaria who also had fever and chills at presentation for a week. His urticaria-like papules and plaques and a red, raised, itchy rashes especially in his face continue after his fever has stopped. In a vast array of laboratory investigation brucella serology, using both eliza and serum agglutination method, was positive. Only treatment of underlying infection has leaded to resolution of symptoms.

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Bronchial hyperresponsiveness in patients with chronic urticaria

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Background: Involvement respiratory system in patients with chronic urticaria is still a controversial subject. Large number of patients report dyspnea. The aim of this study was to investigate the present of bronchial hyper responsiveness in patients with chronic urticaria.

Methods: A total of 23 patients (17 women, 6 men, age range 19–56 years) who had urticaria for at least 6 weeks, without other known causes of urticaria, were enrolled in the study. Diagnostic programme of urticaria include: eosinophil count, skin prick test with inhalation and nutritive allergens, test for NSAID intolerance, pulmonary function tests and methacholine provocation. Patients are investigating during a phase of remission.

Results: Three patients had (13%) abnormal pulmonary function tests, ten patients (43,4%) showed significant bronchial hyperresponsiveness on methacholine provocation, three (13%) had elevated eosinophil count, four (17,3%) had positive skin prick test with inhalation and/or nutritive allergens and twelve (52,1%) had NSAID intolerance. Airway hyperresponsiveness was not associated with eosinophil count, skin prick test with inhalation and nutritive allergens, NSAID intolerance.

Conclusion: Significant number of patients with chronic urticaria show bronchial hyperresponsiveness, and performing of this test is reasonable in patients with chronic urticaria.

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An audit of referrals from public sector primary care physicians to an allergy clinic in Singapore

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Background: An electronic registry for all new referrals (New Case Registry) to our adult Allergy Clinic was set up in July 1998 to determine the sources of referrals, referring diagnoses and final diagnoses. Sources of referrals include primary care physicians in the public and private sectors, specialists, national dermatology centre, military services and self referrals.
Aim: To describe the pattern of referrals from primary care physicians in the public sector to an Allergy Clinic in Singapore.
Methods: All new referrals were prospectively captured in the New Case Registry during the study period 1 July 1998 to 31 December 2006.
Results: There were 227 referrals from public sector primary care physicians comprising 8.4% of all referrals. More than half (57.7%) were females and mean age was 36 ± 18 years. The majority (79.7%) were Chinese. The most common referring diagnoses were drug allergy [DA] (35.7%), urticaria and angioedema (23.3%) and food allergy [FA] (10.1%). In contrast, the final diagnosis of the attending allergist was DA in 9.7%, NSAID intolerance in 22.5%, and FA in 7.5% respectively. Among cases with primary care physician diagnosed DA, the allergist's diagnosis was DA in 23.5%, NSAID intolerance in 40.7% and not DA in 35.8%. Among cases with primary care physician diagnosed FA, the allergist's diagnosis was FA in only 31.6%. Chronic idiopathic urticaria comprised 19.1% of all cases of urticaria/angioedema. The following conditions were under-diagnosed by family physicians compared to allergists: anaphylaxis (1.8% versus 4%), asthma (2.6% versus 6.6%), atopic eczema (1.8% versus 6.6%), allergic rhinitis (7.5% versus 19.8%). Among all referrals for suspected allergy, 5.7% were deemed not to have any allergic disorder upon completion of evaluation.
Conclusion: DA, urticaria/angioedema and FA were the most common reasons for referral. NSAID intolerance accounted for the majority of cases of 'drug allergy' referred by primary care physicians. Allergic rhinitis, asthma, atopic eczema, and anaphylaxis were under-diagnosed, potentially leading to patients being deprived of appropriate therapy for allergic airway disease and life-saving epinephrine autoinjectors. Increasing the awareness and education may be useful in improving the diagnosis and treatment of allergic diseases in the community.

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Chronic urticaria: alternative treatment with beta-agonists

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Background: To describe a 46 year old patient with chronic urticaria, unresponsive to standard therapy, who responds to oral beta agonists and to propose possible mechanisms of action.
Methods: We performed basic bloodwork and checked eosinophil levels, ANA, ESR, CBC, CMP, anti-thyroid antibodies, TSH and complement levels. Skin biopsy was performed as well.
Results: Patient with history of diabetes mellitus presents for treatment of chronic urticaria x 15 years. Initially, patient responded to corticosteroids but since then no medications have helped, including Allegra, Doxepin, and Periactin. Test results revealed normal eosinophil levels, negative ANA, normal ESR, CBC, CMP, Anti-thyroid antibodies, TSH and complement levels. As patient was unresponsive to other medications and requiring frequent oral corticosteroids, albuterol sulfate 4 mg tablets were given. Patient took approximately 32 mg of albuterol orally each day for approximately 14 days in addition to her anti-histamine. On return visit, patient's symptoms had resolved completely.
Conclusion: Beta agonists have a good side effect profile compared to oral corticosteroids and may be of great benefit in reducing urticarial symptoms. Possible mechanisms of action include inhibition of anti-IgE induced histamine release thru desensitization of beta adrenoceptors. They may be a promising pharmacotherapy in the treatment of chronic urticaria.

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A study on the impact of chronic idiopathic urticaria on the quality of life in korean patients

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Background: Chronic idiopathic urticaria (CIU) is a relatively common disease, and yet not much is known about the causative factors or the pathomechanism, which makes it difficult to cure. Due to its chronic nature, many patients suffer from significant detrimental effects on their quality of life (QOL). The purpose of this study is to assess the impact of CIU on the QOL of Korean patients, and to determine whether a relationship exists between QOL and the severity of disease.
Methods: One hundred twenty four patients with CIU, who first visited our out-patient clinic from August 2005 to July 2007, were asked to complete a questionnaire designed to assess the effects of CIU on the daily lives of patients. QOL was divided into six different categories: mental status (MS), daily living activities (DLA), leisure activities (LA), self-perception (SP), treatment-induced restrictions (TIR), and social restrictions (SR). All of the QOL scores were recalibrated to a 0–100 scale, with 100 indicating the worst QOL, and 0 the most favorable. Pearson and Spearman correlation coefficients were used to analyze the relationship between the six different QOL categories, as well as the association between disease duration and QOL. One way ANOVA was used to analyze the relationship between disease severity and QOL. A p value less than 0.05 was considered significant.
Results: The average QOL scores obtained from the questionnaire were as follows: MS (50.64), DLA (47.58), LA (39.85), SP (37.06), TIR (35.54), and SR (33.23). The results showed that patients were most affected mentally, and that social restrictions were the least significant. The six different categories showed a positive correlation amongst themselves, which demonstrates that CIU exerts an influence on not one, but many aspects of QOL. LA and TIR were significantly affected in longer term CIU cases ($p < 0.05$), while SP, MS, DLA, and SR did not show a significant relationship with disease duration. Disease severity was found to have a significant effect on the SR, LA, and TIR categories ($p < 0.05$).
Conclusion: Based on these results, it is suggested that CIU has a negative impact on the QOL of Korean patients. Therefore, it is important to recognize the effects CIU can have on QOL and consider them as candidates for treatment as well.

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Neutrophil activation finding was noted in patients with ASA induced urticaria

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Background and Objective: ASA ingestion could induce acute and chronic urticaria, however their pathogenic mechanisms are not understood. We compared the level of neutorphil activation and related cytokine between ASA intolerant acute urticaria (AIAU) and ASA intolerant chronic urticaria (AICU).
Methods: The 88 patients with AICU, the 51 patients with AIAU and the 102 patients, normal control (NC) were enrolled. The clinical and laboratory findings including serum myeloperoxidase (MPO) level, IL-8 and IL-18 levels were compared among AIAU, AICU, and NC groups. The correlations between MPO and, IL-8, IL-18 were observed.
Results: The level of serum MPO were significantly higher in AIAU than in AICU and NC ($p=0.032$, $p=0.014$ respectively). The level of IL-18 was significantly higher in AIAU than in NC ($p=0.006$), while no significant difference was noted in IL-8 level. Within the AIAU patients, significant correlation were noted between MPO and IL-8 ($r=0.36$, $p=0.01$), not with IL-18.

Conclusion: Increased neutrophil activation finding in association with IL-8 was noted in AIAU patients, which may be involved in the pathogenic mechanism of AIAU.

Key words: ASA intolerant acute urticaria, neutrophil activation, IL-8

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Most cases of urticaria are caused by gastrointestinal disorders

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It is known that about 20% of people develop urticaria at least once in their lives. Chronic idiopathic urticaria affects up to 3% of the population. The incidence of acute urticaria is higher in people with atopy, and the incidence of chronic urticaria is not increased in people with atopy. This study shows that most cases of urticaria are related to such gastointestinal disorders as active gastritis, esophagitis and peptic ulcer. One of the most interesting things is that urticaria is likely to be only a single symptom of a severe and maybe life-threatening gastrointestinal disorder.

Methods: 10 patients with acute (3) or chronic (7) urticaria passed gastroscopy after 5–14 days of the onset of urticaria. In addition to urticaria, 3 had angioedema, 1 suffered from severe pruritus with desquamation and 1 developed palmar eczema.

Results: All patients showed to have different gastrointestinal problems, which correlated very well with the anamnesis data. 7 had gastric erosions, among which 3 with esophagitis; 1 had esophagitis with atrophic gastritis, 1 esophagitis alone, 1 patient with severe generalized urticaria had 2 big (1 cm each) duodenal ulcers. The patient with concomitant pruritus had lots of severe gastric, esophagal and duodenal erosions. Only 3 patients suffered from ocasional epigastric pain or heartburn, but none said that symptoms were severe. Gastrointestinal cause was suspected because episodes of urticaria developed or increased after irritating or acid food (spice, citruses, strawberries), drinks (alcohol, soda) or drugs (NSAIDs, acetyl-cysteinum, ascorbinic acid) but trigger factors were always different and dose-dependent. 9 patients that underwent anti-reflux, antacid, antisecretory and some (2) antibiotic therapy along with symptomatic antihistamine drugs recovered (8) or showed much relief (1). 1 patient with severe pruritus didn't pass prescribed treatment and chose his usual intramuscular injection of diprospan instead. As always, he reported a short 2-week relief of symptoms.

Conclusion: These data show that urticaria alone or with angioedema very often occurs as a symptom of a gastrointestinal disorder. It is very important to prescribe gastroscopy and adequate treatment not only to stop urticaria, but moreover to prevent gastrointestinal bleeding or perforation, very genlty operating with steroids, especially oral, in such patients.

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Etiology and prognosis of acute urticaria in a university dermatology clinic in China: a follow-up study

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Background: Etiology and prognosis of acute urticaria has not been reported in China.

Objective: To investigate the etiology and outcome of acute urticaria in a university dermatology setting.

Methods: A prospective follow up study was performed. All patients with acute urticaria seen by authors between March 2005 and July 2006 in department of dermatology, Peking University Third Hospital were asked to participate. Etiologic agents and triggers were evaluated by comprehensive and detailed history and physical examination, supplemented with selected laboratory testing and follow up. Prognosis was assessed at 9 months after the first visiting. Cure was defined as complete free of symptoms for more than 6 months without relapse after cessation of the treatment.

Results: 75 patients participated. There were 22 male, 53 female, with age ranged 1.5 to 83 years. Of those, 60% were between 18 to 40 years; 32% had a history of acute urticaria during childhood; and 6.5% were atopic. Etiologic agents or triggers could be identified in 54.7% of the patients including medications (21.4%), infection (20%), food (8%), physical stimuli (2.7%), insect bite (1.3%) and contactant (1.3%). In 71 patients finished the follow up, 58 cases (81.7%) cured. 13 patients, all etiology and triggers unidentified, progressed to chronic urticaria, but 6 of them free of symptoms at the end of evaluation. The overall cure rate in patients with disease duration less than one week was 94%, which was much higher than that of patients with a longer duration (41.2%, $p < 0.05$, chi-square test). Sex, age and severity of the disease had no effect on the prognosis.

Conclusion: Etiology and triggers can be identified in more than half of acute urticaria patients. Medicaments, infection and food are common triggers. The outcome of acute urticaria is favorable. The longer disease duration (over a week) is an important risk for poor prognosis.

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Angioedema in a department of medicine. IgE-mediated mechanism the most suspected cause

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Background: In angioedema deep dermal and subcutaneous tissue is affected.Even mucus membranes are involved and may be part of life-threatening anaphylactic reactions. These patients are often seen in departments of medicine.

Materials: 123 patients with angioedema remitted to the department of medicine for evaluation and treatment during the period August 2002 – May 2007. Mean age 45, range 17–86 years. F/M ratio 2.3. 54%(64) had urticaria as well. Method. Pathogenetic mechanisms and the cause were evaluated. IgE-mediated allergy, aspirin-induced allergic-like reactions, direct drug induced adverse reactions and c1-esterase inhibitor deficiency were considered.

Results: Among 152 patients remitted for evaluation of allergy to penicillins, 47(31%) had angioedema, but in only 39 (26%) penicillin was not excluded as the cause (exclusion by controlled administration of penicillins and s-tryptase). Aspirin (11), nonsteroidal antiinflammatory drugs (6), ACE inhibitors (10) and other drugs (10) were seen as the cause mediated by leukotrienes, bradykinin and direct mast cell degranulation. Food was suspected in 28. Insect sting the cause in 16/40 (honeybee 1/9 and wasp 15/31) and occupational agents in 5/43: latex (nurse), fish protein, flour, dyes, nikel. In only one the c1-esterase inhibitor was slight reduced. Angioedema was associated to allergic rhinoconjunctivitis and asthma. Infections and physical stimuli were well known inducers in rather few. Most patients with non-drug induced angioedema had prophylactic anaphylactic medication with corticosteroid and antihistamines at home. 48 had epinephrine (epipen, autoinjector).

Conclusion: Urticaria and angioedema are the most frequent manifestations of severe allergic reactions (anaphylaxis). Many different causes may induce angioedema. However, IgE-mediated mechanism is the most frequent suspected cause of angioedema in a department of medicine.

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Successful treatment of chronic urticaria with low molecular weight heparin

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Background: Autologous serum skin test (ASST) has been used to detect circulating vasoactive and histamine-releasing factors in patients with chronic urticaria (CU). Recently, it has been observed that autologous plasma skin test (APST) scores positive more frequently than ASST in CU patients, and its positivity is associated with the activation of coagulation cascade.
Aim: We report a case of chronic urticaria successfully treated with anti-coagulant therapy using low molecular weight heparin (LMWH) to see whether symptom control is associated with reduction of coagulation activation.
Methods: A 63-year-old woman, with a 3-year history of unremitting CU, was evaluated in our outpatient allergy clinic. No other diseases were present in her clinical history. CU improved only with prednisolone 0.5 mg/kg/day, plus anti-H1 antihistamine, while therapy tapering led to symptom recurrence. Known causes of CU were ruled out with laboratory and instrumental examinations. ASST, APST, negative control with saline solution and positive control with histamine were performed after a 5-day discontinuation of steroid and anti-H1 therapy, and repeated after 15 days of LMWH therapy. Prothrombin fragment F1 + 2, marker of coagulation activation, and D-dimer, marker of fibrinolysis, were measured by ELISA before starting LMWH therapy and 15 days later.
Results: At baseline, ASPT gave an unequivocal positive response (20 mm mean wheal diameter), whereas ASST was negative; prothrombin fragment F1 + 2 value was 833 pmol/l (normal range 69-229 pmol/l) and D-dimer value was 27.15 nmol/l (normal range 0.50–4.00 nmol/l) confirming an activation of the coagulation cascade and fibrinolysis. LMWH therapy (enoxaparin 4000 UI twice a day sc) was started. After few days CU improved and the patient stopped anti-H1 and steroid therapy, without recurrence of CU. Fifteen days later the patient was still asymptomatic, while receiving only LMWH therapy. APST became negative and we observed a marked reduction of both prothrombin fragment F1+2 (287 pmol/l) and D-dimer (2.84 nmol/l).
Conclusion: The activation of coagulation in this patient suffering from CU can be counteracted by anticoagulant treatment, leading to symptom control. Properly designed studies are needed to evaluate this new therapeutic approach.

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Angioedema associated with the use of Angiotensin Converting Enzyme: clinical features and insights into pathogenetic mechanisms

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Background: Angiotensin converting enzyme inhibitors (ACE-Is) are known cause of angioedema. Its incidence is between 0.1% and and 2.2%, but its clinical relevance is due to risk of life-threatening involvement of upper respiratory tract. In the present study we reported the clinical features of patients complaining of ACE-Is-angioedema referred to our Unit in a 5 year period. In a subgroup of them we studied the potential mast-cell involvement by measuring serum tryptase.
Methods: Descriptive, observational on ACE-Is-induced angioedema during a 5-year period (January 2001– December 2006). Serum tryptase level was evaluated by ELISA assay during the angioedema attacks, and in baseline conditions in three different groups: Group 1 = 12 patients complaining of previous ACE-Is angioedema; Group 2 = 15 patients with previous non- ACE-Is angioedema; Group 3 = 10 patients treated with ACE-Is without angioedema.
Results: We observed 32 patients (19 male; mean age 62.3 yrs, range 42 – 88 yrs) with ascertained ACE-I-induced angioedema. Only one patient was African, whereas the other were white. Patients reported an average of 3.8 attacks of angioedema (range 1– 9). Upper respiratory tract involvement was observed in 2 subjects. Eleven patients reported a concomitant use of NSAIDs or aspirin (five at low dosage). One patient reported previous cough due to ACE-Is. Drugs responsable were: lisinopril (7), fosinopril (7), enalapril (6), quinalapril (6), rampril (4), captopril(2).

The period of time between the beginning of ACE-Is therapy and the first angioedema attack ranged from 10 days to 4 years (mean 1.4 years). Most of patients (72%) had an isolated angioedema (lips 53%, face 21%, tongue 9%). Only two patients were admitted to the medical intensive care unit and one of them was intubated. Follow-up information were available only for 16 subjects and 12 of them had no relapse after stopping ACE-Is treatment. During the attacks only one patient showed a slight increase of serum tryptase (12 ng/mL), whereas no significant change was observed in baseline condition in the three groups (Group A : 5.7 ng/mL; Group B : 4.9 ng/mL; Group C : 3.9 ng/mL of serum tryptase).
Conclusion: ACE-Is are a relevant cause of angioedema mainly in adult and elderly population. Physicians should be aware of the potential risk of upper airway involvement. Mast-cells seem not to be involved in the pathogenesis.
Keywords: Angiotensin converting enzyme inhibitors, angioedema, tryptase.

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Icatibant –an upcoming subcutaneous treatment for acute attacks in hereditary angioedema

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Rationale: Hereditary angioedema (HAE) is characterised by skin swellings in various parts of the body, laryngeal edema or abdominal pain. Besides the deficiency or malfunction of plasma C1-inhibitor as cause of HAE, bradykinin is considered to be a key mediator of symptoms in HAE attacks. A new treatment for acute HAE attacks might be Icatibant, a synthetic and highly selective bradykin-B2-receptor antagonist which can be administered sub-cutaneously. We report on the effect of Icatibant in treating an acute attack in one patient in our hospital.
Methods: In the open-label extension phase of a phase III, double blind, randomized multicenter trial (FAST-2) which compared Icatibant versus tranexamic acid, patients with abdominal or cutaneous attacks were treated with Icatibant 30 mg sc. Symptom score by patient, documentation of time for the onset of symptom relief measured by a visual analog scale and time to complete resolution of symptoms were the main assessments.
Results: The patient treated in our hospital was a 22 year old, otherwise healthy male with known HAE since the age of 6 month. He has been having frequent attacks about twice per month which usually were treated with tranexamic acid. The patient experienced in total 11 attacks (7 abdominal, 2 cutaneous and 2 laryngeal) with an average duration of 4 days during the last 6 months. The patient came to the clinic with a severe cutaneous edema of his genitals. He received one single injection of 30 mg Icatibant sc. The onset of symptom relief was reached rapidly after approximately 10 minutes. Complete resolution of symptoms occurred within 5 hours, so the patient could leave the hospital. Icatibant was well tolerated, only mild injection site reactions such as pruritus were observed. No systemic adverse events were observed.
Conclusion: In the present case Icatibant provided fast and complete resolution of severe cutaneous HAE symptoms and was well tolerated. The treatment course of the HAE attack in our case complies with the results of the double blind phase of the FAST-2 trial. The time to first improvement was 0.8 hours for Icatibant vs. 7.9 hours for tranexamic acid ($p < 0.001$).

The primary endpoint (median time to onset of symptom relief) was met with 2.0 hours for Icatibant compared with 12.0 hours for tranexamic acid ($p < 0,001$). The end of attack (almost complete symptom relief) was reached after 10 hours during treatment with Icatibant compared to 51 hours for tranexamic acid ($p < 0.001$).

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Evaluation of the receptor occupancy by desloratadine and levocetirizine in allergic subjects

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Background: The inhibitory activity of the new generation H1antihistamines levocetirizine (L) and desloratadine (D) on allergen-induced wheal & flare reaction at different times after administration of therapeutic doses has been evaluated in a previous study (Frossard et al. Br J Clin Pharm 2007). L suppressed the cutaneous allergic reactions with a higher potency than D. The receptor occupancy (RO) also evaluated for both drugs 24 h post-dose was higher for L than for D. The purpose of the present study was to further analyze the RO for both drugs at 12 h and 24 h after administration.

Methods: During a double-blind, randomized, single dose, cross-over, placebo-controlled study in 18 allergic subjects (9 males, 9 females, all Caucasians, age 18–48 yrs, weight 54– 93 kg), L and D plasma levels were measured at 12 and 24 h post-dose. RO was calculated as RO % = ROmax x Cfree¨M (Cfree + Ki), where ROmax = maximal % of binding sites (set to 100%), Cfree = free concentration of drug at the histamine H1 receptor, and Ki = equilibrium inhibition constant. Free concentration of the drug at the histamine H1 receptor was considered as the free plasma concentration since only the unbound drug is capable of entering and leaving the plasma and tissue compartments (assuming the absence of active transport mechanisms, at least in the target tissues containing histamine H1 receptors). As the Ki may vary with pH (Gillard & Chatelain Eur J Pharmacol 2006) and acidosis appears to be a hallmark of inflammatory processes [e.g. the exhaled breath condensate pH in children (Profita et al. JACI 2006) and adults with allergic rhinitis is acidic compared with healthy controls], RO was also calculated using the Ki values obtained in acidic conditions. The statistical analysis was performed using the Student-t-test for paired data.

Results:

Parameter	Desloratadine (5 mg)	Levocetirizine (5 mg)	Difference (95% CI) (* = statistically significant, p < 0.001)
Plasma elimination half life t½ (h)	27	8	
Plasma protein binding (%)	85	91	
Ki (nM)			
Ki (t 37°C, pH 7.4)	0.4	3	
Ki (t 37°C, pH 5.8)	0.63	1.3	
Free drug conc. (nM) (mean ± SD)			
C 12 h	0.394 ± 0.118	11.3 ± 2.61	
C 24 h	0.215 ± 0.075	3.90 ± 1.59	
Receptor occupancy (%) for Ki at pH 7.4 (mean ± SD)			
RO 12 h	48.6 ± 7.6	78.3 ± 4.4	29.8 (25.7–33.8)*
RO 24 h	34.0 ± 7.7	54.1 ± 11.7	20.1 (14.3–25.9)*
Receptor occupancy (%) for Ki at pH 5.8 (mean ± SD)			
RO 12 h	37.7 ± 7.0	89.2 ± 2.6	51.5 (48.0–55.0)*
RO 24 h	24.9 ± 6.4	72.2 ± 10.2	47.4 (42.3–52.4)*

When evaluated at pH 7.4, a statistically significant difference of 30 and 20 between the percentages of RO by L and D was obtained at 12 and 24 h post-dose, respectively. This difference reached 52 and 47 when RO computation was performed using the Ki values obtained at pH 5.8 (see table).

Conclusion: In spite of the higher affinity (Ki) and longer t½ of D compared with L, the higher free plasma concentrations of L at 12 and 24 h post-dose produced higher percentages of RO probably accounting for the statistically significant higher inhibition of allergen-induced wheal & flare induced by L.

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The impact of hereditary C1-inhibitor deficiency on the development of atherosclerosis

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Background: Hereditary angioedema (HAE) is a rare disease characterized by recurrent swellings of the subcutaneous or submucosal tissues caused by the inherited deficiency of the C1-inhibitor (C1-INH) –a main regulator of the complement system. Atherosclerosis is an inflammatory disease and the complement system plays a major role in the development of the plaques. The aim of the present study was to investigate the possible impact of C1-INH deficiency on atherosclerosis.

Methods: In a cross-sectional prospective study we included 57 adult patients and 20 age and sex matched healthy individuals. The prevalence of cardio- and cerebrovascular atherosclerotic diseases as well as distinct atherosclerotic risk profiles were determined. In addition, the thickness of the intima-media (IMT) of the common carotid artery was also determined. As severe HAE patients receive long-term danazol prophylaxis –an attenuated androgen-, patients were divided into two subgroups according to prophylactic treatment.

Results: Low prevalence of atherosclerosis was found in HAE patients: only 2 subjects were affected (1-1 in the danazol treated and not treated group, respectively) according to the medical history. Patients with long-term danazol prophylaxis had an increased risk profile for atherosclerosis: significantly higher body-mass index, LDL/HDL ratio, creatine kinase activity, creatinine, GPT and hemoglobin levels were found compared to the other HAE patients and healthy controls as well. However carotid IMT did not differ relevantly in study subgroups.

Conclusion: Long-term danazol use results in a highly proatherogenic risk profile in patients with HAE. However, it seems that it does not lead to early atherosclerosis. The prevalence of cardio- or cerebrovascular diseases is low in HAE patients irrespectively to their prophylactic treatment and carotid IMT is comparable to healthy controls. We hypothesize, that the functional deficiency of C1-INH might be a protective factor in these patients against atherosclerosis – which outlines the importance of the plasma enzyme systems in the process which are regulated by C1-INH.

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Patient perception of levocetirizine in urticaria: a multicenter study in Taiwan

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Background: Levocetirizine (Levo) is a potent latest-generation, non-sedating oral H1-antihistamine for which no data in Taiwanese population has yet been published.

Objective: Primary: patients' perception of Levo in the treatment of urticaria. Secondary: adverse events (AEs), onset of action, and improvement in quality of sleep and daily activities.

Methods: A multicenter observational study (6 medical centers) was conducted from May 2006 to March 2007 in Taiwan assessing the treatment perception of urticaria patients on Levo. Pruritus severity (0=absent, 3=severe) and duration (mild < 1h; moderate 1–6h; severe > 6h), number (mild:1–10; moderate: 10–20; severe: > 20), size (mild = 1.5cm; moderate: >1.5cm <3cm; severe: > 3cm) and duration (mild < 1h; moderate 1–6h; severe >6h) of wheals and the frequency of symptoms (mild30min, <1hrs), and moderate (>1hrs). A visual analogue scale (VAS) was used to evaluate global satisfaction of patients and physicians (range: 0–10).

Results: 97 patients were included and 95 completed the study. 2 patients (2%) were lost to follow-up due to AEs, none serious. 67 patients (69%) had acute

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Symptom improvements	Pruritus severity (n = 95)	Duration of pruritus (n = 97)	Number of wheals (n = 96)	Duration of wheals (n = 97)	Size of wheals (n = 97)	Frequency of symptoms (n = 97)
Complete or partial relief	80%	64%	73%	66%	60%	72%
Unchanged	19%	36%	26%	32%	37%	27%
Worse	1%	0%	1%	2%	3%	1%

Overall efficacy and tolerability were assessed as "good/excellent" by 75% and 68% of patients, respectively. At least 60% of all patients and 75% of those with moderate/severe symptoms reported complete recovery or marked improvement of any individual allergic symptom. 65% of subjects reported the onset of action as very rapid or rapid. Levo was reported as better than their previous therapy by 72% of patients. Good to excellent improvement in quality of sleep and daily activities was reported by 61% and 74% of patients without concomitant medications, respectively. Global satisfaction with Levo was high for both physicians and patients (VAS=7.4). Most frequently reported AEs were somnolence (7%) and fatigue (3%).

and 30 (31%) chronic urticaria. The use of concomitant antihistamines was reduced from 58% (baseline) to 27% at end of treatment.

Conclusion: Results confirm western studies that levocetirizine potent, effective and well tolerated also in Taiwanese patients with urticaria. Most of the patients considered it as better than their previous treatments.

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Levocetirizine has a better overall efficacy than desloratadine in chronic idiopathic Urticaria: A double-blind, randomized, clinical trial

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Background: Levocetirizine (Levo) and desloratadine (Deslo) are H1-antihistamines of the latest generation. Levo has been shown to be significantly more potent than Deslo in either histamine or allergen-induced wheal and flare studies. So far, no head-to-head clinical studies in Chronic Idiopathic Urticaria (CIU) have corroborated these findings.

Methods: A multicenter, double-blind, 2-parallel groups (438 adults on Levo, 448 on Deslo), randomised trial over 4 weeks (w) of treatment to compare the clinical efficacy and safety of Levo 5mg vs. Deslo 5mg, both once daily. Primary endpoint: pruritus severity score (0 = absent to 3 = severe) assessed daily over the first week (w1) of treatment. Secondary endpoints: pruritus severity over all 4 study weeks (w4); pruritus duration, number and size of wheals over w1 and w4 (all on a 0–3 scale); global satisfaction with treatment (0 = very dissatisfied to 10 = very satisfied), and safety. CIU composite score, representing the drug's overall efficacy, was also assessed.

Results:

Conclusion: In this first head-to-head comparison of Levo and Deslo in CIU, patients satisfaction with Levo was significantly higher, onset of action of Levo was faster and the overall efficacy of Levo was significantly better than Deslo in relieving CIU symptoms.

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Urticaria and angio-oedema due to the daily 80 mg dose of acetylsalicylic acid

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Background: Low dose acetylsalicylic acid have been widely prescribed to prevent or reduce the risk of strokes and heart attacks. There are many reports concerning its known side effects, such as: acute urticaria. However, reports of intermittent urticaria and angio-oedema due to the daily 80 mg intake of acetylsalicylic acid are not common.

Methods: We have seen 4 patients (56y old man, 64y old man, 77y old woman and a 79y old woman) who used a daily dose of 80 mg acetylsalicylic acid for more than 6 weeks. They complained about multiple episodes of swelling of the tongue, cheeks and urticaria. The frequency varied from once a week or once in 2–4 weeks, within a period of 6 to 12 months. Some patients consulted a dermatologist and another physician without any results. Routine screening investigations were negative.

Results: The symptoms disappeared in 3–6 weeks after they stop taking acetylsalicylic acid and taking clopidogrel instead.

Urticaria and angio-oedema will reappear within 3–5 weeks, after the patients continue taking the 80mg daily dose of acetylsalicylic acid, instead of the replacement clopidogrel.

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		Baseline Mean (SD)	Over week 1, Adj. Mean (SE)	p value	Over 4 weeks, Adj. Mean (SE)	p value
CIU Composite score (SD)	Levo	4.03 (1.07)	1.98 (0.08)	0.005	1.71 (0.07)	0.041
	Deslo	4.07 (1.05)	2.23 (0.08)		1.88 (0.07)	
Pruritus severity score (SD)	Levo	2.20 (0.53)	1.02 (0.04)	<0.001	0.86 (0.04)	0.004
	Deslo	2.22 (0.53)	1.18 (0.04)		0.99 (0.04)	
Pruritus duration (SD)	Levo	2.11 (0.63)	1.08 (0.04)	0.002	0.93 (0.04)	0.009
	Deslo	2.13 (0.64)	1.24 (0.04)		1.05 (0.04)	
No. of wheals (SD)	Levo	1.83 (0.66)	0.96 (0.04)	0.054	0.85 (0.04)	0.353
	Deslo	1.85 (0.65)	1.05 (0.04)		0.89 (0.04)	
Size of wheals (SD)	Levo	1.94 (073)	1.01 (0.04)	0.025	0.89 (0.04)	0.085
	Deslo	1.93 (0.71)	1.12 (0.04)		0.97 (0.04)	

The improvements in the mean scores were: pruritus severity 54% and 61% (w1, w4) for Levo and 47% and 55% (w1, w4) for Deslo; pruritus duration 49% and 56% (w1, w4) for Levo and 42% and 51% (w1, w4) for Deslo. The patient satisfaction with treatment was significantly higher in the Levo (7.62) compared to the Deslo (7.33) group (p=0.021) over w4. The time to first symptom improvement reported by the 1st quartile of subjects was shorter with Levo (3h00) compared to Deslo (4h15). Drug-related adverse events (DRAEs): 13.7% for Levo and 11.2% for Deslo, without any serious DRAEs.

Conclusion: We conclude that the intake of low dose acetylsalicylic acid may have caused urticaria and angio-oedema in these patients. The reason behind the attack of urticaria and angio-oedema that only happened in once a week or once in the 2–4 weeks is still unclear. The intermittent nature of urticaria and angio-oedema attacks may also be the reason why many physicians are not aware that the daily 80 mg dose of acetylsalicycic acid may cause of this problem.

We suggest replacing the daily 80 mg use of acetylsalicylic acid with clopidogrel, for those patients with urticaria and angio-oedema possibly caused by acetylsalicylic acid.

Further studies are needed to investigate the mechanism of this disorder.

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Distribution of HLA DRB1 specifities in patients with chronic urticaria

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Background: In 50–80% of patients with chronic urticaria (CU) the origin of disease is unknown. In most of CU cases autoimmune mechanisms are involved. In CU patients auto antibodies such as aDNA, p-ANCA, ANF, RF often detected along with the clinical manifestations of autoimmune pathology (arthralgia, myalgia, photodermatitis, sub febrile body temperature). Predisposition to autoimmune diseases is connected to certain variants of HLA genes. Relative risk index ≥2 demonstrates that individuals bearing this HLA gene variant have higher risk of autoimmune pathology development than other individuals. The goal of the study was to investigate association between certain HLA specifities and CU development.

Methods: In our study patients were divided into 3 groups. First group - patients with acute atopic urticaria (N = 30), second group - patients with CU with clinical manifestations of autoimmune pathology (N = 31), third group - normal individuals (N = 135). HLA-genotyping was performed with SSP-variant of PCR.

Results: In the first group half of chronic CU cases revealed from childhood. Weals persisted less than 6 hours in all cases, atopic background in 37% of cases, 100% efficacy of AH drugs application. In the second group: urticaria debut at the age of 24 to 51,urticaria persistence for 2–4 years in average, existence of autoimmune pathology manifestation in reumo-tests. In the second group statistically significant increase of autoantibodies was found in comparison to the first group. According to HLA-genotyping data patients with CU demonstrated statistically significant increase in the level of HLA DRB1*04 in comparison to normal individuals.

Conclusion: We demonstrates that relative risk in patients with CU for DRB1*04 specifity is 2,33. Data obtained points out possible autoimmune mechanism of urticaria development.

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Desloratadine reduces symptoms and improves quality of life in patients with chronic idiopathic urticaria: a multicenter, practice-based study in China

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Background: Chronic idiopathic urticaria (CIU) is associated with a heavy symptom burden, which translates into significant impairment in quality of life (QOL). Non-sedating antihistamines are first-line therapy for CIU. Desloratadine (DL), a non-sedating, selective H1 receptor antagonist has been shown in multiple placebo-controlled studies in Europe and the United States to be effective in reducing the severity of pruritus and wheals in CIU, while improving QOL measures. The impact of DL on CIU has not been studied in a Chinese population.

Methods: This open-label, multicenter study evaluated the efficacy and safety of DL 5mg QD administered for 28 days in patients with CIU treated during everyday practice in China. Patients aged ≥18 yr and of either gender, with a ≥6 week history of CIU were eligible. Patients had to have wheals present for ≥3 days/week, and pruritus, wheals and overall CIU severity scores had to be ≥2 (0=none to 3= severe). At baseline and days 8, 15, and 29 patients rated the severity of pruritus, wheal size/number, total CIU symptoms, interference with sleep and daily activities and overall CIU condition using a 4-point scale. QOL was assessed at the same time-points using the Dermatology Life Quality Index (DLQI). At day 29 the global response to therapy was rated (1=complete relief to 5= treatment failure); adverse events (AE) that occurred during the trial were collected and rated for severity/relation to study drug.

Results: The study included 460 subjects (54.1% female), with a mean age of 37.2 years and a mean duration of diagnosed CIU of 25.7 months. Pruritus, wheal size, wheal number, total CIU score, interference with sleep and daily activities and overall condition of CIU were all significantly improved from baseline at days 8, 15 and 29 ($P < 0.0001$ for all). DLQI also improved significantly from baseline over the course of the study ($P < 0.0001$). The percentage of patients with complete or marked relief of CIU rose from 64.8% at day 8 to 80.3% at day 15 and 82.3% at day 29. DL was well tolerated and no serious AEs were observed. The most frequent AEs rated as possibly/probably related to treatment were drowsiness (3.7%), fatigue (1.7%), headache (1.5%), thirst (1.5%) and dizziness (1.3%).

Conclusion: This practice-based study in Chinese subjects confirms that DL is effective and well tolerated in the treatment of CIU and is associated with improved QOL.

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Fresh frozen plasma for the treatment of acute hereditary angioedema

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Background: Chronic prophylactic therapies for hereditary angioedema (HAE) have existed for some time but in the United States acute therapies are lacking. Fresh frozen plasma (FFP) has been used to treat acute attacks but its use is controversial because reports of FFP worsening HAE symptoms exist.

Methods: Twenty-three patients with HAE answered survey questions regarding past experiences with FFP. The purpose of the survey was to determine whether FFP administration during acute HAE attacks ever lead to a worsening of symptoms. Additionally, patients were asked whether FFP was perceived to be beneficial. Demographic data and data on the types of attacks treated was also collected.

Results: Six of 23 patients received FFP for acute attacks and an additional three patients received FFP for surgical or dental prophylaxis. Greater than 71 acute attacks were treated with FFP. One individual accounted for approximately 50 treatments. Attack locations included abdominal, peripheral, facial, genital, and laryngeal. There were no episodes of acute worsening after administration of FFP or spread to the larynx. There was perceived benefit from FFP in all but five instances.

Conclusion: Until more specific therapies for the acute treatment of HAE are widely available, FFP should be considered as a treatment option particularly in non-laryngeal attacks.

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Hereditary angioedema prognosis in non treated Algerian patients

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Background: Hereditary angioedema (HAE) is an autosomal dominant disease that is characterized by quantitative or qualitative deficiency of a plasma protein called C1 esterase inhibitor(C1INH). The disease is manifested by episodic attacks of nonpitting, nonpruritic, localized oedema that progresses rapidly without urticaria or erythema. Swelling of the intestine can cause intense abdominal cramping associated with vomiting and diarrhoea. Laryngeal oedema may prove fatal.

The main of the present retrospective study is to report the follow-up care of HAE non treated symptomatic patients.

Patients and Methods: For each patient clinical score was established and complement system investigation including antigenic quantification of C1 INH, C3, C4 and C1q by nephelometry (Dade Behring, Germany). And determination of C1 inhibitor function by chromogenic assay (C1 – INH, IMMUNO AG, Vienna, Austria) were performed.

Results: The two most striking matters of HAE spontaneous course in our patients comparing to literature data before androgenic therapy are, firstly a very low mortality rate (about 5% versus 30%) and secondly, only few of them (less than 10%) had developed moderate attack following dental care even without preventive drug.

Conclusion: Our population seems to be more resistant to C1 inhibitor deficiency than European and American patients. We suppose that treated patients would become more sensitive to minor disorders than non treated ones, and also this difference could be due to environmental factor (s) such as microbes.

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Effect of cetirinax on the skin reactivity in patients with chronic urticaria

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Background: Cetirinax® (Actavis), a derivate of piperazine, belongs to the family of the second-generation antihistamines. It is a fundamental pharmacological active metabolic product of hydrazine – a first-generation antihistamine. Beside its antihistamine activity, Cetirinax® posseses also other varied qualities that augment its antiallergic properties. Suppression of erythema and papula is traditional pharmacodinamical mechanism used to determine activity of H1 receptor antagonists.

The purpose of that survey is to get in touch with suppressive effect of Cetirinax® on erythema and papula caused by histamine in patient with chronic urticaria.

The aims that we put were to survey suprressive effect according to time, clinical condition of patients, sex and age.

Methods: The study involved twenty patients with chronic urticaria, 18–63 years of age. Histamine sensitivity of all patients was given by prick-test with 10 mg/ml histamine 20th minutes before treatment with 10mg Cetirinax®. After that the same sensibility was determined on 30th minutes, 1st, 3rd, 6th, 9th, 12th and 24th hour.

Results: The suppressive effect of Cetirinax® set in 30th minutes and is highest marked in 3rd hour on the erythema and in 9th hour on the papula. The difference were statistic important compared to basic size of papula and erythema. Depression of erythema was equal like papula in various hours. Depressive effect was marked clearly in 24th hour.We determined that histamine papula and erythema were similar at patients independently ages, sex, and clinic condition. The clinic effect correlates to the excellent suppressive functions.

Conclusion: Cetirinax® is a medicine with excellent quality on the skin reactivity.

EPIDEMIOLOGY - RISK FACTORS

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Prevalence of bronchial asthma in children in Saudi Arabia

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Background: The impact of childhood asthma on the health care system is considerable. As one of the most chronic diseases of children, asthma is frequent caused for emergency room visits and hospitals admissions. Clinical observations indicate that atopic diseases, particularly bronchial asthma and allergic rhinitis are also common in Saudi Arabia. However, there were no formal studies until recently on the prevalence and etiology of childhood asthma in the country.

Methods: Prevalence of Bronchial Asthma in school children were studied in different part of the country using a standardized protocol of 32 questionnaires. The questionnaire was designed by a committee of international expert and first

used in 1985–1986, before ISAAC launched its first phase of limited age prevalence studies. Our questionnaires, however is very similar to ISAAC questionnaires but include children from 06 to 16 years of age. For comparative purposes, we have continued using the same protocol for other parts of the country. The questionnaires were translated in local language and distributed through the Ministry of Primary Education to different urban schools of the Kingdom.

Results: Their studies were completed at different intervals within the past several years. The combined data revealed varying prevalence of asthma with highest 24% being in a coastal city bordering Yemen called Gizan (n = 362) followed by Taif 23% (n = 594) and Hail 22% (n = 507) an agricultural city. The prevalence rate of asthma for other places where Al-Gazim 16% (n = 384), Abha 13% (n = 485), Dammam 12% (n = 889), Hofuf 14% (n = 923), Jeddah 12% (n = 531) and Riyadh 10% (n = 988). The prevalence for allergic rhinitis was much higher than asthma.

Conclusion: Bronchial Asthma emerged to be one of the prevalent diseases in Saudi Arabia and showed regional diversity. The socio-economic differences between asthmatic and non asthmatic children were none striking. Environmental and geographical influences were considered to be the reasons of the variations in the rate of prevalence.

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Hygienic hypothesis and asthma in early adolescence

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Aim: A negative association between the number of siblings and older siblings especially with allergic diseases symptoms through the impact of infections on Th1/Th2 immune response is suggested. The study was aimed to examine the influence of the presence of siblings and the presence of older siblings on asthma in young adolescents.

Materials and Methods: The self-reported data by 5507 adolescents aged 12/16 years from 8 cities in The Republic of Macedonia obtained through the ISAAC phase 3 questionnaires on asthma and environmental risk factors in 2006 were analysed. The presence of siblings and older siblings separately (adjusted for sex, passive smoke exposure at home, gas/wood cooking at home, wood/coal/oil heating at home, duration of TV watching daily, cat ownership) were correlated to wheeze ever, current wheeze, current speech-limiting wheeze, exercise-induced wheeze and ever-diagnosed asthma. Odds ratios (OR, 95% CI) in binary logistic regression were performed for statistical analysis of data.

Results: A significant association of the presence of siblings in general and the presence of older siblings especially with asthma and its symptoms was not found ($p > 0.05$ for all the parameters).

Conclusion: It seems that the presence of siblings and older siblings do not influence asthma in young adolescents.

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Environmental tobacco smoke: a risk for bronchial hyperreactivity and pediatric asthma

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Background: Fifty passively smoking chiidren of whom at least one parent was a chronic smoker were included in the present study their ages ranged between 5–15 years old.Exposure to cigrette smoke was recognized from personal history and was confirmed by measurement of urinary cotinine(the major metabolite of nicotine in urine).

Aim: To evaluate the effect of household environmental tabacco smoke on bronchial reactivity and subsequent lung functions in passively smoking children. We also aimed to correlate pulmonary functions in passive smokers children with the number of smoked cigarettes of thier parents as well as the duration of thier exposure to cigrarettes smoke. Urinary cotinin was studied as a mirror image to the number of smoked cigarettes. Its level was also correlated to bronchial hyperreactivity and lung functions in the studied children.

Methods: To asses bronical hyperreactivity,metacholine challenge test(MCH) was done by using dosimeter, Pulmonary functions test were performed before and after metacholine challenge test. Challange test was stopped when there is a reduction in forced expiratory volume in 1st second by 20% from base line,or when the maximum concentration of metacholine was reached (25mg/ml). pulmonary function tests was performed by Med Graphics TM SPIROMETERY 1070 series 2,hard disc computer system. Urinary cotinin was measured by high -performance liquied tomography.

Results: 68%(34 children) were found to have bronchial hyperreactivity,14 children out of the 34 did not perform challange test as they had basel bronchial hyperreactivity. 20 chidren showed postive metacholine challange test at various concentrations.

There was a significant relation between bronchial hyperreactivity (BHR) AND THE NUMBER of smoked cigrattes.Also there was a significant relaton between mean forced expiratory volumm one second(FEV1) reduction as well as cumulative dose(CD) OF MCH and number of smoked cigarettes by the parents.duration of tabacco smoke was found to be of great influence on the chiidren pulmonary function tests.

Conclusion: Passively smoking children has subclinical bronchial hyperreactivity with various degrees, and some of them may turn to be asthmatics. Pulmonary functions of those children depends on multifactors in particular, number of smoked cigarettes and duration of exposure to tobacco smoke. Health education programs to explain the hazardous effect of passive smoking are recommended.

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Association of skin allergy test with bronchial asthma patients

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Purpose: Asthma is a major public health problem in Bangladesh. Allergy plays an important role in asthma. Asthmatics tend to be more atopic and thus more allergic skin test reactivity than non- asthmatics. Hence skin prick test may serve as a useful adjunct in the diagnosis and management of bronchial asthma. Results of Skin Prick Test are an important component in the formulation of preventive plan for the management of bronchial asthma.

Methods: This was a prospective case control study carried out in the Asthma Centre, National Institute of Disease of the Chest and Hospital, Dhaka, Bangladesh. 90 bronchial asthma patients were taken as case and 80 subjects having no history of asthma or allergy were taken as control. The mean age of cases was 25.06 ± 1.28 with male female ratio 1.57: 1 and of control 23.09 ± 1.31 with male female ratio 1.5:1

Results: Percutaneous skin test performed with 30 allergens. Frequency of immediately reacting skin (positive) found in group A were, Mite (78.89%). House dust (78.89%), Cockroach (33.33%), Mosquito (25.56), Fungi (16.67%), Grain dust rice (32.22%), Paper dust (27.78%), Straw dust (33.33%), Cow dander (30%), Cat dander (15.56%), Dog dander (12.22%), Wool mixed (23.33%), Chicken feather (14.44%), Beans fresh (26.67%), Coconut (13.33%), Ladies finger (14.44%), Chicken (12.22%), Egg white (13.33%), Prawn (34.44%), Mutton (15.56%) etc. In Group B (Control), positive skin test was detected in mite (7.50%), House dust (5%), Cockroach (2.5%), Mosquito (3.75%), Ladies finger (2.5%), Tomato (1.25%), Chicken

(2.5%), Egg white (1.25%) and Prawn (5%) In control non-allergic subjects' 1.25%–7.5% shows positive skin test result. Cause of this positive reaction in non-allergic control may be due to differences in selection of subjects, standardization of extracts and recording of results.

Conclusion: Skin allergy test is the most sensitive method for detecting specific IgE antibody. Positive skin test along with clinical relevance determined the consideration of asthmatic patient for immunotherapy. High prevalence of positive immediate hypersensitivity skin test reactions in the asthmatics population is a strong argument that it should be included in the initial evaluation of patients with asthma. Identification of allergen by positive skin test may help in the preventive plan for the management of bronchial asthma patients.

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The additional reasons of cardiovascular risk increase in asthma exacerbation

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A vascular disturbances has a great importance in asthma pathogenesis. Research of mechanical properties of vessels, in particular of arterial stiffness has special interest for the estimation of cardiovascular system function. It is not clear whether increase arterial stiffness observes in patients with asthma.

Methods: We examined 54 patients with severe and moderate asthma by non-invasive arteriography (arteriograph TensioClinic TL1 (TensioMed, Hungary)). Control group included 25 age- and sex-matched healthy volunteers.

Results: The aortal stiffness in asthma exacerbation was significant more, than in healthy persons. It was expressed in increase of aortic pulse wave velocity (aPWV) and augmentation index (IA). APWV in patients with severe asthma a two fold surpassed aPWV in healthy persons and was 10,5 ± 1,3m/s. IA in such patients a 6 fold surpassed control level and was 14,4 ± 5,8%. Significant raised ($\delta < 0{,}001$) arterial stiffness can take part in increasing cardiovascular risk during asthma exacerbation. Independently of increasing arterial stiffness in asthma exacerbation, aPWV and IA essentially improved and came nearer to control level in asthma remission. That suggests functional character of these changes. Augmented arterial stiffness in asthma correlates to lung ventilation dysfunction and hypoxia, disease duration and severity.

Conclusion: Described changes can explain cardiovascular events rising in patients with exacerbation of asthma. Our data suggests transitory character of arterial stiffness increasing in asthma exacerbation.

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Relations of arterial stiffening in asthma exacerbation

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Arterial stiffness is an independent factor of cardiovascular risk. Exacerbation of asthma associates with increase arterial stiffness. The mechanisms responsible of this association are not clear.

Methods: We examined 54 patients with exacerbation of severe and moderate asthma and 25 age- and sex-matched healthy volunteers. Arterial stiffness was estimated by noninvasive arteriography (arteriograph TensioClinic TL1 (TensioMed, Hungary)). Plasma IL-10 and TNF-a levels were determined by the immune-enzyme analysis. Inflammation index calculated as ratio TNF-a/IL-10. Systemic oxidative disbalance was estimated by ratio of oxidative and antioxidative plasma activity, assessed by spectrophotometry. Hypoxemia was assessed by digital pulsoximetry. NOn- plasma level was determined by Greiss's method.

Results: Central arterial stiffness was higher in patients with exacerbation of asthma, than in healthy persons. It was expressed in increase aortic pulse wave velocity (aPWV) and augmentation index (IA). As assessed by correlation analysis both (aPWV and IA) was strongly correlated with hypoxemia ($r = -0{,}921$ and $r = -0{,}876$ ($\delta < 0{,}01$)) and inflammation index ($r = 0{,}902$ and $r = 0{,}868$ ($\delta < 0{,}01$)). Less strongly but clearly aPWV and IA correlated with NOn- level ($r = -0{,}543$ and $r = -0{,}367$ ($\delta < 0{,}01$)) and systemic oxidative disbalance ($r = 0{,}611$ and $r = 0{,}459$ ($\delta < 0{,}01$)).

Conclusion: Our results suggests that hypoxemia, systemic inflammation, hyponitrooxidemia and oxidative disbalance can be involved in raising arterial stiffness in asthma exacerbation. This question requires future researches.

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Cockroach hypersensitivity in asthmatic patients

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Background: Exposure to cockroach was reported to a cause of asthma in many parts of the world.

Objective: To determine the prevalence of cockroach sensitivity in asthmatic children less than 5 years of age in Iranian children with asthma.

Methods: Ninety two (33 female /59 male) patients were asked to complete a questionnaire covering demographic characteristics, and were subjected to skin prick testing for cockroach allergen. Blood samples were also withdrawn for the assessment of total serum IgE and eosinophilia.

Results: Twenty seven percent (n = 25) of subjects had positive skin test results for cockroach allergen. There were no significant correlations between the prevalence of cockroach skin test positivity in male and female children, rural and urban areas or infants under 2 years and olders. The age of youngest subject with positive skin test for cockroach was 2 months. More over, the parent's levels of education and the types and ages of children's residence had no effects on the prevalence of cockroach skin test positivity. There was also no significant correlation between total serum IgE and eosinophilia and the prevalence of cockroach skin test positivity.

Conclusion: Cockroach is an important source of domestic infestations in the city of Shiraz. Sensitization to cockroach allergens may develop early in life.

Skin test for cockroach allergen in children with infantile asthma may provide useful information for institution of environmental controls.

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Relationship between sensitization and duration of asthma associated with allergic rhinitis

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Objective: The aim of the study was to assess sensitization to common inhalant allergens among adult patients with asthma associated with allergic rhinitis (AR) and to find a relationship with the medication and the duration of asthma/AR

Methods: A screening survey questionnaire including questions assessing prevalence of symptoms of asthma over 12 months and questions relating 24 months prevalence of AR symptoms, were distributed to 763 of 16–87 year old (median age 46 yr) patients presented in the outpatients department during of 1 years. The response rate was 83%. Asthmatic patients were diagnosed according to the GINA definition of asthma. AR was diagnosed due to the clinical symptoms (sneezing, itching, rhinorhoea, nasal congestion) and sensitization to allergens.

Results: Hypersensitivity to inhalant allergens (house dust mite, animal dander, pollen) was assessed by skin prick test (positive: > 3 mm) in 32 responders having a clinical history. 73% of patients with asthma and 64% with AR were sensitized to house dust mites (D.pteronyssinus and D.farinae). Hypersensitivity documented by positive skin prick test in patients with asthma and AR was noted to pollen in 47% and 49%, to cat and dog dander in 27% and 11%, respectively.

Conclusion: There was a significant correlation ($r = 0.29$, $p < 0.05$) between frequency of sensitization to house dust mites and duration of asthma. The house dust mite sensitivity is the most prevalent allergen sensitization in adult patients with asthma and AR, establishing control of this allergen as critical for optimizing their treatment.

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IL13 Haplotype and association with atopy

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Background: Bronchial asthma (BA) is a chronic disease associated with elevated IgE levels. It was demonstrated that IL-13, as well as IL-4, plays a crucial role in the regulation of IgE production. The aim of our study was to reveal the association between C-1055T, Arg130Gln (G + 2044A) IL-13 gene polymorphism and level of total IgE in BA patients.

Methods: The polymorphisms in IL-13 gene were analyzed by the PCR-RFLP in 136 asthma patients and 64 healthy control subjects.

Results: Total IgE and IL-13 levels were measured by ELISA. The total serum IgE level was found to be elevated in BA patients group: 290(47,5;495) *kU/L, and within normal range in the control group: 25,5(19;100)kU/L. The levels of total IgE in BA patients and healthy donors differ significantly, $p < 0{,}001$. The frequency of alleles combination - −1055T and + 2044A is 6.9% and reliably is different from that in the control group ($p = 0.008$). Studying the influence of the polymorphisms on the total IgE level we revealed that this level was considerably elevated (up to 315(290;1077)kU/L) among the patients with -1055T and +2044A alleles compared with a group who has allele +2044A and −1055C, and the group who has allele +2044G and −1055C. Total IgE level in this groups was 230(200;440)kU/L and 205(190;377)kU/L, respectively. Patient with allele −1055T and +2044G were not identified. The link between disease and studied traits (−1055T and +2044A) determined by the Wolf's coefficient of association , which was 0.9.

Conclusion: Thus in our study we revealed the association between C-1055T, Arg130Gln IL13 gene polymorphism and total IgE levels and atopy.

*Data are presented as interquartile range: median (25% quartile; 75% quartile).

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Asthma in the elderly in Korea: a comparison according to the onset of asthma in the elderly

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Background: Despite its high prevalence, elderly asthma remains unrecognized and misunderstood because its symptoms may not be typical as in young patients. And because other cardiac or respiratory diseases may be frequently associated. Only a few studies on the clinical characteristics of elderly asthma have been reported till now.

Methods: To investigate whether the characteristics of elderly asthma were different according to the onset of asthma, one hundred eighty asthmatic patients over the age of 65 were enrolled who attended the Asthma and Allergy Clinic, Seoul National University Bundang Hospital.

Results: Of 180 patients, ninty nine patients have developed asthma over the age of 65. This group with late-onset asthma (LOA) had a mean duration of disease of 3.4 ± 3.5 yr. The remaining group with early onset asthma (EOA) had a mean duration of illness of 18.8 ± 17.1 yr. These groups were indistinguishable by the symptoms, the medication requirements and the serum level of total IgE. These with EOA had a greater degree of airway obstruction in FEV1 of predicted value ($p < 0.028$) and a greater likelihood of neutrophils in induced sputum. Moreover, the novel indicator, namely, neutrophil/eosinophil ratio of induced sputum was inversely related to several PFT indices, including FEV1, FEV/FVC, FEF25-75%, and FEF25-75%/FVC. These findings suggest not only eosionphil but also neutrophil could play an important role in the airway inflammation and airflow limitation.

Conclusion: Our study sggested that cellular compositions of airway inflammation, and lung function depends on the onset and possibly duration of disease in elderly asthmatics. These findings suggest that early onset and long-standing asthma may lead to chronic persistent airway obstruction and thereby mimic chronic obstructive lung disease.

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APAPARI: questionnaire based assessment of monitoring and treatment of asthma by physicians in infants, preschool and older children

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Background: The APAPARI questionnaire was designed to assess the treatment patterns of childhood asthma in the Asia-Pacific region.

Methods: A total of 262 doctors treating asthma were invited to respond to the APAPARI questionnaire evaluating treatment patterns of childhood asthma.

Results: Of the 262 doctors, 80 completed the questionnaire while 2 did not complete it as they did not treat children. 59 were pediatricians, 14 were pediatric pulmonologists/ allergists, 2 were pulmonologists and 5 belonged to other specialties. 37(45%) were in private practice or hospitals, 22(27%) were in teaching hospitals while 23(28%) worked in other settings. Responses to the use of symptom cards to monitor asthma were as follows:

use of symptom score card to monitor asthma	number	percentage (%)
never	28	42.0
seldom used	19	23.2
frequently used	21	25.6
always used	13	15.9

Responses for the criteria to monitor childhood asthma are tabulated below:

monitoring tool for childhood asthma	number	percentage (%)
only peak flow meter	33	40.2
peak flow meter and spirometry	24	29.3
neither of the two	24	29.3

For treating an acute attack, 62.8% used nebulised salbutamol/ terbutaline while 15.8% used inhaled salbutamol/terbutaline with spacer and 30.2% used oral corticosteroids. Criterion used for hospital admission in an acute attack of asthma is given below:

critieria used for hospital admission	number	percentage (%)
used no criteria / asthma score	73	89.1
no improvement in sympton / signs	50	60.9
clinical judgement	48	58.5
peak flow meter reading < 50% of best / predicted	26	31.7
desaturation (Sao2 < 90%)	48	58.9

Pulse oximetry as a basis for oxygen therapy was used by 31 (37.8%) in all patients, 40 (48.8%) used oximetry only in severe asthma while 10 (12.1%) did not have pulse oximetry facilities. A favourable outcome was reported by 31 (37.8%) doctors while 8 (9.75%) reported unfavorable results in severe acute asthma with ventilatory support as compared to those treated at a tertiary/university hospital. 36 (43.9%) did not have access to ventilatory/ ICU facilities to treat severe acute attacks. Maintenance therapy in asthmatic infants was always used by 13 (15.8%), frequently used by 34 (41.4%), seldom used by 25 (30.4%), while 8 (9.75%) never used it. Responses on the first

choice of maintenance therapy for asthma among infants, preschool and older children were as follows:

maintenance therapy of choice	infants: n (%)	preschool: n (%)	older children: n (%)
inhaled corticosteroids with long acting β2 agonists	23 (28%)	30 (36.5%)	35 (42.6%)
inhaled long acting β2 agonists only	8 (9.8%)	30 (36.5%)	12 (14.6%)
inhaled corticosteroids only	43 (52.4%)	40 (48.7%)	35 (42.6%)
did not use long acting β2 agonists	52 (63.4%)	49 (59.8%)	52 (63.4%)

58 of the 82 respondents (70.7%) knew but had never used any form of immunotherapy, 11 (13.4%) occasionally used it while 9 out of 82 (10.9%) had no knowledge regarding the availability of immunotherapy.

Conclusion: A significant segment of the doctors do not follow the current management guidelines for childhood asthma.

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Expectancy of Bronchial Hyperreactivity (BHR) in children, simply by the use of a short questionnaire

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Background: The provocation test for assesing BHR is time consuming and sometimes impossible to be performed. Does our questionnaire offer an high expectancy for BHR? Does this questionnaire has any practical value, to avoid the provocation test in certain cases?

Methods: 200 children 9–11 years old from Tirana performed bronchial provocation test with 4.5% saline, according ISAAC protocol. These children answered in the same day to our short and original questionnaire of 4 questions: Have you ever had any shortness of breath or cough in these situations: 1- During or after physical effort (gym hour, going up the stairs, etc) 2- In exposure of tobacco smoke? 3- In exposure of car emanations, heating and cooking gases? 4- By laughing loudly? Every positive answer was coded with scores 1,2,3,or 4, with 0 score if no positive answer, and with 5 score if all positive answers.

Results: Subjects with positive result of saline provocation (change of FEV1 > 15% pre-post provocation) are divided according the scores 0,1, 2, 3, 4, or 5. The same is done for subjects with negative BHR. Not surprisingly, 65% of subjects with negative result after saline provocation had score 0, comparing to 10% of subjects with positive saline, also only 10% of subjects with negative saline resulted with score 5, when more than 50% of subjects positively reacting to saline provocation had score 5. When the subjects with negative result to saline, regardless to result of score questionnaire (nr = 183) are compared with the sub-category of subjects which, beside the negative saline, had also score 5 (nr = 19), we found in the last group a higher rate of personal and familiar history of asthma and allergy. Sensibility, Specificity, Positive Predictive Value and Negative Predictive Value of this questionnaire to forecast the BHR are calculated: 60%, 89%, 24% and 97% for the score 5 (presence of all symptoms) and for score 0 (absence of all symptoms).

Conclusion: 1-This short questionnaire is able to forecast a negative BHR in 97% of cases, if all symptoms are missing (score 0). 2- Subjects with score 5, regardless the provocation result, have an intriguing clinical profile, with positive personal and familiar history for allergy, requiring more attention by doctors and deeper investigations in the future. 3- By only asking 4 simple questions, the time consuming bronchial provocation test could be avoided in everyday medical practice most of the cases, if all the answers are negative.

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Use of asthma control test in clinical practice

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Introduction: The 2006 Global Initiative for Asthma (GINA) reports provide a new asthma classification which is based not only on the severity but also on the level of asthma control. Bronchial asthma is a word-wide problem, with an estimated of 300 million affected individuals. There are more than 200,000 asthmatic patients in our country. Classification of asthma by severity is useful when the doctor has made a decision about management and treatment of a patient. The goal of asthma treatment is to achieve and maintain clinical control. Asthma Control Test is one of a validated measurement to analyze the controlled, partly controlled and uncontrolled levels of asthma.

Aim: The aim of the study was to investigate the level of asthma control in our country.

Methods: In our outpatient clinic, we studied a group of 133 patients (female: 91, male: 42; mean age: 44y) with bronchial asthma. Asthma Control Test was done by all patients and they also have an other eleven questions regarding smoking habits, asthma medications, side effects, exacerbations, exercise induced dyspnoe.

Results: Only 50% of our patients (n = 67 pts) had controlled and partly controlled asthma level, an other 50% (n = 66 pts) had uncontrolled asthma based on Asthma Control Test. The total control level has been achieved only by 18% (n = 24) of our patients. There were unable to work and to have a job the 16 % of the patients and 12% were not able to work even in their household because of suffering asthma. Seventy eight % of patients have reduced quality of life. However, 64% of our pts believed and felt they have total asthma control and only 10% of pts subjective opinion were the uncontrolled asthma.

Conclusion: In our study, we found that patient's opinion about their own condition of asthma did not correlate to an objective test. Asthma Control Test show us the reality of the level of bronchial asthma. The test is very easy to use even alone by the patient. The Asthma Control Test help to achieve and maintain the clinically controlled asthma level.

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Obesity - a risk factor for asthma severity?

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Recently obesity has reached epidemic proportions in many countries. Evidence is now mounting that obesity is also a risk factor for asthma. The aim of our study was to assess relation between obesity and asthma severity in children. We conducted cross-sectional study in the out-patient department at M. Guramishvili Pediatric Clinic in 2005–2006yy. In the study were enrolled 63 children aged 12–15 with a mean age of 13.75, 41 boys and 22 girls. We used body mass index (BMI) to detect overweight and obesity. Weight categories were defined by BMI number position on BMI-age growth chart - risk of overweight - 85th to less than the 95th percentile, obesity - equal to or greater than the 95th percentile. Asthma severity definition was based on patients clinical and/or spirometeric parameters according to GINA guideline. Children with a history of other lung disease were excluded. The prevalence of obesity in our study population was 31.7%. Among girls 55% and among boys only 19.5% were obese. Mean BMI of females with asthma were significantly higher than males ($p < 0.0001$). The obese children had a difference in baseline asthma classification (more likely to have persistent asthma). Despite similar severity of illness at admission, they had a longer length of stay at hospital ($9.8 +/- 7.0$ vs. $6.5 +/- 3.4$ days, $p < 0.01$). We can conclude that there is relationship between body weight and asthma severity and obesity may be a potentially modifiable risk factor for asthma or asthma exacerbation.

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Overweight affects eotaxin levels in asthma patients

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Background: Recent data from animal studies indicate that fat tissue is a source of eotaxin, which may play an important role in asthma. However, the link between obesity and airway inflammation in asthma is not well understood.

Objective: We aimed to investigate the impact of obesity on eotaxin levels in patients with stable mild/moderate asthma.

Methods: 20 steroid-naive asthmatics (mean age: 57.2 ± 2.2 yrs) and 9 control subjects without lung diseases (mean age: 59.4 ± 9.7 yrs) were investigated. According to the body mass index (BMI), subjects were divided into the 2 groups: subjects with normal weight (BMI < 25kg/m^2) and subjects with overweight (BMI ≥ 25 kg/m^2). Spirometry, bronchial provocation test with methacholine (PD_{20}), sputum induction and bronchoscopy were performed. Eotaxin-1, -2, -3 concentrations in serum, sputum supernatant and BAL fluid were measured using commercial ELISA kits.

Results: In asthma patients with overweight eotaxin-2 concentration in induced sputum was significantly higher comparing to asthma patients with normal weight (118.8 ± 46.6 vs 20.1 ± 9.3 pg/ml) and control subjects (118.8 ± 46.6 vs 16.7 ± 7.2 pg/ml; $p < 0.05$). Whereas, eotaxin-1, eotaxin-3 did not significantly differ between studied groups, and there was no correlations between eotaxin levels and number of eosinophils in induced sputum and BAL fluid.

Conclusion: Elevated sputum eotaxin-2 concentration in asthmatics with overweight let suggest, that increased body mass may upregulate inflammatory markers in asthma patients.

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Assessment of quality of life in Iranian asthmatic children and young adults and their caregivers

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Background: Interest in the impact of illness on day to day function is leading investigators to include both disease specific and generic health related quality of life (HR QOL) questionnaires in a broad range of clinical studies and to gain a full picture of the impact of asthma on the lives of children with this condition, it is necessary to make direct measurement of health related quality of life.

Methods: In response to this need, we used the Juniper's pediatric asthma quality of life questionnaire (PAQLQ) and Juniper's Pediatric Asthma Caregiver's Quality of Life Questionnaire (PACQLQ) that has been developed based on guidelines for construction of over a dozen validated disease specific quality of life instruments.

The PAQLQ consists of 23 items that in children with asthma have been identified as troublesome in their daily lives and PACQLQ that contains 13 items in two domains of emotional and activities disturbances. The study design consisted of an 18 month single cohort study. Patients participating in the study were 113 children, 7–17 years of age, with a wide range of asthma severity and their caregivers. For each patient a PAQLQ and for each caregiver a PACQLQ was completed. One week before visit patients recorded morning peak flow rates, medication use and symptoms in a diary. After complete physical examination, for determining of asthma severity, spirometry was performed.

Results: The questionnaires after statically analysis showed good levels of both longitudinal and cross sectional correlations with the conventional asthma indices and with general quality of life. We found that consistently QOL in boys were more disturbed than females, a good relevancy between severity of asthma and QOL scores and more disturbances of QOL in caregivers of male asthmatic patients than caregivers of female asthmatic patients. We could not find any significant relevancy between FEV1 percentage of predicted and overall scores of QOL.

Conclusion: Smaller airways, and higher airway resistance and more activity of males than females may explain why boys have more disturbed life style than females.

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A Iranian asthmatic research

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Asthma and quality of life in iranian asthmatic patient asthma is a chronic disease,that make many health problems in every where in the world. The aspect of quality of life in iranian asthmatic patient are variable.

Objective: To determine factors influence to quality of life in patients with asthma, who reffer to lung clinic of general hospital in tehran.

Methodology: This research is a descriptive study.the subjects consisted of 386 patients.samples selected by sample randomized.the method of collecting data was by questionnaire.the data were collected in one stage and analyzed with descriptive and inferential methods and spss soft ware.

Findings: The results showed that ,the quality of life inpatients from physical aspect was inappropriateand, in social and economic aspects were rarely appropriate. the most of patients from mental and sleep aspects were inappropriate.

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Neither sex nor overweight are risk factors for having asthma symptoms in adolescent population of Argentina

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Background: A positive association between body mass index (BMI) and asthma has been recently suggested; however, the sex-dependence of this association remains controversial. The aim of the present study was to explore the relationship between BMI and asthma, as well as its sex-dependence in young adolescents.

Methods: Self-reported data obtained from the standardized International Study of Asthma and Allergies in Childhood (ISAAC) Phase Three. Written questionnaires of 5579 young adolescents aged 13–14 years old from randomly selected private and public schools in Salta and Cordoba (Argentina) were used. The BMI for each individual was calculated; international cut-off points for BMI for overweight and obesity by sex and age were used (Normal: 18 until 24.99 kg/m2, Overweight: 25 until 29.99 kg/m2 and Obesity > 30 kg/m2). Because of the very low prevalence of obesity (0.86 %), they were included in the overweight group (4.6%). Each variable of the ecological questionnaire considered as probable confounder was confronted in 2x2 tables with having wheezed in the last 12 months(13,1%) and BMI, and those variables found significantly associated were included in the final analysis. The data were statistically analyzed by the chi-square test, P and odd ratios (OR, 95 % CI) in binary logistic regression.

Results: 51,4% of responders were male. Being a boy was associated with overweight (OR 1,35. 95%CI 1,06-1,71; P 0,013), and exercising one or less/week was associated with obesity (OR 2. 95%CI 1,07-3,70; P 0,025). After adjusting for each one, both were significantly associated to an increased BMI (P 0,02 and 0,01 respectively). Watching TV one or less hour/day was related to have a lower BMI (OR 0,72. 95%CI 0,55-0,93; P 0.01). Exercising once or less per week was the only factor associated to wheezing after adjusting for BMI and sex (P 0.0001).

Conclusion: In our population, neither sex nor overweight were associated to wheezing symptoms; however sex was significantly associated with overweight. Maybe the scarce obese population in our cities contributed to these findings, however sedentary profile contributed to an increased BMI and to wheezing symptoms also, as was expected.

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A three-year follow-up study of asthma, airway symptoms and self-reported allergy among pilots and cabin crew at commercial aircraft

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Background: Cabin air quality is important especially to employees with sensitive airways. There are few studies on incidence of asthma and allergy in pilots and cabin crew. The aim of the study was to study the relations between occupational factors and personal factors (age, gender, atopy, smoking habits) and incidence of asthma and allergy.

Methods: A standardized questionnaire (Örebro, MN040 NA) was answered in 1997 by pilots (n = 577) and cabin attendants (n = 936). Three years later, in 2000 the same questionnaire was sent to pilots (n = 436) and cabin attendants (n = 698) on duty. The new onset of asthma and allergies (three year incidence) were calculated by logistic regression related to personal factors with logistic regression.

Results: The prevalences of airway symptoms and asthma were two to three times lower in pilots than in cabin attendants. The three year incidence of doctors diagnosed asthma was 1.2% among pilots, and 0.6% among cabin attendants. The incidence of any airway symptom (wheeze, attacks of breathlessness) was 4.6% among pilots, and 12.5% among cabin attendants. The incidence of chronic bronchitis (daily productive cough) was 1.1% among pilots, and 3.5% among cabin attendants. Further, the incidence of pollen allergy and furry pet allergy was 5.4%, and 1.6% respectively among pilots and

5.7% and 2.4% respectively among cabin attendants. The incidence of food allergy/ intolerance was 1.8% among pilots and 3.2% among cabin attendants. The incidence of airway symptoms was lower in pilots ($p < 0.05$). The incidence of doctors diagnosed asthma and food allergy was higher in atopics ($p < 0.01$). Younger cabin crew and pursers had a higher incidence of pollen-/ furry pet allergy ($p < 0.01$). No relations was seen as to gender and own tobacco smoking.

Conclusion: Age, atopy, and type of occupation was related to the incidence of asthma and self-reported allergies. The incidence of doctors diagnosed asthma among pilots (0.4%/year) was somewhat higher than expected. Airway symptoms in genral were two to three times more common among cabin attendants. The study started before the stop of smoking on intercontinal flights.

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Parietaria pollinosis (seasonal allergic rhinoconjuctivitis or/and asthma) in Greece: an increasing problem during 20 years (1987–2006)

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Background: It is a well-known phenomenon that the prevalence of Seasonal Allergic Rhinoconjunctivitis or / and Asthma (SAR/A) is increasing. But there is little data about the changes in frequency of sensitization to pollens in new patients with SAR/A during the time (years).

Objective: To determine the frequency of sensitization to three important pollens (Grasses, Parietaria, Olive) in new patients with SAR/A, each year, from 1987 to 2006(20 years) in Greece.

Methods: We have studied, each year (from 1987 to 2006), all new patients with solely seasonal typical symptoms of rhinoconjuctivitis or/and asthma and positive Skin Prick Tests (SPT's) to Grasses, Parietaria and Olive (one or more). SPT's were considered as positive if the mean of diameters was at least 5-mm larger than the negative control. The proportion of men/women and the mean age (years) in each year, from 1987 to 2006, were similar. For statistical analysis we used the ÷2 for trend.

Results: Patients with SAR/A-positive SPT's ÷2 for trend test Grasses x2 = 28,83, p > 0,05, Parietaria x2 = 63,78, p < 0,001, Olive x2 = 18,96, p > 0,05.

Years	Number	Grasses(%)	Parietaria(%)	Olive(%)
1987	76	85.5	30.3	56.6
1988	141	68.8	34	60.3
1989	161	75.8	37.3	60.9
1990	148	63.5	39.9	52.7
1991	166	77.7	41.6	44
1992	205	69.3	50.7	57.1
1993	137	67.1	57.7	53.3
1994	173	64.7	58.4	54.9
1995	168	65.5	60.1	54.2
1996	217	68.2	56.2	40.1
1997	216	65.3	50.5	41.2
1998	189	70.4	51.8	40.7
1999	164	64	56.1	46.3
2000	182	60.4	59.3	52.2
2001	191	61.8	62.8	48.2
2002	238	61.3	60.5	50.8
2003	184	66.8	60.3	44.6
2004	147	63.3	64.4	42.9
2005	185	60.5	55.7	47
2006	178	55.6	55.1	43.3

Conclusion: Our results indicate that Parietaria pollinosis is more and more frequent, from 1987 to 2006, in Greece among patients with SAR/A. Parietaria pollinosis appears to be an increasing problem during the last 20 years, in Greece. This phenomenon may represent the influence of environmental factors.

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Prevalence of asthma, rhinitis and eczema among Brazilian and Japanese adolescents (AD) in the city of São Paulo, Brazil

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Objective: To evaluate the importance of the genetic background on the prevalence of asthma (A), rhinoconjunctivitis (R) and atopic eczema (AE) among AD born in Brazil and from different genetic backgrounds.

Methods: ISAAĆs written questionnaire was applied to two groups of 12–15-year-old AD of similar socioeconomic status living in the city of São Paulo: Brazilian (N = 381) and Japanese (third generation, born in Brazil, from families without mixed marriages, N = 221). Affirmative response to "Have had wheezing in the last year" identified AD with A, to "Nasal and ocular symptoms in the last year" those with R, and to "Eczema in the last year evidenced in flexural areas" those with AE. Questions "Speech problem in last 12 months during acute attack", "Nasal symptoms interfering with daily activities", and "Kept awake at night by this itchy rash in the last 12 months" characterized severity of A, R, and AE respectively.

Results: There were no differences according to gender in both groups except for the prevalence of R that was higher among Brazilian females. The prevalence of A (12.7% x 19.4%, $p < 0.05$) and the prevalence of R (10.4% x 24.4%, $p < 0.05$) were higher between Brazilian AD when compared to Japanese AD, respectively. There were no difference in A severity (0% x 1.1%) but R's severity was significantly higher between Brazilian AD (10.4% x 24.4%, $p < 0.05$). The prevalence of AE was higher between Japanese AD (7.7% x 3.1%, $p < 0.05$) but without differences in its severity (1.8% x 1.8%, $p > 0.05$). The prevalence of these diseases is not different from those observed in Japan and in São Paulo (ISAAC, phase 3).

Conclusion: The prevalence observed among the third generation of Japanese AD without miscegenation born in Brazil, showed similar results to those obtained in ISAAC phase 3 recently published in Japan. Considering that both AD groups evaluated have been exposed to the same environment we might speculate that the genetic background in this Japanese population is more important than environmental influences on the development of asthma and allergic diseases.

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Establishment of Cohort for Reality and Evolution of Adult Asthma in Korea (COREA), a large multi-center long-term cohort study of whole spectrum of asthma: Clinical characteristics according to severity of asthma

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Background: Asthma is a chronic inflammatory disorder which has widespread, variable airway obstruction and airway hyperresponsiveness (AHR) leading to dyspnea, chest tightness, wheezing and cough. Because it often presents heterogeneous features and various phenotypes according to its severity, it is needed to find good predictable clinical factors concerning severity. Also it cannot be discriminated from chronic obstructive pulmonary

disease, many researchers used to recognize them as a spectrum of 'chronic obstructive airway disorder' (COAD). Accordingly, it is most important that we can reveal prognostic factors influencing on natural course of asthma and prohibit from progressing to severer and irreversibly uncontrolled asthma. It is thought that this is possible only in case asthmatic patients should be observed through long term follow up such as cohort study.
Objective: To define the important prognostic factors contributing to asthma severity and suggest new clinical understanding and paradigm of COAD.
Methods: Researchers from 11 university hospital have established asthma cohort named 'Cohort for Reality and Evolution of Adult Asthma in Korea' (COREA) since year of 2005 in Korea, which will be continued up to 2013. We have collected patients having chronic persistent respiratory symptoms such as dyspnea, cough, sputum or wheezing more than 3 months and airflow limitation by FEV1/FVC less than 0.7 or AHR confirmed by methacholine bronchial provocation test (MBPT). As the first study of our cohort, we analyzed various clinical factors influencing on the severity of asthma for registered 644 patients at the time of first-visit. Severity was assessed by special allergists by the definition of 2002 GINA guideline.
Results: Severity of asthma was associated with skin prick test positivity ($p < 0.006$), history of hospital visit due to asthma exacerbation ($p = 0.001$), symptom onset ($p = 0.037$) and duration of diagnosis ($p = 0.041$). Severity also was significantly correlated with FVC, FEV1, and bronchodilator response (all $p < 0.001$). However, smoking didn't affect the severity of asthma, even in case of passive smoking.
Conclusion: We suggest that history of hospital visit due to asthma exacerbation may be an important clinical index reflecting severity of asthma. Furthermore, it is expected that many valuable findings can be derived from our cohort study

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Allergy in Latin America. A public health problem who's time has come

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In Venezuela, asthma causes more than one million crises per year, a rather significant impact to the health system compared to USA reports. Our demographics situation are similar to the rest of Latin America, with a young and mostly urban population living under variable conditions of poverty; application of massive vaccination programs and other public health measures will ensue that non-communicable diseases occupy major roles in the near future. For Venezuela ISAAC data shows an eighteen percent of wheezing in the past year with an overall prevalence of thirty two percent is reported.
Methods: In order to explore such previous high prevalence, we carried out a multidisciplinary and ongoing study (approved by the ethical committee of the Institute of Biomedicine, ratified by the academic council of the medical Faculty of Central University of Venezuela) in 1000 unselected school children, 6–12 years old, from low to middle socioeconomic background covering different areas from Caracas, Venezuela. After an informed consent was signed from parents or guardians, all children from randomly selected schools were evaluated with a validated modified Graffar's socioeconomic questionnaire and an allergic rhinitis, asthma and atopic dermatitis validated questionnaire after ARIA, GINA and Hannifin criteria; skin prick testing (ALK-ABELLO) for common food and inhalant allergens plus total IgE (ELISA), complete blood count (COULTER) and serial feces examination for ova and parasites were performed in all children. Pre and post bronchodilator FEV1 and PEF spirometric measurements values were obtained (MICROLOOP), and included in data analysis.
Results: We found that 60% of the child evaluated had allergic rhinitis, 40% had asthma, and 21.9% Atopic Dermatitis with or without other allergic pathologies. We also found that 64.8% of the child evaluated had parasites and 41.1 had respiratory infectious disease associated.
Conclusion: Our data confirms the high prevalence of allergic diseases in Venezuela employing questionnaires, a clinical exam and immune diagnostic evaluation methodology. Hence, our health system and medical faculties should be highly aware of these and similar findings for establish a proper education programs, both at primary health care personnel and also in communities. Developing nations are waiting for simpler but effective approaches in the management of a public health problem who's time has come.

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Validation of an instrument for epidemiologic studies of recurrent wheezing in infants

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Background: Large international studies on asthma and allergies in childhood have found notorious variation in prevalence and temporal trend among countries. However, there is no international studies on the epidemiologic characteristics of wheezing in the first year of life. The aim of this study is to validate a questionnaire to assess the prevalence of recurrent wheezing infants.
Methods: This study was undertaken in pediatric emergency rooms (PER). A randomized sample of parents visiting PER for lower respiratory illness, answered a questionnaire on wheezing developed by the International Study of Wheezing in Infants (EISL, from spanish Estudio Internacional de Sibilancias en Lactantes). During visit, all infants were examined by one of the authors (blind) who reported, or not, the presence of wheezing on chest auscultation. Sensitivity, specificity, predictive positive value, predictive negative value and concordance (κ) level were calculated from parent and physician reports.
Results: Two hundred and nine infants aged 12 to 15 months participated in the study. Fifty six parents reported current wheezing and 43 were confirmed by physician; 153 parents did not report current wheezing and 146 had not wheezes at physical examination ($\kappa = 0{,}74$, CI 95% 0,64 – 0,85). This questionnaire showed high sensitivity (86%), specificity (91,8%), positive predictive value (76,8%) and predictive negative value (95,4%).
Conclusion: Regardless of previous experience with wheezing episodes, parents can reliably inform when their infants are currently wheezing. A simple and convenient questionnaire confirmed by physical examination produce an accurate tool to asses the prevalence of asthma symptoms in infants.

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Different seasonal (monthly) variation in asthma exacerbations in patients with allergic and non allergic asthma

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Background: The recognition of clear seasonal (monthly) patterns of Asthma Exacerbations (AE) in a particular area may allow preventive strategies to be developed.

Objective: The aim of this study was to determine the seasonal (monthly) variation in AE in patients with allergic and non-allergic asthma.

Methods: Data were obtained from our department in Athens, between October 1986 and September 2006 (240 months). We evaluated all AE occurring this period for patients with Allergic Asthma (AA) and Non-Allergic Asthma (N-AA). The diagnosis and classification in AA and N-AA were made according to the results of skin prick testing (SPT) to common aeroallergens. Patients with AA defined as those with > 1 positive SPT and patients with N-AA defined as those with negative SPT. A positive SPT was defined as a wheal size at least 3 mm greater than the negative control. AE were defined as a deterioration in asthma resulting in an unscheduled visit (i.e patient-initiated) leading to change in asthma treatment or the need for oral steroids for > 3 days and/or emergency room visit/hospitalization. Data are presented as monthly averages of these years of combined data, as a percent above (+) or below (−) the average monthly value (%) for the 240 months under study.

Results: There were 3.570 AE (2.313 in males, 1257 in females) in 2.345 patients with AA (1.569 males, 776 females). Also, there were 2.585 AE (1.335 in males, 1250 in females) in 1.518 patients with N-AA (736 males, 782 females).

Percent above or below an average monthly value (%).

Months	AA	N-AA
January	−42.9	+15.6
February	−52.2	+11
March	−19	+13.7
April	+37.6	+10
May	+307.1	− 9.9
June	+22	−19.7
July	−44.5	−42.4
August	−75.1	−68
September	−19.5	+15.6
October	−35.1	+43.4
November	−27.3	+31.4
December	−50.2	−0.6

Conclusion: These findings suggest that AE have a different clear seasonal (monthly) variations between patients with AA and N-AA. Patients with AA showed an increase in AE in April, May and June (spring and early summer). Especially in May occurred a great increase (peak) in AE (+307,1). Patients with N-AA had an increased AE from January until April (winter and spring) and from September until November (autumn). These results may offer significant opportunities for improved disease management.

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Seasonal (monthly) variation in asthma exacerbations in Greek children: the effect of age

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Background: The recognition of clear seasonal (monthly) patterns of Asthma Exacerbations (AE) in pediatric patients, in a particular area, may allow preventive strategies to be developed.

Objective: The aim of this study was to determine the seasonal (monthly) variation in AE in pediatric patients with weezing and/or asthma according the age.

Methods: Data were obtained from our department in Athens, between October 1986 and September 2006 (240 months). We evaluated all AE occurring this period for pediatric patients 0–4 years, 5–9 years and 10–14 years. AE were defined as a deterioration in wheezing/asthma resulting in an unscheduled visit (i.e patient-initiated) leading to change in wheezing/asthma treatment or the need for oral steroids for > 3 days and/or emergency room visit/hospitalization. Data are presented as monthly averages of these years of combined data, as a percent above (+) or below (−) the average monthly value (%) for the 240 months under study.

Results: There were 2.014 AE (1.285 in males, 729 in females) in 1.071 wheezer/asthmatic patients (658 males, 413 females). From 2.014 AE, 642 were identified in patients 0–4 years, 854 in patients 5–9 years and 518 in patients 10–14 years.

Percent above or below an average monthly value (%)

Months	0–4 years	5–9 years	10–14 years
January	−6.4	−15.5	−32.6
February	−13.9	+4.2	−25.6
March	+4.9	+2.8	+23.2
April	−15.7	+22.5	+7
May	−19.5	+35.2	+137.2
June	−28.8	−16.9	+34.9
July	−68.2	−33.8	−60.5
August	−77.5	−74.6	−58.1
September	+19.9	+15.5	+44.2
October	+77.9	+39.4	−9.3
November	+102.2	+33.8	−30.2
December	+27.3	−9.9	−25.6

Conclusion: These findings suggest that AE have a different clear seasonal (monthly) variation between the three age groups in pediatric patients. Children under 5 years (0–4 years) showed an increase in AE in March and from September until December (autumn and early winter). Children 5–9 years showed an increase in AE from February until May (late winter and spring) and from September until November (autumn). The group of older children (10–14 years) had an increased AE in spring (March to May with a peak in May +137,2%) and early summer (June) while in the rest of the months only in September. This study suggests that difference in allergic and environmental factors implicated in provoking AE in pediatric patients and that clear seasonal (monthly) variations exist in AE which are specific to age. These results may offer significant opportunities for improved disease management.

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Seasonal (monthly) variation in asthma exacerbations in patients allergic to pollens: the May epidemic of asthma in Greece

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Background: The recognition of clear seasonal (monthly) patterns of Asthma Exacerbations (AE), in a particular area, may allow preventive strategies to be developed.

Objective: The aim of this study was to determine the seasonal (monthly) variation in AE with allergy to pollens, in Greece.

Methods: Data were obtained from our department in Athens, between October 1986 and September 2006 (240 months). We evaluated all AE occurring this period for patients allergic only to pollens, according to the results of skin prick testing. A positive SPT was defined as a wheal size at least 3 mm greater than the negative control. AE were defined as a deterioration in asthma resulting in an unscheduled visit (i.e patient-initiated) leading to change in asthma treatment or the need for oral steroids for >3 days and/or emergency room visit/hospitalization. Data are presented as monthly averages of these years of combined data, as a percent above (+) or below (−) the average monthly value (%) for the 240 months under study.

Results: There were 1.723 AE (1.099 in males, 624 in females) in 1.219 asthmatic patients (798 males, 421 females). From 1.723 AE, 218 were identified in patients 0–14 years, 615 in patients 15–29 years, 600 in patients 30–44 years and 290 in patients >45 years.

Months	AE (number)	Percent above or below an average monthly value (%)
January	54	−62.3
February	45	−68.6
March	93	−35.1
April	230	+60.4
May	826	+476
June	192	+33.9
July	60	−58.1
August	37	−74.2
September	43	−70
October	53	−63
November	51	−64.4
December	39	−72.8

Conclusion: These findings suggest that AE in the Athens region have a clear cut seasonal (monthly) variation in patients allergic to pollens. An increase in AE occurred in three months (April, May, June) and a decrease in the rest of the months. It is of great interest to note that especially in May occurred a huge increase (peak) in AE (+476%) in patients allergic to pollens (May epidemic). This study suggests that aeroallergens (pollens) can exacerbate asthma, especially in May, in Greece and the results may offer significant opportunities for improved disease management.

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Is there seasonal (monthly) variation in asthma exacerbations in patients allergic to mites (D.Pteronyssinus, D.Farinae) in Greece?

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Background: The recognition of clear seasonal (monthly) patterns of Asthma Exacerbations (AE), in a particular area, may allow preventive strategies to be developed.

Objective: The aim of this study was to determine the seasonal (monthly) variation in AE with allergy to mites (D. Pteronyssinus, D. Farinae), in Greece.

Methods: Data were obtained from our department in Athens, between October 1986 and September 2006 (240 months). We evaluated all AE occurring this period for patients allergic only to mites (D. Pteronyssinus, D. Farinae), according to the results of skin prick testing. A positive SPT was defined as a wheal size at least 3 mm greater than the negative control. AE were defined as a deterioration in asthma resulting in an unscheduled visit (i.e patient-initiated) leading to change in asthma treatment or the need for oral steroids for >3 days and/or emergency room visit/hospitalization. Data are presented as monthly averages of these years of combined data, as a percent above (+) or below (−) the average monthly value (%) for the 240 months under study.

Results: There were 590 AE (370 in males, 220 in females) in 342 asthmatic patients (214 males, 128 females). From 590 AE, 193 were identified in patients 0–14 years, 231 in patients 15–29 years, 116 in patients 30–44 years and 50 in patients >45 years.

Months	AE (number)	Percent above or below an average monthly value (%)
January	32	−34.7
February	41	−16.3
March	58	+18.4
April	59	+20.4
May	74	+51
June	55	+12.2
July	37	−24.5
August	10	−79.6
September	73	+49
October	67	+36.7
November	54	+10.2
December	30	−38.8

Conclusion: These findings suggest that AE in the Athens region have a seasonal (monthly) variation in patients allergic to mites (D. Pteronyssinus, D. Farinae) in Greece. An increase in AE occurred in the spring months (March, April, May) and the June (peak in the May +51%) and also in the autumn months (September, October, November) with a peak in September (+49%). A decrease in AE occurred in the rest of the months with a nadir in August (−79,6%). Although AE may be triggered by a number of other factor (i.e respiratory viral infections), this study suggests that asthmatic patients allergic to mites represent a seasonal (monthly) variation in AE, in Greece. These results may offer significant opportunities for improve disease management.

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Epidemiology of bronchial asthma in Sverdlovsk region

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Background: The goal of the research was to study the prevalence of bronchial asthma (BA) and bronchial asthma risk factors (RF) in Sverdlovsk region.

Methods: The BA prevalence and RF were studied with the help of the ECRHS questionnaire and the RF questionnaire, which were answered by the rural and urban population in 2000 and 2006.

Results: It was found that the asthma prevalence rate in Ekaterinburg is 5,9%, in villages - 3,6%. BA atopic form was 71% and 76% correspondingly. The most spread asthma RF are: allergic rhinitis, a strain of asthma and allergy, bad factors in production facilities, presence of cockroaches, birds and fish aquariums in living apartments. In rural region the most spread asthma RF are: allergic rhinitis, a strain of asthma and allergy, presence of fish aquariums in living houses.

Conclusion: In Sverdlovsk region BA is spread in the urban area wider than in the rural region. The BA atopic form prevailes both in urban and rural areas. In the city the important asthma RF are bad factors in production facilities and presence of cockroaches and birds. Asthma RF common for urban and rural areas are: allergic rhinitis, a strain of asthma and allergy, presence of fish aquariums in living houses/ apartments.

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Outcome of childhood asthma in 205 asthmatic children

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The aim of this study was to investigate the outcome of childhood in asthma. 205 asthmatic children aged 2 to 14 years seen between the years 1990–91 were followed prospectively till 2005. The severity of asthma was classified: mild, moderate or severe. The severity was evaluated every three years by changes in respiratory symptoms and spirometry. Initially 32p.100 of children have mild asthma, 25p.100 moderate asthma and 43p.100 ware considered as severe asthma. 67p.100 of children who had initially mild asthma have the same severity ten years later, 21p.100 have moderate asthma and 2p.100 have severe asthma. 70p.100 of children who had mild asthma did have mild asthma ten years later. 60p.100 of children who had severe asthma are considered severe ten years later. The final prognosis was influenced by the initial severity of the asthma and presence of associated atopic disease.

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Effects of nitric oxide synthase inhibitors on airway hyperresponsiveness and inflammation in a murine asthma model

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Introduction: Excessive airway production of nitric oxide (NO) has been suggested to play a role in bronchial asthma. It is still not clear which iNOS isoform is involved in eosinophilic airway inflammation.

Objective: A selective Inducible nitric oxide synthase (iNOS) inhibitor 1400W (N-3-(aminomethyl) benzyl) acetamidine) or a nonselective NOS inhibitor L-NAME (N-nitro-L-arginine methyl ester) have been tested in a murine acute asthma model.

Methods: Six week-old female BALB/c mice were sensitized and exposed to aerosolized ovalbumin (OVA). BALB/c mice (each group, n = 4) were treated with indicated dose of 1400W (2 mg/kg, 4 mg/kg, 8 mg/kg, intraperitoneal) or L-NAME (10mg/kg, intraperitoneal) for five times before allergen challenge. Methacholine bronchoprovocation test and bronchoalveolar lavage (BAL) fluid analyses were performed.

Results: Airway inflammation was decreased only in 1400W- treated groups. BAL fluid total cell counts (positive control 428 x 10^3/μl, 2 mg/kg 273x10^3/μl, 4 mg/kg 210x10^3 /μl, 8 mg/kg 154 x 10^3/μl, each $p = 0.012$, $p < 0.001$ $p < 0.001$) and eosinophil counts (positive control 318x10^3 /μl, 2 mg/kg 150 x 10^3/μl, 4 mg/kg 142x10^3/μl, 8 mg/kg 105x10^3/μl, each $p = 0.016$, $p = 0.009$, $p = 0.003$) were decreased in 1400W- treated groups compared with positive control group. But there were no significant differences in airway hyperresponsiveness (Log PC200 $p = 0.719$) regardless of treatment.

Conclusion: The results demonstrate that a selective iNOS inhibitor 1400W is effective in improving the airways inflammations through reducing total inflammatory cell counts in the OVA-sensitized BALB/c mice.

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Relationship with respiratory function tests and psychological and somatic status in females with bronchial asthma

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Background: To assess the relationship among the sexual, psychological, hormonal, and physical status of women with bronchial asthma (BA).

Methods: Twenty-nine women with BA and 18 healthy women as the control group were enrolled in the study. The patients were asked to complete the Female Sexual Function Index, General Health Questionnaire, and Medical Outcomes Study Short Form 36-item Health Survey (SF-36). The mental and physical component summary scores were calculated using the answers on the SF-36. The same questionnaires were given to this group as well. Statistical analysis was performed using the Mann-Whitney U test and Pearson correlation tests.

Results: Statistically significant differences were observed for all questionnaire scores ($P < 0.01$). The most common female sexual dysfunction was diminished arousal (n = 21, 72.41%) in women with BA. In the correlation analysis, the total Female Sexual Function Index score had a statistically significant and positive correlation with the mental component summary score ($r = 0.517$, $P = 0.001$) and a negative correlation with the General Health Questionnaire score ($r = -0.368$, $P = 0.01$).

Conclusion: The results of our study have shown that bronchial asthma can be a cause of female sexual dysfunction with mental and psychiatric mechanisms.

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Oxide stress index and psychosomatic characteristics in children with Bronchial Asthma

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In the difficult pathogenesis of Bronchial Asthma is important nerve inflammatory mechanism and unbalance of the various group of autonomic nervous system. The process made by bronchial asthma cause lipid phase cell membrane changes. The importance is the oxidation process reinforcement and to development an antioxidant syndrome.

The study aimed to asses the last product of lipid peroxide oxidation (LPO) - enzyms: Malon Dealdehyde (MDA), ceruloplasmin (CP) and superoxidismutas (SOD) and the function of autonomic nervous system. The situation and personal nervous status were fulfilling the diagnosis tests of C. Spilberg, the emotional status were studied with Lusher's color test. The investigation of some index of oxidation stress and psychosomatic characteristics in children with bronchial asthma. 90 patient (median age of patients was 7–15 years) with bronchial asthma and practically, 100 healthy children were investigated. The mathematical treatment of dates had made with program package SPSS,v.12.

Results: In the blood serum of the patients the contemt of mda was 63% high than in control group. ($p < 0,05$). Mda level in the patient with eytonic type of vegetative regulation was lower than in vagotonyc patients ($p < 0.05$). Especially important was the coefficient dates. LPO product quantity in the patients with 0,05). At < vagotonyc tape is straight contact with emotional stress ($r = 0,63$; p the end of the study was carried out, that bronchial asthma is conducted the high level of LPO products in blood serum. The LPO activate has shown at almost everytime – from 1 to 5 year and more. Results of the investigation show us the LPO prosess activateing at the time of bronchial asthma. It's established the coordibation of the LPO level in the blood serum

and the severity of disease of the patients with bronchial asthma, which confirmed the antioxidant decrease in the patients. Low antioxide guard in the patients with eytonic type of vegetatic regulation give us a chance to seperate them as biochemical «nontrustworthy » lipid peroxide oxidative system.

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Relation of BCG-Scar diameter and asthma in children

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There are conflicting ideas about the inverse relationship between delayed type hypersensitivity reaction of BCG and atopic state. Stronger response to PPD test as an indicator of more potent TH1 response is supposed to influence TH2-modulated allergic reactions, but it is still doubtful whether scar of BCG vaccine can also be supposed as an indicator of TH1-immune response or not. In our previous study PPD test response was significantly smaller in asthmatic patients, now in this study relationship between BCG scar and asthma is investigated.

Methods: 100 patients younger than 5 years old, vaccinated at birth, with variable severity of asthma were compared to the same number of age-adjusted healthy control group in a case-control study. Mean of vertical and longitudinal diameter was determined. At the same time, the severity of asthma in the case group was evaluated (according to classification of national institutes of health, national institutes of heart lung and blood), by asking mothers about the frequencies of night and day symptoms or intervals of attacks. The patients were divided to three groups: Mild, Moderate and Severe groups (Mild intermittent and mild persistent groups were classified as one group).

Results: In 97 case group the mean size of BCG scar was 4.93mm +_1.89mm, again in 97 control group the mean size was 4.94mm + _1.46mm (P = 0.949).

Conclusion: Against the result of our previous research which showed a significant reverse relation between PPD-skin test response and asthma, but no significant relation was found between BCG scar and asthma, this may indicates that BCG-scar is not a good indicator of TH1 response and is probably related to other mechanism such as wound healing.

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Asthma control test (ACT), FeNO and FEV1 correlation in asthma control

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Backround: The ACT is a validated, 5-item questionnaire with a total score ranging from 5 (poor control) to 25 (total control) (Nathan RA et al, JACI 2004).

Aim: The correlations of ACT, FeNO and FEV1 were evaluated in not well controlled asthma patients for a three month follow-up period.

Methods: ACT, FeNO and FEV1 are measured in 77 female and 20 male persistent asthma patients followed in outpatient clinics of asthma. In 32 of them with ACT<20 and/or FeNO >25ppb and/or FEV1<80% add-on therapy were given and after 3 months, all parameters were remeasured. The correlation of those parameters before and after add-on therapy were evaluated.

Results: Mean ACT scores, FeNO and FEV1 before (period 1) and after (period 2) add-on therapy of 23 female and 9 male asthma patients with a mean age of 40,1±13,3 are in the table. In 16 (50%) of the patients all three parameters, in 6 (19%) FeNO and ACT, in 3 (9%) only FeNO, in other 3 (9%) ACT are under normal values. In the remaining 4 patients comorbid diseases are found as the reason of uncontrolled asthma.

	ACT scores	FeNO (ppb)	FEV1
Period 1	18,9±5,3	32,1±17,8	80,3±17,6
Period 2	20,6±3,6	23,4±13,4	81,9±16,8

As a result; increases in all three parameters are significantly well correlated.

Conclusion: We conclude that because these three parameters are well correlated with each other, ACT by itself may be sufficient to observe the patients alone although it would be best to measure all 3 parameters if they are available.

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Global approach to the psychological aspects of the child with asthma

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Background: Although a significant incidence of various psychological variables has been recognized for a long time in asthmatic children, its true role is the subject of strong controversies. Stress, suggestion and external conditioning can affect the respiratory tract. The chronic aspect of the disease, its disruptive effects on the quality of life and the sensation of threat to their life, can condition the appearance of different psychosocial alterations in the patient, or in their immediate family environment. Experimental tests have shown that the severity of the disease, multiple hospitalizations and the absence of specific IgE mediated symptoms, upon exposure to allergens, could contribute to increased emotional disorders in children.

Objective: To search for an efficient intervention procedure which includes the improvement of these children in school and personal, familial and social adaptation, and to help them modify their inadequate attributions and attitudes towards their disease.

Materials and Methods: This was a random stratified prospective study including a population of 168 pediatric patients with ages comprised between 9 and 11 years of age (100 boys and 68 girls); 74 had asthma, 56 rhinitis and 38 asthma and rhinitis. Patients were divided into active and control groups. The following tests were used: multifactorial autoevaluation test for infantile adaptation (TAMA), questionnaire for attributions and attitudes toward the disease (ATAC), and a questionnaire for the evaluation of the physical aspects of the disease (EVEO). The program applied to the case group consisted of an emotional/instructive program (PIE_ASA).

Results: The psychotherapeutic approach to the treatment of these asthmatic children using the program PIE_ASA achieved a global improvement of the children, fulfilling the objectives of the program. The modification of the negative attributions and attitudes towards their disease was accomplished. No changes in other attitudes such as increase food intake and compliance with medication were obtained. Better results were obtained in boys than in girls, regarding social and school adaptation aspects. However, girls showed a reduction in the number of emergency room visits.

Conclusion: We believe that the use of this type of evaluation en asthmatic children is recommended, since it contributes to an overall improvement in the asthmatic symptoms of children.

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Autoimmunte uveitis, 16 years experience (1988-2004) at Instituto nacional de pediatria in Mexico city

Mario Rafael Duarte-Abdala

Introduction: Autoimmune ocular diseases can be divided in two categories:

external diseases that affect to the cornea and the conjunctiva and internal disease that is denominated uveitis. The term "uveitis" encompasses a wide range of intraocular inflammatory disorders primarily or secondarily involving iris, ciliary body or choroids. Later this term was used to describe any intraocular inflammation that affects the uvea, retina, sclera and vitreous humor.

There are few reports of the disease in latin children.

Materials and Methods: The records of 59 patients with the diagnosis of uveitis, younger than 18 years treated in the service of Immunology at the Instituto Nacional de Pediatria in Mexico City from 1988 to 2004 were reviewed retrospectively.

Results: Among 59 children with uveitis 37 (62.7%) were boys and 22 (37.3%) were girls. Mean age was 8,7 years (standard deviation, 3.8, range 2–17). Most frequent affected site was anterior uveitis in 19 (32.2%) patients, intermediate uveitis in 18 (30.5%), panuveitis in 15 (25.4%) and posterior uveitis in 7 (11.9%) patients. The process was bilateral in 36 (61%), Unilateral right eye affection was established in 13 (22%) patients, and unilateral left eye affection in 10 (17%) patients. Familiar history was present in 4 (6.8%). Uveitis was associated with systemic disease in 18 (30.5%), non-associated with systemic disease in 4 patients (6.8%). Systemic diseases were juvenile chronic arthritis in 9 (15.3%), spondylitis in 3 (5.1%), Vogt Kayanagi Harada disease, nodosa poliarteritis, Wegener granulomatosis, sarcoidosis in 1(1.7%). Infectious etiology agent was identified in 4(6.8%) patients. Clinical signs described were diminution of the visual sharpness in 42 (71.2%), ocular pain in 11 (18.6%), fotofobia in 13 (22%), red eye in 28 (47.5%). We found antibodies antinuclear positive in 17 patients, but with weak pattern. Immunosupresor treatment used was cyclophosphamide in 50 patients, azatyoprin in 8 patients; methotrexate was used in 11 children. Complications included synechias in 8, cataracts and keratopathy in 6 patients. 11 patients left the treatment (18.6%).

Conculsion: The clinical and demographic characteristics of the presentation of this disease in latin children in a hospital of third referral hospital are described.

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Respiratory allergic diseases: Italian monitoring study of GINA and ARIA guidelines (ARGA)

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Background: Asthma is a chronic inflammatory airway disease closely associated with atopic diseases like allergic rhinitis. Both diseases are increasing to epidemic proportion, with increasing medical costs, with a reduced patients' quality of life (QoL) and lower productivity.

The correct management of asthmatic and rhinitic patients would be ensured by the use of international World Health Organization (WHO) guidelines as GINA (Global Initiative for Asthma) and ARIA (Allergic Rhinitis and its Impact on Asthma). Many studies prove that the guidelines recommendations are not or only partially applied within the clinical practice.

Objective: To evaluate the applicability of the GINA and ARIA guidelines and their impact on patients' QoL in General Practice; to evaluate the short and long term efficacy of a Continuing Medical Education (CME) intervention for General Practitioners (GPs) in Italy.

Methods: Prospective observational study in different Italian areas (North, Centre, South-Islands). 168 GPs (71 attended an educational intervention and 97 didn't attend it) will enrol their patients with diagnosis of asthma/rhinitis and with prescription of anti-asthmatic or anti-rhinitic drugs or asthmatic/rhinitic-like symptoms in the previous 12 months.

The GPs and their patients will fill in a questionnaire about respiratory allergic diseases and adverse drug events. The patient will fill in also the Rhinasthma questionnaire about the QoL and the Asthma Control Test. A follow-up will be done after 12 months.

Expected Results: 1. epidemiological data on asthma and rhinitis burden; 2. evaluation of applicability, utility and adherence to GINA and ARIA guidelines by GPs and patients; 3. short term and long term efficacy of a CME intervention; 4. evaluation of prescriptions appropriateness and treatment cost/efficacy ratio.

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Respiratory and psychiatric comorbidity in an Italian primary care population: *EPIDEA study

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Aim: To evaluate the relationship between psychiatric disorders and respiratory diseases (chronic obstructive pulmonary diseases (COPD) and asthma) in an Italian primary care population.

Methods: Cross-sectional survey carried out in 6 Italian geographical areas. 2083 subjects (68.1% of expected sample, mean age 44 + 13 yrs), randomly selected by 143 (70.1%) general practitioners, participated in the study. Each subject answered to a physician-administered interview, based on the standardized National Research Council questionnaire, for the detection of psychiatric disturbances, respiratory diseases and symptoms, relative risk factors and pharmacological treatments, and filled out the General Health Questionnaire (GHQ).

Results: Prevalence of current and lifetime psychiatric disturbances (affective disorders and anxiety) were 27.5 and 46.0%, respectively. COPD and asthma were significantly associated with the presence of anxiety alone and, above all, with the co-presence of anxiety and depression.

By logistic regression models adjusted for independent effects of age, area, smoking, education, working position, GHQ score, and sex, COPD resulted associated with anxiety and depression in combination (OR: 2.8, CI: 1.8–4.6) and with anxiety alone (OR: 2.6, CI: 1.7–4.1). Asthma was also associated with co-presence of anxiety and depression (OR: 2.2, CI: 1.3–3.8) and with anxiety alone (OR: 2.1, CI 1.2–3.5).

Conclusion: EPIDEA study confirms the large diffusion of psychiatric disorders in an Italian primary care population. The association between respiratory diseases and affective and anxiety disorders suggests to take into account these aspects in the clinical practice of pneumologists, allergologists and psychiatrists.

ANAPHYLAXIS

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Exercise-induced anaphylaxis related to Cannabis sativa smoke exposure

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Background: Exercise-induced anaphylaxis is a life-threatening condition that can be food-dependent (specific or non-specific) and drug-dependent; sometimes the only trigger is exercise. We report here a case in which the trigger of the episode was the previous smoking of marijuana (*Cannabis sativa*) cigarettes.

The patient, a 23 years-old-man, had previosly been diagnosed of cereal-dependent, exercise-induced anaphylaxis. In spite of strict avoidance of these foods, he had recurrent episodes of anaphylaxis during exercise (jogging). When he was reevaluated he recognized he used to smoke marijuana cigarettes very often, sometimes before exercise. 2 years before this evaluation he begun presenting sudden onset rhinoconjuctivitis each

time he smoked a marijuana cigarette. He gave up marijuana smoking; since then he hasnát had any other anaphylactic reactions. He has been jogging, eating cereals before exercising, without any problem for more than one year.
Methods: Skin tests (prick-prick test) with fresh *Cannabis sativa* leaves, skin tests to a leave extract, IgE immunoblotting to a *Cannabis sativa* leaves extract.
Results: Skin tests were positive, while they were negative in 10 controls On the immunoblotting, the patient's IgE recognized a 35 KD band.
Conclusion: We report here the first case of exercise-induced anaphylaxis in which the trigger is the inhalation of *Cannabis sativa* smoke. The skin tests and *in vitro* results, as well as the clinical picture and evolution after smoke cessation are consistent with a "*Cannabis sativa* smoke dependent, exercise-induced anaphylaxis", in a patient that also has a IgE mediated immediate rhinoconjuctivitis due to marijuana smoking. The mechanisms of the anaphylactic reaction are obscure; there are a few previous reports that confirm that *Cannabis sativa* smoking can cause immediate hypersensitivity symptoms, like bronchospasm, rhinoconjuctivitis or generalized itching in sensitized individuals, but to our knowledge there are no previous reports of an exercise-induced reaction that appears to be dependent on marijuana smoking.

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The usefulness of serum tryptase in the diagnosis of shrimp anaphylaxis in children

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Background: Anaphylaxis is a life-threatening allergic reaction, and foods are one of the most common culprits. Serum tryptase is marker of mast cell activation and could use to confirm anaphylaxis to shrimp.
Objective: To determine the utility of serum tryptase in the diagnosis of shrimp-induced anaphylaxis.
Methods: Twenty-one patients with previous allergic reaction from shrimp were recruited into a prospective study for shrimp challenge. Twelve patients developed mild allergic reaction and nine patients developed anaphylaxis. Serum tryptase were obtained before shrimp challenge and 1 hour after the onset of symptoms.
Results: In both groups of patients, median tryptase levels were significantly elevated after the onset of shrimp challenge as compared to baselines (baseline tryptase - 1.08, peak tryptase - 2.33 μg/L in anaphylaxis group; baseline tryptase -1.49, peak tryptase 1.56 μg/L in non-anaphylaxis group, $p < 0.05$). The delta tryptase (peak minus baseline) values in the anaphylaxis group was significantly higher than non-anaphylaxis group (1.33 VS 0.125 μg/L, $p = 0.0004$). The tryptase ratio (peak divided by baseline) values in the anaphylaxis group was also significantly higher than non-anaphylaxis group (2.11 VS 1.055, $p = 0.0001$). Using the recommended cut-off range (peak tryptase < 12.0 μg/L) the sensitivity of such cut-off was 0.11 with specificity of 1.0.
Conclusion: We recommend using serial tryptase values, including tryptase ratio and/or delta tryptase values, for the diagnosis of food-induced anaphylaxis. The latter two values may be helpful than peak serum tryptase.

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Anaphylaxis to peach

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Food is considered the most common cause of anaphylaxis in children. The common cause of food allergy and anaphylaxis are cow's milk, hen's egg white, wheat, peanut, tree nuts, fish, shellfish and soya. Fresh fruits is also common especially peach (*prunus persica*).
Case: 8 years old boy presented to emergency room with generalized itchy erythematous skin rash all over the body after 30 min. of eating peach. There was no history of having medication, new food or an insect bite. There were no history of shortness of breath, wheezing, stridor, abdominal pain, vomitting, swelling of tongue or mucus membrane or loss of consciousness. the was significant hypotension noticed during obtaining the vital signs as low as 92/27 mmHg, lasted for about 24 hrs instead of 2 appropriate doses og adrenaline injections, chlorpheneramine maleate IV and intravenous fluids.
Other history: No atopic dermatitis, allergic rhinitis, asthma. Breast-feded for 2 years and recieved goat milk as a child, mother had allergic rhinitis, father and other 5 siblings are healthy.

This child continue to have oral allergy symptoms to accidental peach ingestion in small amounts after the anaphylaxis incident, but never anaphylaxis again to peach or other fruits or highly allergenic foods.
Conclusion: Rosaceae fruit allergy (typically peach) is a true fruit allergy. The spectrum of symptoms range from oral allergy syndrome to rarely anaphylaxis case, as in our patient. Anaphylaxis can be the initial presentation of fresh fruit allergy even if it was consumed before. Avoidance and epinephrine auto-injectors can be an appropriate management.

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Epidemiology of Anaphylaxis among children and adolescents in Korea

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Background: There was little information studied about the epidemiology of anaphylaxis among children and adolescents in Korea until now. We identified the epidemiology of anaphylaxis in Korea and established the on-line registration of anaphylactic cases through the official internet site of Korean Academy of Pediatric Allergy and Respiratory Disease (KAPARD).
Methods: We used the sample data from two sources. One from the National Health Insurance Review Agency (NHIRA), and the other from the questionnaire by the pediatric allergy specialists in the university hospitals, all from the KAPARD. The data from the NHIRA was based on the diagnosis codes, specific for anaphylaxis and other related diagnoses between 2001 and 2005.
Results: According to the data from NHIRA, the incidence of anaphylaxis was 0.8–2.9 episodes per 100,000 person-years. There was no increase in incidence between 2001–2005.0The causes of anaphylaxis were identified as 45% for unknown cause, 25% for insect, 15% for food, 10% for drug, 1% for serum. According to the questionnaire from KAPARD, we identified 88 episodes of anaphylaxis. The causes of anaphylaxis were identified as 40% for food, 20% for drug, food and exercise for 5%, exercise for 1%, bathing for 4%, bee sting for 1%, unknown for 17%. We could not identifty the incidence and the increase of anaphylaxis.
Conclusion: Further research and more reliable databases are necessary to establish the epidemiology of anaphylaxis in Korea. We also encourage the registration of anaphylaxis for pediatricians in the future.

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Wheat-dependent, exercise-induced anaphylaxis: A report of 5 cases among Thai children

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Background: Wheat is not an uncommon cause of food-dependent, exercise-induced anaphylaxis. The aim of this study was to describe common clinical characteristics, laboratory manifestations and natural history of the disease.
Methods: Children with history of wheat-dependent, exercise-induced anaphylaxis were identified. Skin prick test and specific IgE for wheat were done. A three-day challenge program including open challenge for wheat, exercise and exercise challenge test after a meal containing wheat was performed.
Results: Five children, aged 5-14 years (mean: 8 + 3.74 years) were evaluated. Atopic history was found in 40% of patients. All patients had symptoms involved skin and respiratory systems and two had hypotension. Sera specific IgE for wheat were determined in 3 patients among which two were positive. Three patients completed the three-day protocol. Anaphylaxis occurred in 2 out of 3 patients with the amount of wheat consumed prior to exercise being more than 100 grams.
Conclusion: Wheat-dependent, exercise-induced anaphylaxis is more common in male. Skin and respiratory symptoms are major manifestations. A three-day wheat challenge protocol is a definitive diagnostic tool. However the amount of wheat required for challenging should be high.

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Anaphylaxis due to fish hypersensitivity in an exclusively breastfed infant

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Two cases of anaphylaxis in exclusively breastfeeding are reported.
Case1: 5-month-old boy with exclusive breastfeeding suffered from eczema since 2-month-old and his mother restricted intake of egg and cow's milk since 4 month. He was hospitalized with tarry stools at 5 month of age. Within an hour after breastfeeding after his mother had taken raw fish, he developed whole-body rash three times. Skin prick test was positive to horse mackerel, mackerel, salmon and codfish. After his mother restricted fish ingestion no symptom was observed while breast feeding was continued.
Case2: 9-month-old boy with exclusive breastfeeding presented severe eczema at 1 month of age. Thereafter his mother and he avoided eggs, cow's milk, soy, and fishes. At 9 month, he was fed almost breast-fed and ate little solid food. One day he developed rash and cough soon after mother fed him breast milk. He was breast-fed again and 3 hours later presented vomiting and rash over whole body and was hospitalized. Blood examination revealed high titer of specific IgE to egg, cow's milk, sardine and codfish in his serum. Skin prick test was also positive to sardine and eggs. Only small amount of allergens was detectable in breast milk. Although food their mother took was not determined, anaphylaxis due to food allergens in breast milk was most suspected. Systemic immediate type food allergy due to small amounts of food allergens included in breast milk is rare and so far a few case has been reported.

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Anaphylaxis in Chile: a 10-year review

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Background: Anaphylaxis is a severe acute allergic reaction, quickly progressive and potentially fatal, triggered mainly by food antigens and drugs. No prevalence studies of this condition have been conducted in our country, neither of the clinical presentation and/or etiology.
Objective: Determine the clinical characteristics of patients with anaphylactic reactions seen at the Clinical Hospital of the University of Chile. Patients and Methods: A retrospective review of medical records spanning 10 years (May 1997- May 2007) allowed us to obtain data of 127 patients who where derived to the Allergy and Immunology Department of the Clinical Hospital of the University of Chile with a diagnosis of anaphylaxis of unknown origin. Three patients were excluded from this study, two with diagnosis of systemic mastocytosis and the other with hereditary angioedema. We evaluated: sex, age, number of episodes before consulting, interval between first and last episode, history of atopy, time elapsed between exposure to the suspected triggering factor and the appearance of symptoms, form of presentation, severity of the reaction, treatment received and clinical/laboratory work up.
Results: 127 patients, 48 men and 79 women, were included. Age ranged from 11 months to 81 years, with a mean of 27.5 years; 61% were atopic. The average delay before consulting was 4 years, and median number of previous episodes was 2. Etiology was identified in 83% of cases; the main causes were drugs (31%), followed by foods (27%) and Hymenoptera stings (16%). The average time of onset of symptoms was 42 minutes post-exposure. Ninety six percent of patients presented urticaria and/or angioedema and severe manifestations (Grade III and IV) were seen in 70% of the individuals. From the individuals who had indication of management with epinephrine, only 42.1% received it in the Emergency room.
Conclusion: The clinical characteristics of the Chilean patients with anaphylaxis are similar to those reported in other countries. It is relevant to emphasize the great percentage of atopics patients in our series as well as the delay in the consultation.

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Anaphylaxis in Singapore children

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Background: To study the epidemiology and review the treatment of anaphylaxis in Asian Singapore children.
Methods: This is a 2-year retrospective study, which includes all patients with anaphylaxis seen in the KK Children's Hospital, Singapore. Cases were identified by the relevant discharge codes from the Children's Emergency (CE) and hospital's inpatient records, and cross-checked with referrals to the Allergy clinic.
Results: 30 children with anaphylaxis were identified. 19 (63%) were male. 3 (10%) were less than 1 year old at the time of presentation, 8 (27%) were aged 1–5, 11 (36%) were 6–11, and 8 (27%) were 12–16 years. Race distribution corresponded to population demographics. 25 (83%) had features of atopy.

Food triggers were reported in 17 (57%), but food specific IgE sensitization was demonstrated only in 11 children (37%). [Egg (4), peanut (4), cow's milk (1), clam (1), chinese pear (1)]. There were 11 (37%) cases secondary to drugs: [NSAIDs (4), paracetamol (3), traditional Chinese medication (1), asparaginase (1), desferrioxamine (1) and blood product (1)]. Two patients had no identifiable triggers and were diagnosed as idiopathic anaphylaxis.

Most patients presented with dermatologic (87%) and respiratory features (83%). 8 (27%) children had vomiting or abdominal pain, and 8 (27%) had hypotension. There were no fatalities in this study.

Of the 23 (77%) patients that presented to the CE, 74% were admitted for observation. 3 patients required admission to the intensive care unit because of hypotension requiring fluid resuscitation at presentation.

12 (40%) children were treated with epinephrine, but this was administered mostly by the subcutaneous route. 18 (60%) were given antihistamines; 22 (73%) steroids; 17 (57%) bronchodilator therapy; and 9 intravenous fluid boluses.

6 children were equipped with self-injectable epinephrine. Those who did not receive self-injectable epinephrine included cases with drug triggers, patients who defaulted, and parental refusal due to anxiety over self-injection or cost concerns.

Conclusion: Anaphylaxis is predominantly food triggered in Singapore children, with a noticeable increase in peanut food allergy. There is also a significant number of cases attributed to ibuprofen and paracetamol. Whilst epinephrine is the cornerstone therapy for anaphylaxis only 40% of our cases received this treatment. There is an urgent need to improve the awareness and treatment of anaphylaxis in our population.

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The angiotensine converting enzyme activity in patients with an anaphylactic reaction after Hymenoptera sting event

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The renin angiotensine system seems to play a significant role in the response to anaphylactic reaction.

The aim of the study was to evaluate the serum activity of angiotensine converting enzyme (ACE) in patients with anaphylaxis after Hymenoptera sting event depending on the polymorphism of ACE gene and specific IgE.

Materials: The study group consisted of 31 patients, mean age 50±12 years. There were 21 patients with specific IgE to wasp venom, 6 to bee venom and 4 to both venoms.

Methods: The ACE activity was measured according to the method described by Liberman I/D ACE polymorphism was determined based in PCR technique employing insert specific- primers.

Specific IgE was measured with Allergopharma specific IgE ELISA Kit, Germany.

Results: The ACE activity in studied group was 29,11±11,5 IU. Table 1 contains the ACE activity depending of the ACE gene polymorphism.

	n	ACE activity (IU)
DD	9	38,6± 9,1 *
ID	16	28,3±9,7 *
II	6	17,1± 6,1 *

*p < 0,01 Table 2 contains the ACE activity depending of the specific IgE to Hymenoptera.

	n	ACE activity (IU)
wasp	21	30,5± 11,4 **
bee	6	23,6±12,6**
wasp/bee	4	30,0± 10,8 **

**p > 0.01

Conclusion: 1. The serum activity of ACE in the patients with anaphylactic reaction to Hymenoptera is the same as in general population 2. There are significant statistical differences between ACE activity in groups according to the ACE gene polymorphism 3.The specific IgE does not influence on the ACE activity

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I/D ACE gene polymorphism in patients with anaphylaxis after Hymenoptera sting

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The activity of the ACE may play an important role in the anaphylactiv reaction. The ACE activity is genetically determinated. The DD genotype is correlated with arterial hypertension and other vascular disorders. The ID polymorphism in the Caucasian population is 0,53.

The aim of the study was to analyze the frequency of angiotensine converting enzyme gene polymorphism in patients with documented history of anaphylactic reaction to a Hymenoptera sting.

Materials: The study group consisted of 48 patients (38F, 16 M), mean age 48±14 years. There were 29 patients with specific IgE to wasp venom, 7 to bee venom and 12 to both venoms. The control group was 23 healthy person (9F, 14M), mean age 41±7 years.

Methods: I/D ACE polymorphism was determined based in PCR technique employing inser-specific primers. Specific IgE was measured by Allergopharma specific IgE ELISA, Germany.

Results: The ACE gene polymorphism in the control group were: DD n=5 (21.7%), ID n=15 (65.2%), II n=3 (13.1%) The ACE gene polymorphism in the study group were: DD n=16 (33.3%), ID n=22 (45.85%), II n=10 (20.85%)

ACE genotype n(%)	wasp n=29	bee n=7	wasp/bee n=12
DD	9 (31.0)	0	7 (58.3)
ID	16 (55.2)	6 (85.7)	0
II	4 (13.8)	1 (14.3)	5 (41.7)

Conclusion: In the group of patients with documented anaphylactic reaction to Hymenoptera sting there were no differences between the ACE gene polymorphism and general population.

We did not observed differences of this polymorphism between the wasp and bee group, but in the wasp/bee one the allel I was more frequent, and therefore in larger groups of seems justified.

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Milk and wheat: the most common cause of anaphylaxis in children

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Background: Anaphylaxis is a medical emergency requiring immediate recognition and treatment. We report common findings of the first case series of anaphylaxis in Iran, which carried out in children who referred to the center of Immunology, Asthma and Allergy.

Materials and Methods: Children referred to the clinic of allergy with a previous diagnosis of anaphylaxis were considered in this study (2003 - 2006). A specific questionnaire was completed for all of the patients and a detailed clinical history and demographic data were recorded. For all of the patients, Skin prick tests and specific IgE measurement were performed for suspected allergens. Determination of causative allergen was based on patients' history and IgE mediated tests. Challenge test was not performed for any patient.

Results: 39 children ages ranged from 1m to 13 year old with a previous diagnosis of anaphylaxis were referred to the clinic of allergy during 2 years. (48/1% male and 24/1% female respectively). Skin prick tests was positive in 78/9% and specific IgE was positive in 88/2%. The most probable causative agents in this population were foods (92/40%). Milk (55/6%) and wheat (18/5%) were the most frequent causes of anaphylaxis in this study.58% of children had more than 2 episodes of anaphylaxis in their life; also 6 cases of multifactorial food anaphylaxis were identified for the first time. The most common causative allergen for recurrence anaphylaxis was milk in 20 patients Dermatologic signs and symptoms were the most common (92/2%) vs. 76/%, 62/5%, 21/6%, 20/8% for respiratory, gastrointestinal, neurological and cardiovascular signs and symptoms,

respectively. Flushing was seen in 66/7%, prurities in 56/9%, urticaria in 54/9%, preorbital edema in 52/1% of patients, rash in 50%, erythema in 43/1% and angioedema in 41/2%.

Conclusion: These data confirm that foods specially milk and wheat are the most common causes of anaphylaxis in children and dermatologic signs and symptoms are the most common finding, thus parents and physicians should be educated to recognize the most common causative allergens and common presenting clinical manifestation of this disease to avoid the progression of this life threatening syndrome and be able to manage anaphylactic patients.

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Food anaphylaxis: a clinical study

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Background: Food allergy is one of the most common causes of anaphylaxis.

Objective: The aim of this study was to characterize a group of patients with food anaphylaxis followed-up in a food allergy outpatient department during a one-year period.

Methods: Twenty-two consecutive patients observed in our department from June 2006 to May 2007 with a suggestive clinical history of food anaphylaxis were selected. Foods involved in the severe systemic reactions were identified and atopic co-morbidities, occurrence of anaphylaxis in the study period and the medication used were investigated. A standardized questionnaire and skin prick tests to airborne allergens (mites, moulds, cockroach, cat and dog dander, pollens, feathers, latex) and food allergens (egg, milk, seafood, mammals and birds meat, fruits, nuts, cereals, legumes, spices and other vegetables) were performed. Serum specific IgE to airborne and food allergens and/or prick-to-prick with natural food extracts were done in selected cases (if there was evidence of clinical reactivity).

Results: All the patients selected had a past history of an immediate severe systemic reaction after ingestion of food. The mean age of the population studied was 37±10 years. Fourteen patients were female. The mean age at the first anaphylaxis episode was 24±11 years. Foods involved were fruits in 4 patients, crustaceans in 4 patients, cow's milk in 4 patients, nuts in 3 patients, legumes in 2 patients, cereals in 2 patients, fish in 2 patients and egg in 1 patient. All patients were sensitised to the culprit food. Sensitisation to airborne allergens occurred in 19 patients (10 to mites, 7 to grasses, 5 each to weeds and trees, 3 to latex and 1 each to cockroach, dog dander and feathers). Sensitisation to more than 1 food occurred in 16 patients. Eighteen patients had atopic co-morbidities related to airborne allergens (12 patients had asthma). Recurrence of anaphylaxis occurred in 2 patients with cow's milk allergy in the study period. Self-administered adrenaline was used by 1 patient twice.

Conclusion: In the population studied a high proportion of atopic asthmatic patients was found. Different plant and animal foods were implicated. Hidden milk allergens can be extremely difficult to avoid and this fact could explain the recurrence of anaphylaxis in 2 cow's milk allergic patients, in spite of a strictly restrictive diet.

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Two cases of anaphylaxis caused by octopus variabilis

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Octopus variabilis, a kind of Mollusca, is a favorite food in oriental countries. There have been no published report of anaphylaxis caused by octopus and the mechanism of this food allergy has not been understood. We experienced two cases of anaphylaxis developed afteringestion of raw octopus.

The first patient, a 52-year-old man without any previous allergic diseases, experienced generalized urticaria, dizziness, and loss of consciousness at one hour after ingestion of raw octopus. The skin prick test showed weakly responses to octopus extract, *D. pteronyssius*, *D. farinae, oyster*, shrimp (A/H ratio was all 2+). Serum specific IgE antibody to octopus extract was undetectable by ELISA, whereas increased level serum specific IgG4 antibody to octopus extract was noted by ELISA.

The second patient, a 51-year-old woman with allergic rhinitis and allergic conjunctivitis had suffered from recurrent abdominal pain, chest discomfort and dizziness for 5 min after ingestion of steamed or raw octopus. The skin prick test showed negative response to octopus extract, but positive responses to *D. pteronyssius* and, *D. farinae*. Serum specific IgE, IgG1 and IgG4 antibodies to octopus extract were not found by ELISA.

These finding suggest that octopus could induce anaphylaxis via non-IgE mediated mast cell activation.

Key words: Anaphylaxis, Octopus variabilis

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Anaphylaxis: finding the causative agents in a university hospital

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Background: Anaphylaxis is an emergency condition. Attempts should be made to find the causative agents for further prevention.

Methods: We prospectively studied 28 patients with anaphylaxis at Siriraj Hospital, Thailand from January 1st, 2003 to July 31st, 2004. Direct patient interviews and physical examinations were done. Skin prick tests and challenges to the agents according to history, were carefully performed to identify the causes of anaphylaxis.

Results: The causes of anaphylaxis were identified by positive skin test in 24/28 patients. In these 24 patients, oral challenge test and specific IgE wereperformed and found positive in 3/3 and 4/4 patients, respectively. The agents causing positive skin tests were ant (7 patients), seawater shrimp (4patients), immunotherapy (3 patients), wheat (2 patients), wasp (2 patients), seawater mollusk (2 patients), seawater crab (1 patients), wheat-dependent exercise-induced anaphylaxis (WDEIA, 1 patient), cow's milk (1 patient), andciprofloxacin (1 patient). Four cases had negative skin test. In these patients, the causative agents could not be identified in 3 cases and the patientswere diagnosed with idiopathic anaphylaxis. One patient had skin test negative to erythromycin but positive oral challenge to erythromycin. The causes of positive oral challenges in 3 patients were seawater shrimp, seawater mollusk and WDEIA (1 patient each). The causes of positive specific IgE wereseawater shrimp, wheat, cow milk and ant (1 patient each). No patient developed severe anaphylaxis during the tests.

Conclusion: Detailed history taking remains important to find the cause of anaphylaxis. However, skin tests and/or challenges are helpful tools if they are done carefully under a controlled condition.

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Anaphylaxis caused by light, a case report

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We present the case of a woman, 36 years old, who smokes around 20 cigarettes a day, health worker, with a family history of atopy and a personal history of tonsillectomy, left renal colic and habitual consumption of oral contraceptives.

From the last five years she has been presenting episodes of urticaria and occasionally angioedema, immediately after the solar exposure (at any season of the year), located only in the exposed areas. The injuries remain for 2 to 3 hours and disappear without leaving any residual injury.

During three times she suffered head instability and afterwards loss of consciousness, twice because of a heavy solar exposure (despite of having applied sunscreen cream previously) and another time after reciveing an UVA's bronzed session.

Laboratory findings, including complete blood cell count, erithrocite sedimentation rate, lactate dehydrogenase, renal and liver function, glucose, immunoglobulins A, G, M, tryptase, complement C3, C4, rheumatoid factor, total proteins and C-reactive protein did not reveal any pathological values. Test for antinuclear, anti –ENA, anti- mithocondrials, anti-muscular smooth, anti- parietal cell antibodies were negative. The total IgE was 1000 UI/ mililiter. The 24 hours urine and faeces test analysis in order to detect porphyrins were negative.

Skin prick test with a set of common air-allergens were positive for house dust mites. Photobiological examination: urticarial lesions were caused after unique exposure to UVB and to UVA irradiation, but not with the visible light spectrum.

Although the minimal urticaria dose was not determined, the evolution of this case show an extreme seriousness. She continues with clinic symptoms in the area of exposure few minutes after minimal solar exposures in winter (although she has not presented new episodes of anaphylaxis) in spite of the continued treatment: topical broad spectrum sunscreen daily and all through the year, different systemic H1 antihistamines (cetirizine 30 mg/day, hidroxicina, ebastine, desloratadine), beta carotene supplementation, natural desensitisation approach by regular sun exposure.

We suspect this is a case of solar urticaria that, clinically changed into an anaphylaxis when receiving very high doses of radiation.

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Common insects causing anphylaxis in Thailand

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Stinging insects comprised approximately 10 percent of the causes of anaphylaxis in Thailand. We live in a tropical climate, so some of the temperate inhabited insects do not exist in Thailand, such as yellow jackets (Vespula spp.), white faced hornet (Dolichovespula maculata), and yellow hornet (Dolichovespula arenaria). The poster presented pictures and brief description of common insects that have a potential of causing anaphylaxis in Thailand, such as tropical wasps, paper wasps, Giant bee, Indian bee, Small bee, bumble bee, venomous ants, and kissing bugs. In conclusion, the study described the species and characteristics of common stinging insects that can cause anaphylaxis in Thailand.

INFLAMMATORY CELLS & MEDIATORS

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Do mast cell numbers differ in different inflammatory periodontal diseases?

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Background: Mast cells play an important role in allergic reaction, host defense, local homeostasis, inflammation and angiogenesis. The objective of this study was to evaluate the relationship between mast cell numbers and different types of periodontal diseases.

Methods: Gingival specimens were taken from 20 moderate to advanced chronic periodontal and 19 moderate to advanced aggressive periodontal sites as case groups and 18 healthy/gingivitis sites as control group in routine periodontal surgeries (flap and crown lengthening) and were examined after toluidine-blue staining for mast cells counting and hematoxylin-eosin staining for assessing inflammation. Inflammatory and mast cells in 5 micron sections were assessed by two observers 3 times, utilizing light microscope at 100X and 400X magnification. ANOVA and T tests with an alpha error level less than 5 percent were used to analyze data.

Results: Mast cells numbers were higher in chronic versus aggressive periodontitis and healthy/gingivitis ($p=0.000$). The aggressive periodontitis didn't have more numbers of mast cells as compared to healthy/gingivitis ($p>0.05$). There were no relationship between mast cell numbers and degree of inflammation in 3 groups.

Conclusion: The present study indicates more mast cell numbers presence in the chronic periodontitis sites than other sites. The results of this study suggest more studies to evaluate dynamic aspects of host defense in conjunct with other aspects of immune system, simultaneously.

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Exogenous nitric oxide regulates cyclooxygenase-2 expression and prostaglandin D_2 generation in mast cells

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Background: Mast cells (MC) are important effector cells in allergic and inflammatory responses through secretion of various mediators following activation. Nitric oxide (NO) is important signaling molecule that regulates MC function. It can depress MC allergic responses such as leukotriene (LT), cytokine and chemokine production, as well as MC degranulation. However, the involvement of NO in prostaglandin (PG) D_2 production, an important lipid mediator produced in MC, is unclear. In PG synthesis, cyclooxygenase (COX) is an important enzyme and two isozymes of COX, constitutively expressed COX-1 and inducible COX-2, have been reported. It is well established that mouse bone marrow-derived mast cells (BMMC) exhibit biphasic PGD_2 biosynthesis; COX-1-dependent immediate and COX-2-dependent delayed PGD_2 production, when BMMC are stimulated with SCF, IL-10 and IL-1β.

Methods: The effects of NO on COX-2 expression and PGD_2 generation in BMMC were investigated using NO-donors, S-Nitrosoglutathione (SNOG) and S-Nitroso-N-acetylpenicillamine (SNAP). PGD_2 production by stimulated BMMC was assayed using PGD_2-MOX enzyme immunoassay kits. Western blot and real-time RT-PCR were used to measure COX-2 protein and mRNA expression.

Results: Exogenous NO augmented COX-2 protein expression and increased COX-2-dependent PGD_2 generation in response to SCF, IL-10 and IL-1β. The increased expression of COX-2 by NO-donors was reduced by the p38 MAPK inhibitor, SB202190. Downstream of p38 MAPK, NO-donors augmented not only COX-2 mRNA transcription but also its stability. By contrast, NO-donors did not affect COX-1 protein expression. However, in contrast to the augmentation of COX-2 expression and activity, SNOG (100 to 500 μM), but not SNAP (up to 500 μM) inhibited COX-1-dependent PGD_2 generation.

Conclusion: Thus, exogenous NO regulates PGD_2 production by MC through regulation both of COX-1 and COX-2. Furthermore, these findings help us to understand the role of NO in MC function and the regulatory mechanisms of lipid mediator generation in MC in inflammatory diseases.

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Cigarette smoke suppresses the production of cytokines but not chemokines by IgE/Ag activation in mast cells

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Background: Chronic obstructive pulmonary disease (COPD) is a major incurable global health burden and will become the third largest cause of death in the world by 2020. It is currently believed that an exaggerated inflammatory response to inhaled irritants, in particular cigarette smoke (CS), causes progressive airflow limitation. This inflammation, where macrophages and neutrophils are prominent, leads to oxidative stress, emphysema, small airways fibrosis and mucus hypersecretion. Mast cells are important effector cells in anaphylactic reactions and are involved in a variety of immunological and non-immunlogic processes. However, the role of mast cells in pathogenesis of emphysema not yet documented. Recently, we have demonstrated that CSM induces protease expression and chemokine release by primary mast cells. In this study, we further investigated the effects of CSM on the cytokine production of mast cells respect to activation with IgE and antigen.

Materials and Methods: BMMC were cultured from BALB/cBy mice for 3 weeks. Cells were exposed against CSM and activated with IgE and antigen.

Degranulation of cells was assessed by monitoring the release of the granular enzyme beta-hexosaminidase. The amount of cytokines were determined in supernatants and IkB-α\ degradation, P65 and CREP phosphorylation were measured by Western blot.

Results: CSM attenuated degranulation and of cytokines releases of mast cells by a concentration-dependent and non-cytotoxic manner. CSM did not effect on cytokine releases induced by LPS. Moreover, CSM induces phosphorylation of CREB and ATF-1 when activated with IgE and antigen.

Conclusion: Thus, some of the inhibitory effects of CSM on degranulation and cytokine releases may different than chemokine and leukoterine releases.

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The suppressive effect of Houttuynia cordata extract on HMC-1 cell migration in response to stem cell factor

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Hottuynia cordata Thunb. (Saururaceae; HC) is known as a therapeutic drug that has been used in traditional oriental medicine for the treatment of allergic diseases. Mast cells function as regulatory cells in a variety of inflammatory diseases, in particular, asthma and atopic dermatitis. In this study, we examined the effect of HC extracts on the chemotactic activity of the human mast cell line, HMC-1, induced by stem cell factor (SCF). The survival rate of HMC-1 cells was not altered by treatments with the HC extracts at a concentration of 10 μg/ml for 24 h. SCF showed the typical bell-shape curve for the HMC-1 cell chemoattraction with the peak of the curve at the SCF concentration of 100 ng/ml. HC-1, which was the whole plant (Houttuynia cordata) extracted with 80% EtOH, and HC-3, which was the residue successively partitioned with EtOAc, both had inhibitory effects on HMC-1 cell movement. After the treatment with 10 μg/ml HC-1 extract for 6 h and 24 h, the chemotactic index (CI) of HMC-1 cells decreased up to 73% and 63%, respectively. HC-3 extract significantly inhibited the cell movement (72% ± 6% and 44% ± 2%). The HC-1 and HC-3 extracts had no inhibitory effect on the mRNA and protein expressions of c-kit, SCF receptor. SCF tranduces the chemotaxis signaling via NF-kB translocation, and both extracts blocked the activation. Taken together, our results indicate that HC-1 and HC-3 extracts inhibit the chemotactic activity of HMC-1 cells in response to SCF by blocking the NF-kB activation. These substances may be helpful for treating the inflammatory diseases associated with mast cells.

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The effect of IL-3 and SCF mixture on cell proliferation and rat mast cell protease-¥± synthesis of rat bone marrow derived mast cells

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Background: Mast cells are the important immune cells to produce cytokines chemokines and histamine, and closely related to allergic reactions processing. The investigation of mast cell activation enhances the understanding of allergies and requires easy supply of large number of mast cells of culture. Bone marrow derived mast cell (BMMC) are the mast cells to provide sufficient number of cells and are affected by the kinds of cytokine combination and dose both in proliferation and in differentiation. This investigation was about the effect of culture condition on rat BMMC cultivation in serum supplement and cytokines.

Methods: Rat bone marrow cells were cultured with the media conditioned differently by the addition of individual, or mixed cytokines of IL-3 IL-4 IL-6 and SCF. Rat BMMC proliferation was measured using CCk-8 assay. Rat BMMC differentiation was analyzed by the level of RMCP-¥±. Serum supplement effect on rat BMMC culture was investigated by comparison of the media including horse sera with the media carrying rat sera.

Results: In the culture with horse serum supplement, rat BMMC proliferation in the presence of IL-3 (10 ng/ml) mixed with SCF (10 ng/ml) was the highest among the applied cytokine mixture. Rat BMMC with the mixture of cytokine concentration showed relatively high level of RMCP-¥±. In the culture with rat serum supplement, the cytokines condition of IL-3 10ng/ml with SCF 10ng/ml did not induce RMCP-¥± production. However the adjustment of cytokine concentration with the mixture of IL-3 10ngml and SCF 1.25ng/ml enhanced RMCP-¥± synthesis, comparable to the level produced by IL-3 10ng/ml and SCF 10ng/ml in the media supplemented with horse serum.

Conclusion: These results indicated that rat BMMC preparation requires IL-3 and SCF, and that rat BMMC differentiation be affected by the ratio between IL-3 and SCF and by the kind of serum.

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Mast cells involvement in stress-induced epithelial barrier dysfunction in the rat jejunum

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Background: Stress may facilitate intestinal inflammation by promoting epithelial barrier dysfunction allowing luminal antigens to enter the mucosa that may be crucial for the development of food allergy.

Methods: Male WKY rats were submitted to crowding stress (CS) (8 rats/cage) or sham-crowding (2 rats/cage) for 15 days. In addition, ketotifen, a known mast cell stabilizer or saline was injected i.p. at day 14th (1 mg/kg, 2 doses, 12 h interval). At the end of the stress or sham protocols, jejunal segments were mounted in Ussing chambers to measure macromolecular permeability to horseradish peroxidase (HRP). Rat mast cell protease II (RMCPII), an indicator of mast cell activation, was measured in jejunal perfusates and in jejunal homogenates at the end of stress or sham exposure. Plasma corticosterone and CRH (corticotropin releasing hormone) receptor 1 (R1) and 2 (R2) expression in jejunal tissue was measured by an ELISA assay by RT-PCR respectively.

Results: Plasma corticosterone was tree fold increased after the stress protocol in the CS group (CS: 138±27 ng/mL vs. control: 52±16; p=0.0001; n=8/group). CRH-R1 but not CRH-R2 expression was increased in the CS when compared to control. Crowding stress (CS) induced an increase in HRP permeability (CS: 18,2±5 pmol/cm2/h vs. control: 6,2±2.8; p=0.0004;), that was diminished when ketotifen was administered (12,1±5,2 pmol/cm2/h; p=0,015 vs CS; n=8/group). Furthermore, jejunal RMCPII was higher in perfusates from stressed rats (CS: 23.6 ± 5.1 μg/L vs. control: 14.8 ± 9.9; p=0.021; n=8/group) and in tissue homogenates (CS: 456±123 ng/mg of tissue vs. control 274±84; p=0.002: n=8/group) suggesting ongoing mast cell activation. Ketotifen reduced the increased RMCPII in both jejunal perfusates (16,5±7,2 μg/L; p=0,019 vs. CS; n=8/group) and in tissue homogenates

(198±72 ng/mg of tissue; p=0,002 vs. CS; n=8/group). Ketotifen did not induce any change in the parameters evaluated in the control groups.
Conclusion: Stress evoked an epithelial barrier defect that may facilitate the uptake of luminal macromolecules and is mediated by mast cells. This synergistic effect may play an important role in food allergy.

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Importance of matrix metalloproteinase -9 (MMP-9) and its inhibitor (TIMP-1) in development of postoperative complications of patients with degenerative-dystrophic diseases (DDD) of hip joints

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Purpose of our work was to estimate of prediction possibility of development of complications in postoperative period by means of monitoring of MMP-9/TIMP-1 level in patients' blood serum before and after hips replacement.
Materials and Methods: We tested blood of 27 patients before and after hips replacement operation, undergoing, 11 patients (40.74%) from them had postoperative infectious complications being developed, and 16 patients (59.26%) had smooth postoperative course without infectious complications. Blood of 30 healthy donors was as control. It was drawn fourfold blood sampling, as the following: the first blood sampling was made before operation, the second - during first 24 hours after operation; the third - during fifth 24 hours after operation and the fourth - during fourteenth day after operation. Estimation of MMP-9/TIMP-1 was made by immunoenzyme method using of the test-system of "R & D System Inc.", USA.
Results: It was detected the following: before operation patients from group with smooth postoperative course had level of MMP-9/TIMP-1 equal 10.90±0.83 ng/ml that was essentially higher than patients of control group had, being 0.22±0.07 ng/ml ($p<0.05$) typical for healthy people. There was increase of MMP-9/ TIMP-1 (12.59±1.02 ng/ml, $p<0.001$) on the first days after operation with subsequent continued increase up to 12.75±1.25 ng/ml, $p<0.001$ on the 5th day. Essentially decrease of MMP-9/ TIMP-1 up to level of 12.40±0.98 ng/ml was observed on the first day after operation that for certain did no differ from values of first day after operation. In the patient group with complicated postoperative course the index MMP-9/ TIMP-1 before operation was 9.54±1.37 ng/ml, this index was higher in the control group as well (0.22±0.07 ng/ml ($p<0.01$). On the first day after operation it was observed rapid growth of MMP-9/ TIMP-1 (13.37±1.41 ng/ml, $p<0.05$) in above-mentioned patient group, and on the 5th day the level still persisted high (14.82±1.74 ng/ml). On the 14th day the level of MMP-9/ TIMP-1 decreased slightly and was 14.27±1.07 ng/ml ($p>0.05$).
Conclusion: Divergence of MMP-9/ TIMP-1 ratio towards its significant increase already before operation for patients with DDD testifies to essential pathogenetic role of MMP-9 in pathogenesis of dystrophic diseases of hips.

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The role of hyalgan at inhibition of IL-1beta-stimulated production of matrix metalloproteinase

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Background: Hyalgan is now widely used in the treatment of osteoarthritis (OA) by intra-articular administration into the affected joints. Although a clinical benefit of Hyalgan has been demonstrated with respect to pain relief in patients with OA, the level at which the drug acts remains unclear.
Objective: This study was aimed to investigate the mechanism of inhibitory action of Hyalgan on interleukin-1beta (IL-1beta)-stimulated production of matrix metalloproteinases (MMPs) in human articular cartilage.
Methods: IL-1beta was added to human articular cartilage with or without changes of osteoarthritis (OA) in explant culture to stimulate MMP production. Articular cartilage was incubated or preincubated with Hyalgan to assess the effect of Hyalgan on IL-1beta-induced MMPs. Secreted levels of MMPs-1, -3, and -13 in conditioned media were detected by immunoblotting, while intracellular MMP synthesis in chondrocytes was evaluated by immunofluorescence microscopic analysis. Penetration of Hyalgan into cartilage tissue and its binding to CD44 were analyzed by fluorescence microscopy using fluoresceninated Hyalgan. Blocking experiments with anti-CD44 antibody were performed to investigate the mechanism of HA action.
Results: Treatment and pretreatment with Hyalgan resulted in significant suppression of IL-1beta-stimulated production of MMPs in normal and OA cartilage explant culture. Fluorescence histocytochemistry revealed that Hyalgan penetrated cartilage tissue and localized in the pericellular matrix around chondrocytes. Hyalgan -binding blocking experiments using anti-CD44 antibody demonstrated that association of HA with chondrocytes was mediated by CD44. Preincubation with anti-CD44 antibody, which suppressed IL-1beta-stimulated MMPs, reversed the inhibitory effect of Hyalgan on MMP production induced by IL-1beta in normal and OA cartilage.
Conclusion: This study demonstrated that Hyalgan effectively inhibited IL-1beta-stimulated production of MMP-1, MMP-3, and MMP-13, which supports the clinical use of HA in the treatment of OA. Such Hyalgan action on IL-1beta may involve direct interaction between Hyalgan and CD44 on chondrocytes.

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Nasal lavage fluid eosinophil cationic protein gradient reflects the eosinophilic inflammation in RSV infection

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Background and Purpose: It is well known that after respiratory syncytial virus (RSV) bronchiolitis during infancy and early childhood, a number of children develop a persistent wheezing. The study of eosinophil cationic protein (ECP) suggests that, as with asthma, eosinophilic inflammation mechanisms may also play a role in RSV infection.

The aim of our study was to prove the existence of eosinophilic inflammation in infant with RSV infection, by measuring local/systematic ECP.
Methods: ECP was measured in the nasal lavage fluid and serum of 41 infants with RSV infection and 29 other respiratory infections, all who were admitted to our hospital between October, 2005 and March, 2007 (they ranged from 0 months to 24 months of age, except for four pair of twins).
Results: Nasal lavage fluid ECP levels were significantly higher in infants with RSV infection than in the infants with other respiratory infections. (3.148±0.527 vs 2.723±0.461 microgram/L, $p<0.05$). But serum ECP levels did not indicate such a tendency. Moreover, a subanalysis showed that wheezing infants also had significantly higher levels of nasal lavage fluid ECP levels than in the case of non-wheezing infants, (3.240±0.512 vs 2.833±0.491 microgram/L, $p<0.05$), but serum ECP levels did not indicate such a tendency. With regard to both nasal fluid lavage and serum, no significant difference in ECP levels were found whether there was a family history of atopy or not. This suggests that RSV infection may induce local eosinophilic inflammation even during infancy.
Conclusion: Eosinophils are more strongly activated in RSV infection than in other respiratory infectious disease. Theoretically, this result supports the hypothesis that RSV infection may cause reactive airway disease.

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Mechanisms of eosinophil infiltration in the middle ear of patients with eosinophilic otitis media

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Background: Patients with intractable otitis media (OM) with co-existent bronchial asthma have an extensive accumulation of eosinophils in the effusion and mucosa of the middle ear; called eosinophilic otitis media (EOM). Previously, we reported that mast cells can regulate eosinophilic inflammation in nasal polyps. In order to elucidate the mechanisms of eosinophil accumulation in the middle ear, we analyzed eosinopil chemoattractants like RANTES, eotaxin and their ligand CCR3 as well as mast cell mediator tryptase in middle ear biopsies and effusion (MEE) of patients with OM with and without asthma.

Materials and Methods: By ELISA, we analyzed the levels of RANTES and eotaxin in MEE and mucosa of patients with OM with and without asthma (control). The number of tryptase+ cells (mast cells), MBP+ cells (eosinophils), RANTES+, eotaxin+ and CCR3+ cells were analyzed by immunohistochemistry in middle ear biopsies.

Results: Levels of RANTES and eotaxin were higher in patients with OM with asthma as compared to controls. Whereas in the mucosa of controls, MBP and eotaxin+ cells were not detected, they were markedly increased in patients with asthma in concert with high numbers of tryptase+, RANTES+ and CCR3+ cells.

Conclusion: These data suggest that mast cells may regulate eosinophilia accumulation in the middle ear of patients with EOM via RANTES and Eotaxin.

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Eosinophil functions induced by adhesion molecules and leukotriene D_4

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Background: Eosinophils preferentially accumulate at sites of airway inflammation of asthma. For circulating eosinophils to participate in the asthmatic airways, it is necessary to interact with adhesion molecules expressed on endothelial cells and then expose to inflammatory mediators such as cysteinyl leukotrienes(cysLTs). There is evidence that cysLTs including leukotriene (LT)D_4 regulate the functional status of eosinophils.

Objective: To investigate whether interaction with adhesion molecules modifie eosinophil functions induced by cysLTs.

Methods: rh-VCAM-1, rh-ICAM-1, or rh-P-selectin was dissolved in 0.05 M $NaHCO_3$ coating buffer, added to 96-well EIA plates and incubated at 4 °C overnight. Residual fluid was decanted and HBSS/0.1% gelatin was added to reduce non-specific activation of eosinophils. Eosinophils were isolated from blood of healthy donors, incubated in the EIA plates, and then exposed to LTD_4. The generation of superoxide anion (O_2^-) and release of eosinophil-derived neutrotoxin (EDN) were evaluated by cytochrome C reduction assay and ELISA, respectively.

Results: In this experimental condition, neither VCAM-1 nor LTD_4 (100nM) directly induced eosinophil O_2^- generation, however, VCAM-1 and LTD_4 act synergistically to induce eosinophil O_2^- generation. The O_2^- generation induced by combination of VCAM-1 and LTD_4 was blocked by anti-α4 integrin mAb and anti-β2 integrin mAb. ICAM-1 by iteslf induced eosinophil O_2^- generation and this was enhanced by LTD_4. The enhanced O_2^- generation was blocked by anti-β2 integrin, but not anti-α4 integrin mAb. P-selectin did not induce O_2^- generation in the presence or absence of LTD_4. Finally, a combination of LTD_4 and VCAM-1, but not ICAM-1 or P-selectin, induced the release of EDN.

Conclusion: The combination of VCAM-Ior ICAM-I and cysLT effectively induce effector functions of eosinophils. Eosinophil adhesion to and migrate across endothelial cells via these speficic adhesion proteins and subsequent exposure to cysLT may be involved in the manifestations of eosinophil activation in the airways of asthma.

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IL-21 and IL-21 receptor expression in eosinophils

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Introduction: Eosinophils are recruited to late phase inflammation and elicit host defense against parasitic infections. However, in allergy eosinophils contribute negatively by eliciting tissue damage. Our aim was to evaluate the expression of interleukin-21 (IL-21) and IL-21 receptor (IL-21R) in eosinophils.

Methods: Eosinophils where purified from blood samples taken from allergic and healthy individuals using CD16 and CD3 microbeads to deplete neutrophils and remaining T-cells from the granulocyte population. RNA was purified using ABI prism 6100 robot and real-time PCR was performed using applied biosystems Taqman Gene Expression Assays. ELISA assays were performed for IL-21 receptor. Eosinophilic apoptosis was measured in a flow cytometric assay using AnnexinV and propidium iodide for 200,000 cells at 24, 48 and 72 hour time points.

Results: The yield of eosinophils from healthy and allergic persons was $64x10^3$ and $137x10^3$ eosinophils per ml full blood respectively. Using real-time PCR no IL-21 mRNA could be detected; however IL-21 receptor mRNA was detected with a mean Ct-value of 31.16 from 6 donors. No difference was seen between allergic and healthy individuals. The naïve B-cell line Ramos was used as a positive control and yielded a Ct-value of ~20, which is roughly a 1000-fold higher expression of IL-21R mRNA in Ramos compared to eosinophils. The IL-21R mRNA expression in eosinophils was comparable to the eosinophilic cell line HL-60 clone 15.

A sandwich ELISA for detection of IL-21R in eosinophil cell lysate was established using a polyclonal rabbit anti-IL-21R antibody and a mouse monoclonal anti-IL-21R. The ELISA had a detection limit of 1 ng/ml or roughly 1000 receptors per cell and no IL-21R expression was detected in either eosinophils or Ramos cell line. The effect on eosinophil apoptosis in presence of IL-21 or the other Type I cytokines (IL-19, IL-20, IL-22, IL-24, IL-26, IL-28 and IL-29) was examined, however, no apparent effect of either increased survival or increased apoptosis could be seen.

Conclusion: Eosinophils express IL-21R mRNA. Real-time PCR showed a mean Ct-value 31.16 from 6 donors. IL-21R protein expression was examined using both flowcytometry and ELISA, but no IL-21R could be detected. Functional apoptosis assay showed that IL-21 or the other Type I cytokines had no effect on eosinophils, so it is unlikely that IL-21 stimulation of eosinphils is of major significance.

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Anti-interleukin-5 (mepolizumab) therapy for a hypereosinophilic syndrome

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Background: IL-5 is critically involved in proliferation, activation, migration and survival of eosinophils. As such, IL-5 may play a pathogenic role in hypereosinophilic syndromes (IHES), which are a heterogeneous group of disorders, characterized by sustained peripheral blood and/or tissue eosinphilia. We assessed the safety and efficacy of a humanized monoclonal anti-IL5 antibody in a patient with IHES with respiratory involvement.
Methods and Results: The patient is a 25 year-old woman, with a 3 year history of IHES. Biopsies confirmed on various occasions the involvement of lungs and paranasal sinuses. The onset of the disease was an eosinophilic pneumonia (56% eosinpohils in bronchial lavage) with asthma and respiratory failure. The persistent eosinophilia and respiratory symptoms were successfully treated with doses of prednisone (25–75 mg/daily), variable according to eosinophilia. Attempts to shift to inhaled steroids plus montelukast, invariantly lead to relapse. However, withdrawal of steroid treatment was forced by severe systemic side effects and bleeding gastric ulcera. An attempt with initimab was not effective.

A treatment with mepolizumab (5 mg/kg intravenously) monthly was then started. Mepolizumab was well tolerated and rapidly reduced both eosinophilia and symptoms. Eosinophils decreased from 998/mmc to 210 after the first infusion and remained below 250/mmc at the subsequent controls. After 8 weeks the oral steroid could be discontinued.
Conclusion: Mepolizumab is safe, and effective in lowering eosinophil count , has a glucorticoid sparing effect in patients with IHES.
Keywords: hypereosinophilic syndrome, pneumonia, mepolizumab.

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Influence of cetirizine and levocetirizine on granulocyte-macrophage colony-stimulating factor and interleukin-8 secretion by A549 human airway epithelial cells stimulated with interleukin-1beta

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Background: Recent studies suggest that several antihistamines can modulate various inflammatory reactions besides their H1-receptor antagonism. We investigated the effect of cetirizine (Ceti) and levocetirizine (Levo) on granulocyte-macrophage colony- stimulating factor (GM-CSF) and interleukin-8 (IL-8) secretion of A549 human airway epithelial cells.
Methods: A549 cells were pre-incubated with Ceti (0.1, 1, 2.5, 5, 10 μM) or Levo (0.1, 1, 2.5, 5, 10 μM) individually for 16 hours and were then stimulated with IL-1β for 8 hours. The levels of GM-CSF and IL-8 in cultured supernatants were measured by enzyme-linked immunosorbant assay (ELISA).
Results: Our data showed that Ceti 5 and 10 μM and Levo 2.5, 5, and 10 μM significantly suppress GM-CSF secretion by 29.29%, 38.45%, 39.94%, 38.09% and 42.85%, respectively. While Levo 5 and 10 μM significantly suppressed IL-8 secretion (22.43% and 34.58%, respectively), this was possible only with Ceti 10μM (24.96%).
Conclusion: These results suggest that both cetirizine and levocetirizine at higher concentrations reduce the release of GM-CSF and IL-8 from A549 human airway epithelial cells stimulated with IL-1β. These observations indicate that these antihistamines may exert anti-inflammatory effects beyond their antagonistic histamine H1-receptor activity. Our results suggest that Levo could be more potent than Ceti in terms of anti-inflammatory activity.

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A new interesting property of hydroxyzine, a drug used in the treatment of chronic idiopathic urticaria and anxiety disorders

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Background: The histamine-H1 receptor antagonist, hydroxyzine (H) has a wide variety of therapeutic applications, including the treatment of allergic skin disorders and the control of nausea and vomiting caused by conditions such as motion sickness. It is also used as a tranquilliser for the symptomatic management of conditions such as generalised anxiety disorder and the tension associated with psychoneuroses and in the treatment of alcohol withdrawal symptoms. It is used too in the management of pruritis associated with allergic conditions such as chronic urticaria and histamine-mediated pruritus. Its Ki towards the human histamine H1 receptor is 2 nM. The enzyme semicarbazide-sensitive amine oxidase (SSAO; EC 1.4.3.6) exists as a soluble form in plasma and a membrane-bound form associated with the exterior surface of the plasma membrane in many tissues. The membrane-bound form can function as a vascular adhesion in mediating the early stages of inflammatory responses (Salter-Cid et al. JPET 2005). Monoamine oxidase (MAO) B is involved in the oxidative deamination of an histamine metabolite, N-telemethylhistamine.
Methods: H inhibitory properties towards the bovine plasma and membrane-bound form of SSAO and towards rat liver MAO A and B have been investigated as described by O'Sullivan et al. (J Neural Transm 2006).
Results: H is a relatively potent competitive inhibitor of bovine plasma SSAO (IC50 = 20±0.5 μM; Ki = 1.8±0.4 μM). H was a somewhat less potent competitive inhibitor of MAO B (Ki = 19±0.4 μM) and a very weak inhibitor of the bovine membrane-bound form of SSAO (IC50 ~ 900 μM) and MAO A (IC50 ~ 650 μM).
Conclusion: The inhibitory values towards the bovine plasma SSAO are higher than the reported plasma levels of the drug when administered at the therapeutic doses of 25–100 mg/day (Lader & Scotto Psychopharmacol 1998) [e.g. mean Cmax = 34 ng/ml (91 nM) and 73 ng/ml (195 nM) after a single dose of 25 mg and of 39 mg, respectively (Simons et al. JACI 1984, 1995)]. However, as H plasma concentrations are higher after repeated administration at steady state and as human tissue concentrations are not

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Concentration (μM)	Level of GM-CSF secretion compared to IL-1β (100%)			Level of IL-8 secretion compared to IL-1β (100%)		
	Cetirizine (%)	Levocetirizine (%)	p value vs. IL-1β	Cetirizine (%)	Levocetirizine (%)	p value vs. IL-1β
0.1	101.64±5.12	96.50±2.90		103.83±4.80	101.16±9.27	
1	79.33±9.28	77.15±8.59		97.79±7.63	95.96±9.64	
2.5	77.55±8.44	60.06±19.36	p<0.01 for Levo	98.73±2.60	88.97±5.69	
5	70.71±11.80	61.91±15.00	p<0.05 for Levo & Ceti	89.70±1.06	77.57±6.09	p<0.01 for Levo
10	61.55±5.82	57.15±5.70	p<0.01 for Levo & Ceti	75.04±5.28	65.42±8.59	p<0.01 for Levo & Ceti

Mean ± standard error of mean

known, it would be interesting to examine the inhibitory properties of the drug also towards the human plasma and membrane-bound form of SSAO, in order to estimate the possible significance of such inhibition in terms of the therapeutic actions of H.

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Quantitative and qualitative analysis of circulating Vgamma9/Vdelta2 T cells in newborns versus adults

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Background: Human Vgamma9/Vdelta2 (Vγ9/Vδ2) T lymphocytes are known to participate in the defense against microbial pathogens and have anti-tumor activity. The cellular immune system of newborns is generally considered to be immature and hypo-responsive in comparison to that of adults. In this study we made a quantitative and qualitative comparison of the Vγ9/Vδ2 T lymphocytes, their activation status and production of cytokines in cord blood mononuclear cells (CBMC) versus those in adult peripheral blood mononuclear cells (PBMC).

Methods: We employed flow cytometric analysis to compare numbers and phenotypic characteristics of Vγ9/Vδ2 T lymphocytes between CBMC and adult PBMC.

Results: Numbers of Vγ9/Vδ2 T cells are lower in CBMC than those in adult PBMC while TCR expression levels were similar in both groups. Stimulation of CBMC Vγ9/Vδ2 T cells with isopentenyl pyrophosphate (IPP) did not induce IF-γ, in contrast with adult PBMC positive production of this Th1 cytokine. The addition of IL-2 to IPP did not enhance IFN-γ in CBMC Vγ9/Vδ2 T cells. PMA/ionomycin stimulation did not lead to INF-γ production in CBMC Vγ9/Vδ2 T cells, inversely again, adult PBMC activation produced this cytokine.

Conclusion: In addition to the immaturity of the adaptive immunity in newborns, such as αβ T cells; CBMC Vγ9/Vδ2 T cells are hypo-responsive in comparison with adult PBMC Vγ9/Vδ2 T cells and this may be associated with the predisposition to infection with virus, bacteria and fungus, as well as the development of allergic inflammation in newborns.

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The matrix metalloproteinase 9 and its first type's inhibitor type (hMMP-9/TIMP-1) in bronchalveolar lavage fluid of pulmonary tuberculosis patients

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Significant components of the territorial matrix are the matrix metalloproteinase that is a group of proteolytic enzymes, which are especially active in conditions of an inflammatory response. Matrix metalloproteinase 9 (MMP 9) is produced by normal alveolar macrophages, granulocytes, and neutrophiles. Expression of MMP 9 is increased in tissues affected by the process of remodeling and angiogenesis. Negative influence of this process is restricted by tissular inhibitor of metalloproteinase (TIMP-1). Main function of TIMP-1 consists in regulation of distraction of collagen and basic membrane components. We have researched a quotient of MMP 9 and its first type's inhibitor contained in the bronchalveolar lavage fluid of 23 teenagers suffered from pulmonary tuberculosis. Received results have been processed through statistical analysis; an arithmetic mean (M) and a mean error (m) have been also determined. Fidelity of results has been reckoned through Mann - Whitney nonparametric test. Evaluation was made in accordance with the clinical and laboratory findings and bacteriological test of sputum. The researches have shown that quantity of hMMP-9/TIMP-1 in the bronchalveolar lavage fluid of pulmonary tuberculosis patients comes to 2803, 85 ± 270, 72 pg/ml. It was also elicited a tendency for increase of hMMP-9/TIMP-1 in Koch's bacillus cases (+) (3127, 23 ± 555, 20 pg/ml vs. 2735, 77 ± 315, 58 pg/ml; p>0,05). Differences in hMMP-9/TIMP-1 of patients with local pulmonary tuberculosis and infiltrative phthisis cases haven't been revealed. The received findings are not sufficient for making unambiguous conclusions concerning pathogenetic value of the researched mediators and denote necessity for further searches of diagnostic characters for inflammation in cases of teenagers' pulmonary tuberculosis.

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The cytokines production at the experimental pneumonia

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Activation of cytokines system at different variants of the immune inflammation occurs without dependence from the etiologic factor. At the same time there is the data, testifying that microorganisms can develop the products being similar cytokines, and thus influence on the cytokines cascade at the immune answer. The purpose of the present investigation was to reveal influence of the etiologic factor on parameters of the local and systematic structure of the opposite cytokines (IFNγ and IL-10) at the experimental pneumonia caused by S. aureus and E. coli. Infection of mice of ÑÂÀ line was spent intranasally in the doze of 1x103 mt/ml for S. aureus (I experimental group) and 1x105 mt/ml for E. coli (II experimental group). Production of cytokines by blood cells and mice lungs was investigated for the 10th day after infection in reaction of IFA.

Results and Conclusion: At infection of mice S. aureus level of IFNγ considerably (ð <0,05) decreased (up to 0,84+0,1 pg/ml in blood and up to 36,38+2,38 pg/ml in supernatant of lungs). IL-10 level at mice of tentative group also tended to decrease and made 14,15+1,3 pg/ml in blood and 70,8+2,88 pg/ml in supernatant of pulmonary tissue. At infection of mice E. coli content of IFNγ in blood changed little (3,97+0,28 pg/ml), and decreased in supernatant of lungs, but not so prominently, as at mice of tentative group (70,84+2,17 pg/ml). IL-10 level in blood of mice of the second experimental group was authentic (ð <0,05) below than in group of the control and made 10,8+0,87 pg/ml. And IL-10 level in supernatant of pulmonary tissue at infection of mice by E. ñoli increased practically in 2 times in comparison with the group of the control (186,08+5,13 pg/ml, ð <0,05). Thus, carried out investigation revealed features of local and systemic production of cytokines depending on etiology of the experimental pneumonia. IFNγ level under influence S. àureus prominently decreased. Development of inflammatory process in the lungs, caused by E. coli, rendered significant influence on local production of IL-10 increasing its level more than in 2 times, moreover IL-10 authentically decreased in peripheral blood.

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Rate of antiphlogistic cytokines in blood serum and lacrimal liquid of the patients with ophtalmoherpes

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Ophtalmoherpes is a grand medical and social problem. Herpetic infection of eyes is relapsed in 50% of cases and results to reducing acuity of vision, disablement and deteriorates life activity. Enlargement of conception regarding inflammatory mechanism let us opportunity to optimize respective therapy.

Objective: To work out cytokines content (TNF-á, GM-CSF, IL-12p70, IL-12p40 and IL-2) in blood serum and lacrimal liquor of the patients.

Methods and Materials: Serum and lacrimal liquid of 60 ophtalmoherpes patients were tested conformably (women 38, man 22). Control group embraced 30 practically healthy volunteers without disease of organum visus. Cytokines were quantified with IFA method using specific reagents "R and D Diagnostics Inc." (USA).

Consequences and Discussion: We found polysemantic modification of local and systemic cytokine profile of patients as against reference group. In blood serum were determined acceleration of IFNá 30-50 times more (ñ<0,001), IL-2 level 2-10 times more (p< 0,5), IL-12p40 content decreased 1,5-3 times (p<0,05), whereas GM-CSF and IL12p70 were near to standard. The abovementioned indices in lacrimal liquor appeared in the following way. Authentically increasing of IL12p70 level (10,08+1,29 pg/ml against 4,98+1,1 pg/ml, p<0,01) and GM-CSF (7,57+1,73 pg/ml against 2,15+0,64 pg/ml, p<0,01). TNFá and IL-12 modifications had a tendency to heightening (p<0,2), whereas IL-12p40quantity reduced (p<0,05) in the same way as in blood serum. Obtained results allow us to draw a conclusion, that at ophtalmoherpes disease neutrophiles and macrophages mainly are more active locally and produce GSM-CSF and IL-12p70. System response is characterized by hyper production of TNFα and IL-2. Compensative anti-inflammatory reaction distressed as locally as systematically.

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Streptokinaza indused pulmonary infilltrates with hipereosinophilia-case report

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The various drugs and enviromental agents capable of producing acute pulmonary infiltrates with eosinophilia. We present a man on 48 with acute infarct of myocardi who was treated with streptokinaza besides the usual cardiological treatment. From the anamneza we faund aut that the patient did not have problems with his heart, neither any kind of alergic or some other kinds of ilneses. By the first impresion the patient was conscious, skared,dispnoic, afebril,with strong pain on his chest. By auskultation he had anormalvezicular breathig and heart rate of 110/min. The other phisical finds were in order. Imidiately after the posed diagnoza, a usual cardiological therapy was given and in the same time a streptokinaza (1500 000IE) was included. The patient beard this treatment very well. On the third day of the therapy his health condition deteriorated. He got frequent chaemoptisis, increased temperature(38C), decreased blod presure, and rich find of rales on clinical examinaton. Laboratory examination showed:ESR 42mm/1st hour, strong blod reduction, eosinophilia 49%, leukocytosis. On the x-chest ray both upper lobes were caracteristicaly involved and were described as the "photographic negative" the radiographic shadows seen in pulmonary oedema. Chest CT showed bilateral pulmonary infiltrates mainly in upper and middle lobes. Bacterial examination and cultures of sputum were all negative. The search for parasistic infestation was negative as well as for BK infection. The lung volumens and diffusion capacities werw decreased and hipooxemia was most prominent. We started the therapy with 40mg prednisone daily with successive decrising in the folowing 6 weeks and also antibiotic and oxigen therapy. Some improvement was seen in less than a week but the radiographic shadows had completely resolved for 6 weeks. The number of eosinophils olso came back to normal.

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Serum IgE and Eosinophils(Eo) as an indicators for severity in pulmonary thromboembolismus

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Background: Pulmonary thromboembolismus can be presented through pulmonary infiltration, which manifestation depends on extensiveness of the pulmonary embolisation and the presence of pleural effusion (PE). In a period of 3 years 29 pts with pulmonary thromboembolism were studied with regard to serum concentrations of IgE, D-dimer and Eosinophils in the pleural fluid during acute phase (on hospital admission) and after treatment (recovery phase).

The aim of the study was to estimate the correlation between these parameters during acute and recovery phases.

Methods: The number of eosinophils in the pleural fluid was determined from smear of pleaural fluid colored by May-Grunwald Giemsa. Serum IgE was measured with Radioimmunoassay diffusion (RID).

Results: The serum IgE concentration increased during the acute phase to 422+/- 307 IU/ml and decreased afterwards in all pts. The increase in serum IgE concentration lagged a few days behind that of the serum D-dimer concentration, indicating later IgE production then thrombus formation and lysis. Most of the pts developed a small chaemoragic PE, while 7 of them had more then 30% of eosinophils and highest levels of IgE measured in acute phase.

Conclusion: These results indicated a relationship between serum IgE concentration and patophisiology of pulmonary thromboembolismus. Serum IgE and Eo count may be a good mark of severity for pulmonary thromboembolismus indicating on pathogenesis and prognosis.

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Modulation of peripheral and cord-blood derived gamma/delta T cells immune response after stimulation with newly designed compounds: possible new therapeutic approach

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Immune system, in response to infective or allergic agents, makes use of different cellular populations, such as alfa/beta and gamma/delta (g/d) T cells. The latter are present in both peripheral (PB) and cord (CB) blood, being the second in a immature stage, and respond to a variety of non-peptidic antigens, through a HLA-independent system, by secreting pro-inflammatory and immunomodulant cytokines, expressing membrane receptors and costimulatory molecules, and producing cytotoxic effectors such as perforin and granzyme. Recent studies have suggested that also the less common g/d T cells may play a role as effectors and immunoregulatory cells in the development and perpetuation of allergic inflammation and also in bronchial asthma, rhinitis and eczema We aimed at analysing the capability of g/d T cells derived from PB or CB to respond to non-proteic antigens, in view of a putative therapeutic use in newborn or

infant pathologies. We firstly found that CB-derived g/d cells strongly respond to stimulation with aminobiphosphonate (ABs) compounds, such as pamidronate and zoledronate, in terms of expansion, whereas stimulation with pyrophosphates (PPs) only lead to no or little expansion. The population expanded with ABs is also perfectly functional, since cells are able to produce large amounts of cytokines (TNFalfa e IFNgamma) if secondarily stimulated with he same antigens. Interestingly, also PPs can induce ABs-expanded cells to cytokine production after a secondary stimulation, suggesting the existence of different responsive pathways concerning expansion and cytokine production in CB-derived g/d cells. Once established that CB g/d T cells are functional and can be efficiently stimulated, we focused our study to the stimulation or down-regulation of g/d T cells with newly designed and synthesised compounds (NCs). We therefore characterised a small-compounds library, and selected few molecules for their capability to stimulate or down-modulate g/d T cells from PB. We than used these molecules also for CB g/d stimulation. We found that, following stimulation with NCs, CB-derived g/d T cells strongly respond in terms of both cellular expansion and cytokine production, and this result was confirmed though the use of CB-derived gamma/delta clones. Studies are in progress to evaluate the toxicity of these compounds, and consequently their employment in vivo as g/d T cells immunomodulators in new therapeutic approach on infectious or allergic diseases.

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A novel low molecular weight thiol compound, N-acetylcysteine amide, attenuates allergic airway inflammation and hyperresponsiveness

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Background: Reactive oxygen species (ROS) play an important role in the pathogenesis of airway inflammation and hyperresponsiveness. Recent studies have demonstrated that antioxidants are able to reduce airway inflammation and hyperreactivity in animal models of allergic airway disease. A newly developed antioxidant, small molecular weight thiol compound, N-acetylcysteine amide (AD4) has been shown to increase cellular levels of glutathione (GSH) and to attenuate oxidative stress related disorders. However, the effects of AD4 on allergic airway disease such as asthma are unknown.

Objective: This study aimed to determine the effects of AD4 on bronchial inflammation and airway hyperresponsiveness.

Methods: We have used a mouse model for allergic airway disease. The effects of AD4 on bronchial inflammation and airway hyperresponsiveness studies by measurement of intracellular ROS, GSH, glutathione disulfide, vascular permeability, proteins, tissue inflammation, and airway resistance.

Results: The increased levels of Th2 cytokines, nuclear factor-κB (NF-κB) and hypoxia-inducible factor-1α (HIF-1α), and vascular endothelial growth factor, the increased ROS generation, the increased vascular permeability, and the increased mucus production after ovalbumin inhalation were significantly reduced by the administration of AD4.

Conclusion: These results suggest that AD4 attenuates airway inflammation and hyperresponsiveness by regulating activation of NF-κB and HIF-1α as well as reducing ROS generation in allergic airway disease of mice.

Clinical implication: These findings provide an important molecular mechanism for the use of a novel antioxidant, AD4 to prevent and/or treat asthma and other airway inflammatory diseases.

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Th1-dominant shift of T cell cytokine production, and subsequent reduction of serum immunoglobulin E response by administration in vivo of beta-carotene in a mouse model

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Background: Th1 and Th2 cells, resulting from antigenic stimulation in the presence of IL-12 and IL-4, respectively, are implicated in the pathology of various diseases including allergic and autoimmune diseases. In this study we have asked whether administration of beta-carotene brings about a Th1/Th2 shift in vivo of the mice, and subsequent reduction of circulating IgE.

Methods: Feed containing beta-carotene was administered orally to BALB/c mice immunized intraperitoneally with ovalbumin (OVA) for approximately 1 month. The titers of OVA-specific IgE, OVA-specific IgG1 and OVA-specific IgG2a in the mouse sera were determined. Cytokine productions by the spleen cells and serum Ig concentrations were studied by ELISA. We also examined the effect of fed beta-carotene on active systemic anaphylaxis.

Results: Feeding beta-carotene to mice immunized with OVA inhibited the immediate reduction of the body temperature induced by antigen stimulation. Furthermore, the increase in serum histamine in the mice fed beta-carotene under active systemic anaphylaxis was lower than in controls. We then examined the pattern of cytokine production by spleen cells from mice followed by restimulation with OVA in vitro. The spleen cells showed enhanced Th1-specific cytokine production; spleen cells from the beta-carotene administered mice produced more IFN-gamma as compared with those from control mice in an antigen-specific manner. IL-2 and IL-4 secretions of the spleen cells were comparable between the two mouse groups. Beta-carotene administration did not reduce serum IgG concentration. It markedly reduced total IgE level and an IgG1/IgG2a ratio, reflection of Th1/Th2 balance, in sera. Furthermore, beta-carotene administration reduced ovalbumin (OVA)-specific IgE levels in sera of the OVA sensitized mice.

Conclusion: Thus, beta-carotene enhances IFN-gamma secretion and thus modulates Th1/Th2 cytokine balance, leading to reduction of serum IgE.

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Effect of statins on PBMC and NK cells and on fibroblasts from human nasal polyps and turbinates

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Background: Statins are serum cholesterol-lowering agents used for the prevention of atherosclerotic vascular disease but there is, a growing evidence they might have immunomodulatory activities.

Methods: To evaluate the effect of these drugs in allergic diseases we treated PBMC from healthy and allergic patients in vitro with ParJ1,2 (5 μg/ml), Fluvastatin (0,1 μM), Atorvastatin (1 μM) and Simvastatin (1 μM) alone and in combination.

NK derived from healthy subjects were activated with IL-2 (300 U/ml) alone or with the addition of statins.

We also evaluated their effect on fibroblast cultures derived from human nasal polyps and turbinates stimulated as follows: not treated and treated with FGF (5ng/ml) alone or in combination with statins.

Results: After different days in culture, cells were analyzed by flow-citometry for the evaluation of the following cell surface receptors: CCR3, CCR4, CXCR3 and CCR5 for PBMC; NKp30, NKp44 and NKp46 for NK cells and CD106 and CD54 for fibroblasts.

Our results show that statins have no modulatory effect on cell surface protein expression from both PBMC and NK.

CD54 was not modulated by the treatments in nasal polyp fibroblasts, whereas in turbinate fibroblasts its expression was upregulated by FGF and downregulated after the addition of Fluvastatin or Atorvastatin.

Interestingly, also CD106 expression was not modified in nasal polyp fibroblasts stimulated or not with FGF and statins. Fibroblasts from turbinates showed an increased expression of CD106 after stimulation with FGF and only the addition of Atorvastatin was able to downregulate it.

Conclusion: In conclusion, the different behaviour of statins on the diverse kind of cells in vitro is prompting news in the studies about their use for the treatment of inflammatory diseases.

ENVIRONMENTAL FACTORS

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Latex allergy in operating room; not frequent but severe

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Objective: Latex allergy is becoming a major health concern among healthcare workers, of whom approximately 2.8 to 18% are reportedly sensitized. The aim of this study to determine the prevalence of latex allergy and potential crossreacting foods in operating room stuffs.

Methods: One hundred and four operating room stuffs (aged 24 years to 58 years) including doctors, nurses and technicians completed a latex allergy questionnaire. They were questioned about symptoms of latex reactivity and about other allergies particularly to foods that may crossreact with latex. Informed consent was obtained and skin prick tests were performed with natural rubber latex.and with five potentially crossreacting foods (banana, kiwi, melon, tomato and potato). Specific IgE antibodies against latex and these foods were evaluated (Pharmacia CAP RAST system).

Results: Five personnel (4.8%) described allergic symptoms they attributed to latex exposure. All latex allergic stuffs had skin symptoms, 2 had severe anaphylactic reaction and 2 had severe asthma. All these 5 personnel had positive reactions to both latex and crossreactive foods. Specific IgE against to latex was found to be positive in this group. There was no significant difference between the latex SPT-positive and -negative health care workers according to age, sex and total exposure time to latex.

Conclusion: In this study we found the prevalence of allergy 4.8% among the operating room personnel. Although latex allergy was not more frequent in our population its presence may lead to severe allergic reactions such as anaphylaxis and severe asthma.

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Dog ownership and contact with dogs during childhood and later development of allergy; results of combined German birth cohort studies

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Background: The effect of dog ownership during childhood on the development of allergy has been investigated in few studies with conflicting results.

Objective: We investigate the association between childhood dog ownership, regular contact to dogs, and indoor endotoxin exposure during infancy and the development of allergic sensitization and atopic disease up to age 6 in 2 German cohort studies.

Methods: Data from two ongoing birth cohorts GINI (n=1962) and LISA (n=1193) were analysed. In both studies, information on children's contact with dogs and their allergic symptoms and doctor diagnosed allergic disease were collected at each follow-up using questionnaire. Specific IgE antibodies to common aeroallergens were measured at age 6. House dust samples were collected when the children were 3 months old and the amount of endotoxin in house dust was determined.

Results: Dog ownership in early childhood was associated with a significant lower rate of mixed pollen and inhalant sensitization but had no effect on dog sensitization and the prevalence of allergic symptoms and diseases up to age 6. Regular contact with dog during childhood without ownership was not associated with any of the health outcomes. No associations were found between house dust endotoxin exposure during infancy and sensitization to dog, mixed pollen, and inhalant allergens.

Conclusion: Dog ownership in early childhood protects against the development of inhalant sensitization and this effect cannot be attributed to the simultaneous exposure to endotoxin.

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School-a source for exposure to furry-pet allergens

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Background: The school environment is an important source of exposure to furry-pet allergens for most children in Sweden. This will have an impact on their health and possibly on development of allergy. The aim of this study was to compare two methods of allergen sampling, and to investigate the association between percentage of animal owners and allergen levels.

Methods: Totally 120 classes in 35 primary and secondary schools in Uppsala, Sweden, were randomly selected. A questionnaire, including questions on pet ownership, was answered by 2 355 pupils aged 7–13 years. Samples of settled dust were collected from floors and furniture throughout the classroom with a vacuum cleaner with a special sampling filter (ALK Abello). Airborne particulates were collected passively in Petri-dishes placed in the class-rooms over a week. Allergen levels for cat, dog and horse were determined using ELISA. Correlations between allergen levels in settled dust and air, and number of cat- and dog owners as well as those who ride in each class were analysed by Spearman's rank correlation, with a two-tailed significance level of 5%.

Results: The geometric mean (GM) allergen level in settled dust was 1530 ng/g for Fel d 1, 1420 ng/g for Can f 1, and 1340 U/g for Equ cx. In Petri dish samples, the GM allergen level was 1.8 ng/sample for cat, 1.3 ng/sample for

dog and 4.7 U/sample for horse. The percentage (GM) of cat-owners was 27%, dog-owners 14% and horse contact 8%. Cat allergen level in dust correlated with cat in air (0.33, p=0.000) and percentage of cat-owners (0.30, p=0.001). Dog allergen level in dust correlated with dog allergen in air (0.27, p=0.003), and percentage of dog-owners (0.40, p=0.000). The same was valid for horse allergen level in dust, which correlated with horse allergen in air (0.37, p=0.000) and percentage of those who ride (0.44, p=0.000).

Conclusion: In all cases, we found a correlation between percentage of animal-owners or riders and allergen levels in dust and air samples. A correlation between dust and air allergen levels has seldom been shown before. We believe that sampling over the whole floor, benches and chairs gives a more representative value. However, dust levels are proxy variables, and air levels may be a better measure of allergen exposure.

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Socioeconomic status and environmental factors affecting cockroach sensitization in atopic children

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Background: Cockroach is the second most common aeroallergen in all over the world. However, there were few studies about the correlation of socioeconomic status or environmental factors and cockroach sensitization in Southeast Asia. The purpose of our study was to determine the socioeconomic and environmental factors affecting cockroach sensitization in atopic children.

Methods: One hundred and twenty children aged 3–15 years attending the allergy clinic, Department of Pediatrics, Faculty of Medicine, Chulalongkorn University, with symptoms of asthma, allergic rhinitis or atopic dermatitis, and with skin test reactivity to at least 1 allergen were enrolled. Questionnaires and skin prick testing results were analysed.

Results: There were 80 boys and 40 girls (average age 9). Eighty one percent of patients had allergic rhinitis, 23% had asthma, and 9% had atopic dermatis. Cockroach sensitization was found in 76% of patients, of which 22%, 12%, 42% were sensitized to German cockroach, American cockroach, and both, respectively. There was no statistical correlation between socioeconomic status or environmental factors, including household income, home's character, home environment, amount of cockroaches seen in home, and cockroach sensitization.

Conclusion: There was high incidence of cockroach sensitization in atopic children. However, socioeconomic status, amount of cockroaches seen in home, and other environmental factors were not correlated with degree of sensitization.

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Asthma severity is influenced by indoor dust mites but not endotoxin or nitrogen dioxide exposure in Hong Kong children

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Background: Endotoxin exposure has dual effects on protecting against wheezing disorders in early life but worsening control in Caucasian asthmatics. Exposure to house dust mites (HDM) and nitrogen dioxide (NO_2) were also known risk factors for severe asthma in this population. However, such data is unclear in Asians. This study investigates indoor exposure to endotoxin, HDM and NO_2 in Hong Kong asthmatic children, and its relation to their disease severity.

Methods: Asthmatics aged 6 to 18 years old were eligible, and spirometry and exhaled nitric oxide (eNO) were measured in the clinic. Home visits were done within 10 days. Dust was collected from patients' mattress, and carers completed ISAAC written questionnaire. House dust Der p 1 and endotoxin levels were measured by enzyme-linked immunosorbent and chromogenic limulus amoebocyte lysate assays, respectively. Average NO_2 levels in kitchens were measured over 48-hour periods by Ogawa sampler badge.

Results: 115 asthmatic patients, aged 12.4 ± 3.2 years, were recruited. Their median FEV_1 and eNO were 92% predicted and 50.5 ppb. Fifty-eight percent of these patients wheezed in past 12 months, and 44% of asthmatics received inhaled steroid. During home visits, 39.2 (24.6-65.9) mg mattress dust was collected. The median (IQR) mattress Der p 1 and endotoxin levels were 0.61 (0.24-2.46) μg/g and 12.4 (6.4-19.5) EU/mg, respectively. A trend was observed for the correlation between mattress Der p 1 and endotoxin load (ρ = -0.176, P = 0.060), but neither factor was associated with indoor NO_2 (P > 0.15). Der p 1 levels = 2 μg/g was found in 29% of families. Kitchen NO_2 levels were 27.1 ± 10.4 ppb, and which exceeded 21 ppb in 78% of families. Indoor Der p 1 levels were significantly associated with night awakening (P = 0.049) and eNO (P = 0.010), whereas mattress endotoxin load was higher in patients with low peak expiratory flow rate (P = 0.031). No association was found between indoor NO_2 and any clinical or spirometric parameter.

Conclusion: Our results suggest that asthma severity in Chinese children is affected by current exposure to HDM allergen in mattress but not indoor endotoxin or NO_2 exposure.

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Effect of indoor mold concentrations on daily symptom severity of children with asthma and/or rhinitis monosensitized to molds

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Little is known about the contribution of indoor molds to the symptoms of asthma and/or rhinitis in children monosensitized to molds. We aimed to investigate the effect of indoor mold spore concentrations on daily symptoms of asthma and/or rhinitis in children monosensitized to molds.

Nine-teen children with asthma and/or rhinitis sensitized only to molds recorded their daily symptoms and PEF values to the diaries, from February 2005 to January 2006. In this study period, indoor mold concentrations were measured monthly from the living/bedrooms.

The median indoor mold concentration was 37.5 CFU/m3. Most commonly recovered indoor molds were Cladosporium (26.4 %), Penicillium (24.7%) and Aspergillus (7%). There was not found significant correlation between indoor mold concentrations and daily rhinitis score (r = -0.021, P = .932), daily asthma score (r = 0.155, P = .554), daily morning PEF (r = -0.056, P= .475) and evening PEF (r = -0.057, P= .471).

The effect of indoor molds is not evident on the symptoms of our patients with asthma and/or rhinitis monosensitized to molds.

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Influence of air quality in schools upon the children with chronic bronchopulmonary diseases

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Aim: To emphasize the influence of air quality in study rooms of schools in children with chronic bronchopulmonary diseases.

Methods: The study planned to interview 1384 of pupils of I-IV class from different schools, for selection of children with chronic respiratory diseases. The lot of study included 54 of children with chronic bronchopulmonary diseases. The clinic examination of these children emphasized 18 cases with bronchial asthma, 9 cases with obstructive chronic bronchitis and in 27 cases simple chronic bronchitis. The hygienic condition of occupation conditions in study rooms of pupils was carried out by means of Air Quality Monitor (AQ-5000, USA), that determines the values of microclimate factors (temperature, air humidity, CO2 concentration).

Results: Hygienic evaluation of occupation conditions in study rooms detected a subnormal thermal regime 15,54 ± 0,1°C, at the beginning of first lesson 16,8 ± 0,2°C and at the end of last lesson (optimal t +20 +25°C). The values of air humidity in study rooms were increasing from 51,11 ± 1,8% at the beginning of first lesson to 56,71 ± 2,3% at the end of last lesson (optimal humidity 30-60%). More significant are the modifications of CO2 in the air, that varies during the day 0,07 ± 0, 009% at the beginning of first lesson and 0, 21 ± 0,02% (p < 0,001) at the end of last lesson (optimal concentration of CO2 < 0,1%. The respiratory functional condition of children with chronic and recurrent respiratory diseases certifies the presence of obstructive disturbances (PEF − 72,3 ± 1,7%; FVC/FEV1 −82,6 ± 2,3%; FEF25−75 −91,7 ± 2,1% and minimal restrictive changes FVC – 76,5 ± 2,1% ; FEV1 −82,6 ± 1,9%.

Conclusion: The reduced temperatures, increased humidity and excessive concentrations of CO2 in child microambience lead to installation and perpetuation of respiratory symptoms in children with bronchial asthma and chronic bronchitis.

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Smoking and asthma-like symptoms in schoolchildren

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Background: The prevalence of bronchial asthma has increased worldwide over the last twenty years. This tendency is particularly valid for the adolescent population.

The aim of the presented study is to evaluate the prevalence of adolescent smoking patterns, parental smoke, exposure to environmental tobacco smoke and the asthma-like symptoms in their relation with active and passive smoking among teenage school-children.

Methods: It is done a cross-sectional questionnaire study (a modified version of Compendium of European Respiratory Standard Questionnaires-CERSQ) with 304 students, aged 15–16 from three secondary schools in Sofia-Bulgaria inquired.

Results: Active smokers are 85 students (28,7%), 45% of them smoke regularly. The average age for starting smoking is 13,2 years and average number of smoked cigarettes per day is 7,4. Regular exposure to tobacco smoke is reported by 201 students (66,3%) and the exposition is between less than 5 years and 15 years. Respiratory symptoms (during the last 12 months): wheezing and whistling in the chest are reported by 22,3% of the students; with in addition shortness of breath in 38.2% of them. Night cough is reported by 39,1% and asthma attack by 2,6% of the examined students. Antiasthma drugs have been taken by 1.6%. The statistical analysis of the questionnaires showed a significant association between "wheezing and whistling in the chest" and "active smoking" ($p < 0,01$); between "night cough" and "active smoking" ($p < 0,01$); "night cough" and "active smoking" ($p < 0,05$); "night cough" and "passive smoking" ($p < 0,01$).

Conclusion: The founded associations support the idea that smoking (both active and passive) plays a role in the initiation, development or maintenance of asthma-like symptoms. The intervention strategies for smoking control should include both public policies elements and preventive measures implemented by health professionals, in particular the general practitioners and school health staff.

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School environment and children health impact in developing country

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Background: Developing country like Nepal has poor health status in school students.

Objective: To know and evaluate the indoor environmental condition of government and private-schools and the health impacts on students.

Methods: A cross sectional studied of representative samples of 35 schools of selected region of Nepal including government and private schools. Onsite observation and health check up & interview with students and teachers were done. Specific scores was given in each criteria. The data were analysed and edited in EPI info program.

Results: The results shows that 89% of government school and 45% of private-schools have poor environmental condition and 69% of government schools students are suffering from environmental-health problem while only 22% of private school student are suffering from some kind of diseases especially allergic. Government-school doesn't have the standard classroom, adequate sports facility, safe drinking water, light and ventilation in comparison with private schools.

Conclusion: We conclude that the main causes are poor socio-economic status, illiteracy of parents, negligence, hard housework for children, diseases, malnutrition, incomplete immunisation and lack of health education. The poor environment condition includes crowded students in a classroom, poor ventilation, shortage of clean drinking water, untidy clothes of students, poor nutrition and lack of greenery in the school area, school near by road, air pollution and lack of environmental awareness among teachers and parents. The government schools have limited budget, resources with compared to private schools and most of the lower and lower middle class family children are studying in government schools which covers nation 82% of total students.

Recommendation: The government should allocate the special budget to the government schools and it should be utilised from the available resources such as good ventilation, limited student in a class, awareness among teachers and parents. Last but not the least, this type of programs are helpful to prevent from environmental health hazards also.

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Air-conditioning bedroom and allergic sensitization in atopic patients

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Background: One of the recommendation of environmental control for allergic prevention was air-conditioning bedroom. The purpose of our study was to determine the relationship between air-conditioning bedroom and allergic sensitization.

Methods: Patients with asthma and/or allergic rhinoconjunctivitis attended pediatric allergy clinic, King Chulalongkorn Memorial Hospital age 3-25 years old and had positive prick skin test for aeroallergen at least 1 antigen were studied by questionnaire.

Results: A total of 180 patients were included, 116 (64.4%) males, 64 (35.6 %) females, 96 (53.3%) lived in households with air-conditioning bedroom. American cockroach sensitization was significantly more prevalent in patients lived households without air-conditioning bedroom (58 %) compared with the other who lived in air-conditioning bedroom (44 %) ($p = 0.048$, Odds ratio 1.726). In contrast, Alternaria sensitization was significantly less prevalent in households without air-conditioning bedroom (3.4 %) compared with air-conditioning bedroom (15.6 %) ($p = 0.006$, Odds ratio 0.2). Polysensitization (skin prick test positive at least 3 antigen) was no significantly related among households without and with air-conditioning bedroom ($p > 0.05$). Cockroach is the second most common aeroallergen sensitization from mite but there was no correlation between amount of cockroach seen in the house and skin test results. There was no correlation between cat and dog exposure or cigarette smoking and allergic sensitization.

Conclusion: Patients living in households without air-conditioning were at increased risk of cockroach sensitization but decreased risk of in mold sensitization.

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Efficiency of neem tree extract (Milbiol) in decreasing the level of house dust mite allergens

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Background: Sensitization against house dust mite (HDM) allergens is common in Estonia and the level of D. pteronyssinus (Der p1) and D. farinae (Der f1) is high in the houses. Diminishing contact with HDM reduces allergy symptoms and therefore it is important to decrease the level of HDM allergens. Milbiol (Hexal AG, Holzkirchen, Germany) is an oil extract produced from neem tree (margosa) seeds. Milbiol is harmless for humans and animals, although it hinders growth and reproduction of mites.

Aim: To estimate the efficiency of Milbiol in decreasing the level of HDM allergens.

Methods: Study period was Dec 2003 - Dec 2004. Study group comprised 44 beds from 19 homes. Twenty-five beds were vacuum cleaned and treated with Milbiol (intervention group) and 19 beds were only vacuum cleaned (control group). Frequency of cleaning was identical and it was performed at the same time for beds of both groups. The dust samples were collected on the 1st, 3rd and 12th month and treatment with Milbiol was performed on the 1st and 2nd

month according to the manufactory instructions. The levels of Der p1 and Der f1 in the dust samples were measured by ELISA method. Data about the living conditions were also collected.

Results: In all beds the level of Der f1 (8614 ± 2654 ng/g) was higher than the level of Der p1 (520 ± 260 ng/g, $p < 0.001$). HDM allergens were more common in old brick houses, on lower floors, in rooms with carpets and in beds older than 5 years. Before and after treatment with Milbiol the mean level of Der f1 was 4180 ± 1502 ng/g and 2784 ± 868 ng/g, respectively; $p > 0.05$, however, after one year the level increased up to 6032 ± 2092 ng/g. The level of Der p1 increased after treatment with Milbiol from 556 ± 353 ng/g up to 784 ± 408 ng/g; $p = 0.005$; although after 12 months the level was lower than before treatment (282 ± 163 ng/g). The mean level of HDM allergen decreased also in the control group beds due to vacuum cleaning - Der p1 from 471 ± 394 to 177 ± 121 ng/g and Der f1 from 14447 ± 5628 to 8733 ± 3700 ng/g, $p < 0.05$; respectively.

Conclusion: Milbiol alone was not effective for decreasing HDM allergens in beds during one-year period after treatment; however, more frequent treatment with Milbiol could give better results. Lower level of HDM allergens was gained also by regular vacuum cleaning.

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The analysis of Alpha1-Antitrypsin serum level among people exposed to industrial dust

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Background: In healthy persons, alpha1 antiprotease serves as a protective screen that prevents alveolar wall destruction. Long-term industrial pollutants exposure may lead to imbalance in the oxidant-antioxidant or proteases-antiproteases systems in the pulmonary tissue witch is followed by unimpeded neutrophil elastase digestion of elastin and collagen in the alveolar walls and progressive emphysema.

Methods: We were interested in comparing the level Alpha1-antitrypsin (AAT) between patients with occupational pulmonary pathology, healthy men, working under industrial pollutants exposure more than 15 years and healthy donors. The subjects in the study were 84 men between the ages of 45 and 60. The main group of study included patients with chronic occupational bronchitis (n = 25), electric welder pneumoconiosis (n = 10), silicoses (n = 29); the first (1) control group included 10 patients without occupational lung disorders and the second (2) control group -10 healthy donors. We measured
the level of AAT by immunoturbidimetric assay for quantitative in vitro determination of AAT in human serum on automated clinical analyzes. We used reagents produced by "Roche-Diagnostics". Normal level 0,9-2,0 g/l.

Results: We found that the level of AAT was under normal among patients with electric welder pneumoconiosis (0,78 ± 0,23). It was significantly lower ($p < 0,05$) than the level in both control groups (group 1-1,23 ± 0,16; group 2 – 1,26 ± 0,16). We did not find significant differences of serum concentration of AAT between silicoses (1,09±0,37), chronic occupational bronchitis (1,15 ± 0,27) and control groups. It was even normal. It was a tendency of AAT concentration increasing due to the term of pollutants exposure.

Conclusion: Long-term industrial pollutants exposure may lead to chronic airway inflammation and caused the imbalance in proteases-antiproteases systems. There was an activation of alpha-1-antitrypsin secretion as the most important proteinase inhibitor among men working under industrial pollutants exposure. The level of alpha-1-antitrypsin was normal among patients with chronic bronchitis, silicoses and healthy workers. We found that AAT level could be decreased among patients with electric welder pneumoconiosis. We supposed that it could be the result of oxidative inhibition of the welding aerosol components on AAT secretion.

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Environmental dust granulometry and microbial load in the presence of tobacco smoke

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Background: Several studies describe damage from passive smoking in humans and animal models. However, how different chemical and physical components relate to mechanisms of damage is not completely understood. Cigarette smoke is composed by a gas phase and a particulate phase, in which a lot of toxicologically relevant compounds are present. We focused our investigations on the particulate phase of environmental cigarette smoke, as the amount and dimensions of suspended particles have been only partially studied so far. It is noteworthy that the size of particles is very important in the inhalation process and in their subsequent deposition into the respiratory tract.

Methods: Differences in granulometry of suspended dust and bacterial load were studied in a 120-cubic meter no-smoking room during and after one cigarette smoke in ten different experiments. To evaluate granulometry in suspended dust we used a 6 channel particle counter, which analyzes particles according to their granulometry. We evaluated suspended bacterial load by using a multipierced sampler on a fixed replaceable plate set to aspirate air. Evaluation of sedimented bacterial load was performed by using the Air Microbial Index technique which expresses the total number of microrganisms settled by gravity onto a Petri plate.

Results: An increase of medium and small, but not large suspended particles number was observed. Bacterial and mycotic load decreased during and up to 60 min after cigarette smoking. Gram-positive cocci never decreased while chromogen-bacteria and mycetes decreases at 30 min. Furthermore, we observed an increase of sedimented bacteria and mycetes at 60 min, a decrease at 120 min and a stabilization at 180 min.

Conclusion: We hypothesize that tobacco burning accelerates sedimentation of larger agglomerates of mean-size smoke particles and bacteria which may be resuspended by movements of people in the room and consequently be inhaled. They could have a stronger affinity with airways surfaces, modifying the normal mechanisms of non-specific and immunologic defenses against microbes, above all in children. This could partially explain the increasing of respiratory infections in children exposed to second-hand smoke.

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Proteomic analysis of birch and ragweed pollen artificially exposed to NO_2

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Background: Air pollutants seems to play a fundamental role in promoting allergies and in worsening the health conditions of allergic people. New interesting findings suggest that traffic-related pollutants do not only irritate the airways, but can also make allergens more aggressive, especially pollen allergens. In particular, air pollutants can induce and/or increase the expression of some allergenic proteins. They can also induce post-translational modifications (PTMs). These changes can make proteins more reactive towards the IgE. Our objective has been to investigate the effect of nitrogen dioxide (NO_2) on pollen from birch (*Betula pendula* Roth) and ragweed (*Ambrosia artemisifolia* L.), two very common and important sources of allergens in Europe.

Methods: Pollen samples were artificially exposed to different concentrations of NO_2. Sera of birch and/or ragweed allergic patients were also collected. Pollen proteins were extracted from control (C) and treated (T) samples, separated by 2D electrophoresis, blotted onto nitrocellulose membranes

and incubated: (a) with patient's sera to identify pollen allergens; (b) with an anti-nitrotyrosine antibody to estimate the entity of protein nitration; (c) with a staining solution specific for glycosylated proteins. Finally, a treatment with lambda protein phosphatase (ëPPase) were performed to identify changes in phosphorylation pattern.

Results: NO_2 treatments did not induce new allergen expression, neither in birch, nor in ragweed pollen. Regarding PTMs, the treatments induced modifications in both birch and ragweed phosphorylation patterns. On the contrary, no changes in glycosylation profiles were detected in the analysed plants. Finally, few nitrated proteins were induced in birch treated samples.

Conclusion: Exposure of pollen to NO_2 induced PTMs in allergens. However, future experiments are needed to confirm these first results.

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Mangifera indica a new pollen allergen

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Introduction: A new pollen allergen was suspected in patients who while receiving allergen immunotherapy showed recurrence of symptoms during the period Jan-March.

Methods: Fifteen patients having various allergic disorders were subjected to skin allergy testing, were found to be sensitive to various allergens and they were on immunotherapy. While on IT they developed recurrent symptoms during Jan-March. To confirm presence of the new allergen, safranin stained glycerin coated slides were exposed in the patients' surroundings with "personal volumetric air sampler". With this "sampler" pollen collection on the slide is much better then simple exposure. Mangifera pollen was recorded in abundance. Antigen of mangifera pollen was tested on all patients and strongly positive reaction was recorded. These patients were subjected to allergen immunotherapy course with mangifera pollen allergen.

Results: Patients who showed positive results with mango pollen allergen and then underwent immunotherapy course with this pollen antigens showed significant improvement in their symptoms during the next pollen season of mangifera indica.

Conclusion: 1) Mangifera Indica pollen could play an important role in naso-bronchial allergy.

2) "Air sampler" makes the detection of causative allergen possible & easier when sometimes simple slide exposure may fail.

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Expression of transcription factor genes for T-cell differentiation in the spleen of C3H/HeN and BALB/c mice by the inhalation of low-level toluene

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Background: Since outdoor and indoor air pollutants are environmental risk factors that have been shown to contribute to the development of respiratory infections and allergy, it is speculated that there is a positive relationship between allergic inflammation and inhalation of volatile organic compounds. Effect of low-level toluene inhalation on Th1/Th2 balance in allergic response remains unknown.

Methods: To study the effect of exposure to low-level toluene on transcription factors for T-cell differentiation, male C3H/HeN and BALB/c mice were exposed to filtered air (control), 5, 50 and 500ppm toluene by whole body inhalation for 6 hours per day, 5 days per week, for 3 and 6 weeks. The allergic model group of mice was injected intraperitoneally with 10 μg ovalbumin (OVA) plus 2 mg alum, and each of these mice was then challenged with nebulized OVA as a booster once every 2 weeks (weeks 2, and 4) during the exposure period. Then, by using a quantitative real-time PCR method, we investigated the expression of GATA3, T-bet and FoxP3 genes in spleen.

Results: Exposure to 500ppm toluene for 3 weeks significantly increased the expression of GATA3 and FoxP3 mRNAs in spleen of C3H mice. In C3H mice exposed toluene for 6 weeks, the expression of GATA3, T-bet and FoxP3 mRNAs was increased at 50ppm toluene as compared to that of filtered air control. However, we did not observe any such pattern for the expression of T-bet and FoxP3 mRNAs in spleen of BALB/c mice. The expression of GATA3 mRNA was significantly suppressed by exposure to 500ppm toluene in BALB/c mice.

Conclusion: This is the first study to show in vivo modulation of the expression of transcription factor genes in the different strains of mice by exposure to low-level toluene, in concert with antigenic stimulation.

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Clinical characteristics of asthmatics susceptible to acute exacerbation by environmental triggers in Cohort for Reality and Evolution of Adult Asthma in Korea (COREA)

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Background: Patients with asthma is prone to experience recurrent exacerbation of symptoms especially when they are exposed to environmental triggers. But it is not well defined what kind of clinical characteristics is associated with this predisposition.

Methods: The Cohort for Reality and Evolution of Adult Asthma in Korea (COREA) study is a prospective, observational multi-center study and will be conducted for nine years (2005-2013) in Korea. Cross sectional data of baseline clinical characteristics were compared between asthmatics susceptible to asthma exacerbation by environmental triggers and asthmatics tolerable to those triggers based on self reported questionnaire at the time of enrollment.

Results: Common cold (22.9%) was most frequent trigger followed by polluted or dusty air (16.6%), second hand smoke (11.5%), emotional stress (9.5%), animal (3.8%), and drug (1.0%) in descending order. The environmental triggers-sensitive asthmatics (TSA) showed female dominance (60.2% vs. 50.5%, P = 0.027) and higher prevalence of rhinosinusitis (63.2% vs. 53.6%, P = 0.041) than environmental triggers-tolerable asthmatics (TTA), while there was no significant difference in obesity, duration of symptom, atopic status, and lung function. More patients among TSA reported aggravation of asthmatic symptoms after moving to current house (16.1% vs. 3.9%, P = 0.001) and TSA had been living in the current house for significantly less time than TTA (P = 0.002).

Conclusion: Female sex, having rhinosinusitis, moving into a new house was significantly associated with the susceptibility to asthma exacerbation by environmental triggers.

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A survey of the children often exposed to Passive Smoking (PS) had relatively higher risk of developing obstructive pulmonary & allergic diathesis

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Introduction: Is to pursue the deleterious effects of (PS) on the health of the exposed children.

Methods: In a retrospective study undertaken from1995-2004, including 600 nonsmoking asthmatic children ages 9-15years (mean age 12 years) with history of house hold smoking as passive smokers (PS). They were likely found to have 60-76% increased tendency of allergic & obstructive pulmonary manifestations (eczema, allergic rhinitis, ocular allergies, middle ear suffusion & shortness of breath) than those residing in a nonsmoking household. Boys were more sensitive to (PS) than girls, and likewise manifestations increased with age and duration of exposure. Children of smoking parents had an increased prevalence of respiratory symptoms (cough, sputum, wheezing) & lower pulmonary function tests (PFTs) values than children of non smoking parents, the association of changes in (PFTs) may persist into adulthood. The significance of the maternal smoking was marked as adding to the population of childhood asthma. The later on follow up has evidenced that passive smoking in early childhood increased the risk of lower respiratory tract infection by 1.5 to 2.2 folds. In the same study/follow up children with congenital pulmonary malformations & family history of allergies were likely to be more sensitive to & exacerbated by (PS) than non smokers.

On further follow up into adult life (PS) were. showing relatively increased prevalence of recurrent infection & less productivity of life.

Results: Underestimation of the significance of (PS) in a house was followed by the grave health related hazards, some of which were irreversible.

Conclusion: The impact of Smoking in a community has variable degree of adverse reactions with paramount morbidity index on terminal ages. Marginlization of smoking to restricted places is followed by a positive socio economic impact.

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The significance of the graded improvement in pulmonary function & physical wellbeing amongst college students with smoking cessation

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Purpose: Significant change in respiratory function/physical well being were documented with stepwise cessation of smoking that had socio-economical & prospective impact upon the college students.

Methods: In the study College students ages 17-21 years both sex having had 1-4 years history of smoking, had manifestation like hacking cough, breathlessness & wheezing more so at night. Some had raised temperature & weight loss altered taste etc. Smoking cessation followed under supervision from weeks to months with advise & encouragement. The outcomes had been as under.

Results: Assessment of the pulmonary function Physical wellbeing,

Conclusion: Immediately after quitting, ex-smokers had been advised breathing exercises with steep line increase in tempo. Improvement in pulmonary function/physical well being had been inversely proportional to the period & history of smoking.

Clinical Implication: Cessation advises followed under supervision with incentives for smoker students in the form of credits in study, awards & certificates for quitters of the week, months etc. khan et al aljh nsr nwfp pk.

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Assessment of pulmonary function Physical wellbeing,			
	6 weeks after quitting		
Pulmonary function(FEV1) on an average<65%	Gradual resumption of taste	Sleep pattern, Slight improvement variable degree of craving for smoking	Physical well being definite improvement observed, slight irritability
	12 Weeks after quitting		
On Average (FEV1) gradually resumed to 63*–65%	Improvement more than before	Refreshing	Much improvement than previous
	18 weeks of smoking cessation		
On an average (FEV!) most had 65%	Significant improvement	No Hangover/Lang over	Significant restoration of vigor

*Rapid restoration in Pulmonary function observed amongst students with relatively shorter history of smoking.

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Ocular, nasal and skin signs of aircrews in relation to before and after the ban on smoking on flights - a follow up study

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Background: The main aim of this 3-years follow-up was to gain further insight into ocular nasal and skin symptoms, and to determine whether the incidence was related to personal, psychosocial, and occupational factors.

Methods: To evaluate the differences of ocular nasal and skin symptoms, a self-administered questionnaire was mailed, in 1997 (n = 1857). Three years later, in 2000 (n = 1446), the questionnaire was sent again to airline crew on duty. The participation rate was 81% 1997 (n = 1513) and 79% 2000 (n = 1145). A cohort of 1134 subjects is used in this study. At the time of the questionnaire study 1997, smoking was allowed on intercontinental flights.

Results: Atopy showed a significant reduction, and hay fever showed a significant increase. During both 1997 and 2000 ocular nasal and skin symptoms were more common in the cabin crew than in the pilots. The incidence of ocular nasal and skin symptoms was more common in cabin crew compared to pilots. The new-onset nasal and skin symptoms were more common in cabin crew compared to pilots. Younger age, work satisfaction, and stress due to excess of work were significantly more related to new-onset ocular nasal and skin symptoms. Cabin crew had significantly more new-onset of nasal symptoms compared to the pilots. Airline crew with hay fewer reported significantly more complaints on new-onset ocular and nasal symptoms. Airline crew with doctor's diagnosed asthma reported significantly more complaints on new-onset nasal symptoms.

Conclusion: Age, hay fever, asthma, psychosocial working environment and type of occupation are related to new-onset of symptoms.

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Atopic diseases, allergic sensitisation and exposure to traffic-related air pollution in children

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Background: In vitro studies, animal experiments and human exposure studies have shown increased risks for atopic outcomes. However, results derived from observational studies are inconsistent. To assess the relationship between individual based exposure to traffic-related air pollutants and allergic outcomes in a prospective birth cohort study during the first six years of life.
Methods: We followed-up 2860 children at the age of 4 years and 3061 at the age of 6 years to investigate atopic diseases and allergic sensitisation. Long-term exposure to particulate matter (PM2.5), PM2.5 absorbance and nitrogen dioxide (NO2) was assessed at residential addresses using geographic information systems based regression models and air pollution measurements. The distance to the nearest main road was used as a surrogate for traffic-related air pollutants.
Results: Strong positive associations were found between distances to nearest main roads and asthmatic bronchitis, hay fever, eczema, and sensitisation. A distance-dependent relationship could be identified with the highest Odds Ratios for children living less than 50 m from busy streets. For PM2.5 absorbance statistically significant effects were found for asthmatic bronchitis, hay fever and for allergic sensitisation to inhalant allergens due to pollen sensitisation. NO2 exposure was associated with eczema, while no association was found for allergic sensitisation.
Conclusion: There is a strong evidence for an increased risk of atopic diseases and allergic sensitisation when children are exposed to particulate matter.

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Air cleaners or polyurethane bedcovers for asthmatic children with cat or dog allergy

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Background: We wanted to see if asthma severity could be reduced by air cleaners in bedrooms and living rooms, and/or polyurethane covered cotton bedcovers on pillows, eiderdowns and mattresses, in children with hospital diagnosed asthma and allergy to dogs or cats.
Methods: Firstly, 60 children accepted invitation. Randomised: 49. Completed: 46. Bedcovers vs. placebo and certified H12/13 class HEPA room air cleaners vs. placebo, in a double blind placebo 4 group design. Inclusion criteria: Asthma, age 7 through 17, allergy to dog or cat. Exclusion criteria: Positive skin prick test (SPT) to house dust mite (HDM); smoking. Only 11 of the first 46 children had a cat or a dog in their homes throughout the trial. Tests: Clinical examination, cold air hyperventilation, symptom score, life quality, indoor environment questions, skin prick tests, specific IgE, serum ECP, dust sampling from mattresses, furniture and air filters. Duration: air cleaners 12 months, bedcovers last 6 months of same period. Secondly, to see if the first results on hyperreactivity and ECP were casual, 31 more patients accepted invitation. 22 patients were included, 21 completed. Randomisation either to active or placebo air cleaners, no bedcovers. Duration: 5 months. Inclusion criteria: Asthma, age 8 through 17, allergy to dogs. Exclusion criteria: Positive SPT to HDM, smoking, and this time, dogs or cats at home. Tests as in first part, but no life quality scoring or dust sampling. Last visit in a colder season than the first for all 67 completing children.
Results: No differences between bedcovers and placebo. Difference between mean ECP (5 missing data) from the last and first visits in the air cleaner placebo groups vs. the active air cleaner groups (0.53 vs. -8.04, p = 0.01, SD = 9.74 vs.12.59). Difference between mean % FEV1 fall at the last and first visits in the placebo air cleaner groups vs. the active air cleaner groups (5.46 vs. -1.09, p = 0.02, SD 11.14 vs. 11.37). No differences by Elizabeth Juniper's Life Quality Scores (PAQLQ and PACQLQ) or symptom scores.
Conclusion: No effects found from polyurethane bedcovers for 6 months in homes of children with asthma and allergy to cats or dogs, without house dust mite allergy. No effects on life quality by air cleaners for 12 months, but significant effects on cold air bronchial hyperreactivity and ECP. Few homes had cats or dogs.

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Methodology for field survey on the impact of indoor environment on allergy and asthma

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Background: A comparative multidisciplinary and inter-regional study on allergy/asthma and indoor exposures could give important information leading to an increased understanding of the aetiology of this morbidity. However, the existing national studies followed different methodologies, which hampered the use of the obtained data for potential comparison. Such drawback can be overcome by following identical protocol with regard to questionnaires, inspections and measurements.
Objective: The present paper proposes a methodology for performing cross-sectional and case-control studies on the impact of indoor home environment on allergy and asthma among children based on the recently performed ALLHOME (Bulgaria) and "Dampness in Buildings and Health" (DBH, Sweden) studies.
Methods: The proposed approach includes a strategy for performing an epidemiological study, selection of target population, identification of the sample size. All key steps in performing the cross-sectional and case-control studies are listed. Suggestions for improving the quality of the obtained data are given.
Results: The DBH and ALLHOME studies identified cultural behaviour as an important factor when performing epidemiological studies. Information gathered from the descriptive cross-sectional study could provide clues leading to the formulation of epidemiologic hypotheses, which could be tested in the analytic nested case-control study. A target group of pre-school children was proposed, since children at that age spend most of their time at home. A cross-sectional questionnaire, developed for the DBH study and containing 87 questions on demographic data, health symptoms, housing characteristics and lifestyle factors was proposed. The use of parental questionnaires for information on housing characteristics could be an effective tool in conducting studies on housing and health. The proposed nested case-control study includes dwelling inspections of a non-destructive type (building checklist, sensory and visual evaluation of dampness, dust sampling for HDM, moulds, pollens, phthalates), engineering measurements (air temperature, humidity, CO2 level) and medical examinations (general examination and skin prick test). Special attention should be paid on storage of collected data, statistical analyzes and dissemination of the research results.
Conclusion: The paper can be used as a guideline for performing studies on indoor environment and allergy/asthma.

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Multiplex technologies - expanding horizons for assessment of allergic sensitization and exposure

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Background: Current methods used for the assessment of allergic sensitization and allergen exposure require separate testing for each allergen-specific antibody or allergen. We have applied fluorescent suspension array technology to allow the simultaneous detection of total and allergen-specific IgE in serum or multiple allergens in environmental samples in a single quantitative test.

Methods: Fluorescent multiplex arrays were developed to allow the simultaneous detection of indoor allergens in environmental samples or total and allergen-specific IgE in serum. A 9-plex suspension array measuring Der p 1, Der f 1, Der p 2, Der f 2, Fel d 1, Can f 1, Mus m 1, Rat n 1 and Bla g 2 was developed using monoclonal antibodies covalently coupled to fluorescent microspheres. A 10-plex array for the detection of total IgE and IgE specific to Der p 1, Der p 2, Fel d 1, Can f 1, Bet v 1, Phl p 5, Bla g 1, Bla g 2 and Asp f 1 was created using monoclonal antibodies and purified allergens covalently coupled to fluorescent microspheres. The multiplex arrays were validated by comparing results between the multiplex methods and established enzyme immunoassays.

Results: There were highly significant correlations between results obtained by the multiplex methods and established enzyme immunoassays with $p < 0.001$ ($n = >70$). In addition, the sensitivity, limit of detection, reproducibility, intra assay coefficient of variance (CV), and inter assay CV of the fluorescent multiplex array was shown to be equal to or better than the ELISA methods.

Conclusion: Our results suggest that fluorescent multiplex technology will facilitate epidemiological studies of exposure and allergic sensitization such as birth cohort studies, population surveys and studies of gene-environment interaction. The arrays can, in principle, be expanded to include other allergens and allergen-specific antibodies.

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Reduction in indoor aeroallergen by using an xtreme 3000 air purifier

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The quality of the environment within buildings is a topic of major importance for public health. Presently Indoor Air Quality (IAQ) is a major concern at various work places. Here is an important quote on air quality from a journal: "With all the publicity, more and more people are realizing that pollutants in the indoor air could make them sick. The worst thing that has happened to the indoor air quality marketplace in the last year or so is also mold. This is because much of the media coverage is designed to sensationalize the topic and frighten the public - so much so, that the word 'mold' always seems to be preceded by the adjective 'toxic.' Thus, homeowners and building managers are scared to death of any minor infestation that might possibly be toxic mold, and they often ignore other health issues, such as combustion byproducts, Volatile Organic Compounds, second-hand tobacco smoke and poor ventilation." This study was aimed at examining the efficiency the air purifier 3000 Xtreme in reducing the indoor spore and pollen counts of allergenic taxa present in different office facilities and residential buildings. Sets of the Petri plates and coated slides were placed with distances of 1foot, 2 feet, 4 feet, 6 feet and 12 feet away from the air purifier. Assay was done with the Petri plates prepared from Brain Heart Infusion agar (VWR) and Gelvatol coated slides with air purifier off for the control and on for the treated. The control Petri plates and slides were placed in the room without using the air purification system. The treated sets were assayed after 24 hour, 48 hour and 72 hour treatment. The slides were examined, analyzed, and photographed using a BX-40 Olympus microscope attached to a DP-70 Digital Camera. Petri plates were examined with an SZ-40 stereo-scope to observe and count the colonies. The control set of plates showed vigorous growth of the microbial colonies after incubation in an incubator at 37°C for 24 hours. Petri-plates closer to air purifier (2 ft. and 4 ft.) produced least number of colonies after 24 h., 48h. and 72 hours of treatment of the indoor air with the air purifier. There was very minor trace of inoculums from the Petri-plates from 2 ft. 4ft. and 8 ft. after 24 hours. After a 2-month continuous exposure of the room air with the Xtreme 3000 at "High" setting we found complete eradication of aeroallergens.

EPIDEMIOLOGY

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Prevalence of allergic disease in eastern Mexico city. A study with ISAAC model

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Background: Wide variations in the prevalence of asthma, rhinitis and eczema have been reported between differents countries. The ISAAC (International Study of Asthma and Allergies in Childhood) was thought in order to maximize the value of epidemiological research into asthma, allergic rhinitis and atopic eczema in childhood, stablishing a standarized methodology and facilitating collaboration and comparison between severe countries.

Methods: A standard questionnaires was distributed through schools and self-completed by 13-14 years old children and by the parents of 6-7 years old children.

Results: 1675 children aged 6-7 years ansewered the questionnaire and 3082 in the group aged 13-14 years. In the group of children age 6-7 years, the prevalence of wheezing in the last year was 8.4% and 5.6% in the 13-14 years group. The prevalence of rhinitis in the last year was 55.5% in the 6-7 years group and 31.4% in the 13-14 years group. The prevalence of eczema in the last year was 9.5% in the 6-7 years group and 6.2% in the 13-14 years group.

Conclusion: Prevalence of allergic disease was similar to reported in other sdudies, except for rhinitis with hight prevalence rates with respect to other studies performed worlwide.

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Alarming results of the comparison of the ISAAC Study in 6-7 and 13-14 years old children in Islamabad, Pakistan

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Background: Allergic diseases and asthma are registering an alarming increase all over Pakistan, especially in children. Pakistan has a population of over 160 million, in which almost 40% are children below the age of 15 years. The ISAAC Study conducted in schools in 1997 and in 2001-2 is the only reliable documented study looking at the prevalence of these illnesses in this country. A comparison of the two study groups has been evaluated to assess the rising danger of respiratory allergy and asthma, in Islamabad, the capital of Pakistan.

Methods: The standard Questionnaire based ISAAC protocol was used for this study. Schools were randomly selected for the study, and all children falling within the study age group were included in the study. Children of age 13-14 years answered the questionnaires themselves, while they were answered by parents of the 6-7 year age group.

Results: Total Number of Children Studied.

Conclusion: The ISAAC study conducted in Islamabad has revealed that the prevalence of nasal problems and wheezing is more than double in the 13-14 year age group, as compared to the younger 6-7 year children. The very large number of 13-14 year olds reporting nasal problems is also very alarming, as there is a 3 fold more danger of these children developing asthma later in life.

Age Group	Males	Females	Total Children
6 - 7 years	1741 (58%)	1276 (42%)	3017
13 - 14 years	1954 (60.3%)	1285 (39.7%)	3239

Number of Children Reporting Wheeze Ever in their life

Age Group	Males	Females	Total Children
6 - 7 years	163 (68%)	77 (32%)	240 (8%)
13 - 14 years	355 (67%)	175 (33%)	530 (16.5%)

Number of Children Reporting Nasal Problems Ever in their life

Age Group	Males	Females	Total Children
6 - 7 years	475 (60%)	318 (40%)	793 (26.3%)
13 - 14 years	1294 (64%)	727 (36%)	2021 (62.5%)

Number of Children Diagnosed as suffering from Asthma

Age Group	Males	Females	Total Children
6 - 7 years	61 (67.8%)	29 (32.2%)	90 (3%)
13 - 14 years	125 (63.5%)	72 (36.5%)	197 (6.1%)

Asthma Diagnosed in those reporting Wheeze Ever

Age Group	Reporting Wheeze Ever	Males Diagnosed as Asthma	Females Diagnosed as Asthma	Total Diagnosed as Asthma
6 - 7 years	240	47 (68%)	22 (32%)	69 (28.8%)
13 - 14 years	530	84 (63%)	50 (37%)	134 (25.3%)

The study also highlights the need for better diagnosis of asthma in both age groups, as only 1 in every 4 wheezing children have been diagnosed to be asthmatic. The results indicate the need for increased awareness amongst the general public and the physicians regarding allergic diseases and asthma.

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Analysis of allergic rhinitis patients according to ARIA guideline in Korea

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Background: ARIA guidelines are in the way of general criterion in classifying allergic rhinitis.

Objective: The objective of study is research on Korean patients who are underwent allergic rhinitis according to their symptoms, severity, frequency and distribution patterns.

Materials and Methods: Statistical analysis was done in 610 patients who had been diagnosed as allergic rhinitis at Seoul National University Bundang Hospital and 540 patients at local clinics. All patients were categorized into 4 groups, mild intermittent, mild persistent, moderate to severe intermittent and moderate to severe persistent groups according to their questionnaire research and allergy laboratory results.

Results: The incidence of moderate to severe persistent group was 34.7 % and moderate to severe intermittent group was 17.1%. In the allergy laboratory results, there was a significant difference between groups in eosinophi count ($P = 0.004$). In questionnaire research, there were significant differences at each group concerning olfaction ($P < 0.001$), self-awareness of rhinitis ($P = .013$), previous history of rhinitis ($P < 0.001$), self-awareness of asthma ($P = 0.001$) and conjunctivitis ($P < 0.001$). There were significant differences in frequency of symptoms for allergic rhinitis and severity between groups ($P < 0.001$) and moderate to severe persistent group showed highest score.

Conclusion: According to ARIA guidelines in categorizing patients, moderate to severe persistent group stands out as the biggest part. Consequently differentiated therapeutic approach between groups will be useful.

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Associations between ocular and nasal symptoms, atopy, personality traits and stress

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The aim was to study associations between ocular and nasal symptoms, atopy, personality characteristics measured by Antonovsky's Sense of Coherence (SOC), Karolinska Scales of Personality (KSP), and stress measured through projective drawing. Totally 194 subjects initially working in 19 buildings with suspected indoor air problems were studied. Information on a history of atopy, hay fever, ocular and nasal symptoms was gathered by a postal questionnaire, including KSP, SOC and projective drawings of "a person in the rain". Results on SOC: There were no associations between SOC and history of atopy or hay fever, however those with low SOC had more ocular ($p = 0.005$) and nasal symptoms ($p = 0.04$). On KSP: Those with atopy had higher social desirability ($p = 0.022$), lower irritability ($p = 0.016$), and lower guilt ($p = 0.047$). Those with hay fever had lower guilt ($p = 0.045$). Those with eye symptoms had lower social desirability ($p = 0.018$), and higher indirect aggression ($p = 0.025$).

Those with nasal symptoms had higher: somatic anxiety ($p = 0.002$), muscular tension ($p = 0.007$), and psychic anxiety ($p = 0.019$). On projective drawings: Those with atopy draw clouds ($p = 0.023$) and long rain strokes (0.039) more often and short rain strokes ($p = 0.005$) less often. Those with hay fever drew water puddles ($p = 0.009$) more often. Drawing of long rain strokes was associated with ocular symptoms ($p = 0.005$). Drawings of strong wind and rain was associated with nasal symptoms ($p = 0.045$).

Conclusion: History of atopy is associated with personality traits measured by KSP and projective drawings. Ocular and nasal symptoms during non-pollen season are associated with SOC, as well as KSP and projective drawings. Different scales were associated with atopy and symptoms. Long rain strokes were consistently associated with both atopy and ocular symptoms.

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Sensitization profile analysis in children with either asthma or eczema alone using a panel of mite allergens from Dermatophagoides pteronyssinus and Blomia tropicalis

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Background and Aim: The prevalence of multi-allergen sensitization to 2 mite species in patients with asthma from Singapore and Malaysia has been reported earlier. It is important to discern whether a specific disease symptom of allergy is correlated with different mite species. The present study is a comparison of the sensitization profiles in children suffering from either asthma or eczema alone using a panel of mite allergens from *Dermatophagoides pteronyssinus (Der p)* and *Blomia tropicalis(Blo t)*.

Methods: A total of 60 children (age between 4 to 16 years old), 30 with asthma and 30 with eczema respectively were recruited in Singapore for the present study. The IgE levels specific to a panel of house dust mite-allergens from two mite species were measured using ELISA. The panel of allergens used is *Der p* extract, Der p 1, Der p 2 and Der p 5 from *Der p* mites and *Blo t extract*, Blo t 4, Blo t 5, Blo t 11 and Blo t 12 from *Blo t* mites.
Results: Children with asthma and rhimitis exhibited the highest sensitization to Blo t 5 (77%) with relatively lower sensitization frequency to Der p 1 (50%) and Der p 2 (67%) while children with eczema showed high sensitization to Blo t 5 (73%), Der p 1 (50%) and Der p 2 (70%). Generally, more eczema patients were sensitized to this panel of *Der p* allergens with much higher measured IgE titres as compared to the asthma group.
Conclusion: These two patient groups tend to show differential sensitization profiles to *Der p* and *Blo t* allergens. Patients with asthma and rhinitis show high sensitization to *Blo t* mite allergens, especially Blo t 5 whereas eczema patients showed high sensitization to Blo t 5, Der p 1 and Der p 2. This suggests the possible correlation between the manifestation of different disease symptoms of allergy and the mite species.

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Sensitization and clinical relevance of ragweed pollen in patients with intermittent rhinoconjunctivitis

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Background: In the past years ragweed (Ambrosia elatior) became of interest in Germany since an increasing distribution of this type-1 allergy causing plant was observed in Europe. We investigated the sensitization against ragweed in patients with intermittent rhinoconjunctivitis and its clinical relevance.
Patients and Methods: 516 patients from North Rhine-Westphalia (Hochsauerland) were tested intracutaneously and specific IgE against ragweed were examined. If the i.c. test turned out positive, a placebo-controlled nasal provocation test was performed. To exclude cross-reactivity with mugwort this allergen was also tested intracutaneously.
Results: 189/516 patients had a positive i.c. test reaction on ragweed. All of them underwent nasal provocation test and 66 revealed a positive result. In 21/66 patients specific IgEs against ragweed could be detected. 40/66 developed also a positive i.c. test reaction against mugwort.
Conclusion: 37% of our patients with intermittent rhinoconjunctivitis had a sensitization against ragweed which was clinically relevant in 35%. 60% of ragweed sensitized patients were also sensitized against mugwort. It remains doubtful whether the clinical relevance of the ragweed sensitization can be explained by cross-reactivity to mugwort taking into account that ragweed has not been detected in the Hochsauerland region.

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Prevalence and evaluation of allergic conjunctivitis in population of buenos aires, Argentina

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Introduction: Allergic conjunctivitis is defined as a bilateral conjunctival inflammation, chronic and recurrent that appear in different times of the year, caused by the direct exposure of the ocular mucosa to allergens present in the environment. Patients who suffer this patology present other associate allergic entities that alter their quality of life.
Objective: Evaluation of patients with ocular allergy pathology by ophtalmologic examination, allergic sensitivity by skin prick test (SPT) with Allergo-Pharma allergens, and serum IgE levels and in tears.
Methods: Population: 64 patients (39 women-25 men) aged between 2 and 75 years with suspected of allergic ocular pathology. Evaluation of allergic sensitivity using allergens such as gramineae, trees, cats, dogs, alternaria, cladosporium, dermatophagoides (d), pteronyssinus (pte) and farinae (fa) using SPT. Measurement of serum IgE levels by RIA and IgE in tears by Elisa method in both eyes. Correlation with other allergic diseases and/or purpose of consultation.
Results: The total number of patients with ocular allergy pathology: 41% belongs > 40 year old, 39% between the 16-39 years old and 20% under 15 years old; 61% females and 39% males. As for the allergy sensitivity (+) for d.pte and d.fa (86%), alternaria and or cladosporium (60%), gramineae (37%), trees (33%), cat (33%) and dog (12%). 45% of patients presented elevated IgE in tear with normal serum IgE and 34% presented both high. The purpose of consultation and/or associated pathology was perennial allergic rhinitis (85%), bronchial asthma (23%), eczemas (14%) and pure allergic conjunctivitis (5%).
Conclusion: We observed through our work that the majority of patients consulted our centre for other allergic entities which aroused the suspicion of this ocular affection by means of a complete interrogation and ophthalmology examination and then confirming with the laboratory and/or SPT. We therefore consider that it is a sub diagnosed entities as it is not the most common consulting, but must be suspected by the Allergist in other allergy pathologies and by the trained ophthalmologist by managing it with discipline to improve the patients quality of life and to avoid after effects.

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The incidence of allergic diseases in different age groups in Western-Hungary 2002-2006

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Rationale: In spite of detailed multi-country cross-sectional surveys focused on allergies (1) there are little comparable data on the distribution pattern of allergy-incidences in different age-groups. Our present aim was-like in a former investigation- to determine it again in the Allergy Outpatient Clinic of Western-Hungary receiving patients from a region located between Western and Eastern Europe with a population of 300.000.
Patients and Methods: Including criteria: Patients with newly diagnosed allergy. Besides clinical investigations allergy diagnosis was always confirmed by skin Prick test and/or specific IgE tests as well. Age groups: Newborns - 96 years of age. Period of investigation: 2002-2006. Total number of patients: 15450
Results: Average total incidence during the investigated period was 160,9. The highest peak was found in the age group of 3-6 years (361,8), the second one was in the age group of 6-14 years (296,9). Adolescents (14-18 y.o) and young adults had nearly the same incidences (average: 289). Further average incidences: 0-3 y.o: 134,7, 30-60 y.o: 197,8; 60-80 y.o: 60,7, over 80 y.o: 7,3.
Conclusion: Peak incidences of allergic diseases were found, like in our former investigation in the age group of 3-6 y.o children. Incidence patterns may not necessarily follow the prevalence distributions of the atopic march. Comparative studies are necessary to determine/analyse regional distribution patterns worldwide.

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The prevalence of allergic diseases in rural and urban regions of Poland - preliminary report

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Aim: The aim of the study was to investigate the prevalence of allergic diseases in two selected, rural and urban, regions of Poland.

Methods: The project consisted of two stages: survey using a questionnaire and diagnostic tests in outpatient clinics. The questionnaire study (adapted ISAAC and ECRHS II) was carried out in a randomly selected groups of children, adolescents and adults (age groups: 6-7, 13-14 and 20-44). One third of the subjects underwent medical examination directed towards allergy recognition. The study was carried out in two regions: in Warsaw - the capital city of Poland and the largest urban agglomeration, with high density of population and dense traffic and in the rural region of southeastern Poland - two districts in the province of Lublin (the districts of Zamosc and Krasnystaw), both characterized by a very low level of environmental pollution, low density of population and natural farming methods.

Results: In 2006 (June - October) 3833 questionnaires were completed. In the rural area the number of answered questionnaires reached the level of 2237 (1105 children and 1132 adults) while in the urban area - 1596 (398 children in age group 13-14, 378 children in the age group 6-7 and 811 adults). Bronchial asthma was declared by 2,6% of all respondents from rural area and by twice as many (6%) from urban area. 14,5% of respondents in Zamojszczyzna and 24,8% in Warsaw declared themselves to suffer from allergic rhinitis. The question concerning skin allergy was answered positively by 23% of respondents from rural region and by twice as many of Warsaw inhabitants.

Table presents questionnaire-based results in relation to age and place of residence.

Conclusion: Preliminary questionnaire-based results may indicate higher prevalence of allergic diseases in Warsaw residents in comparison with residents of rural area of the southeastern Poland. Initial analysis of data from medical examination does not confirm this finding, therefore further analysis is essential.

Age group	20-44	20-44	13-14	13-14	6-7	6-7
Place of residence	Rural (n=1132)	Urban (n=811)	Rural (n=592)	Urban (n=398)	Rural (n=513)	Urban (n=387)
Bronchial asthma	1,9%	5,1%	3,4%	9,1%	3,5%	4,7%
Allergic rhinitis	11,7%	22,7%	18,2%	26,4%	16,4%	27,5%

629 subjects living in rural area and 474 residents of Warsaw underwent verifying medical examination. Bronchial asthma was diagnosed in 5,1% of examined subjects from rural region and in 3,2% from Warsaw. 14,8% of examined rural area inhabitants and 16,5% of Warsaw inhabitants appeared to suffer from intermittent allergic rhinitis, while persistent allergic rhinitis was diagnosed in 12,7% and 6,7%, respectively.

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Allergy for cat and sources of the allergens in the urban versus rural areas of Poland

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Background: The aim is the comparison of a frequency of positive skin prick test for cat allergen, and the frequency of animal presence at home in various areas of Poland (rural versus urban). The presented data are a part of the earliest outcome of large epidemiological study in the field of allergies which is now run in Poland.

Methods: Randomized probe of various polish populations differing in the age range (20–44 13–14, 6-7 years old) were included in to the study. Analyzed urban population was gathered in Warsaw (capital of Poland). The second selected region was Zamojszczyzna, being the typical rural area with low industrial density. 3833 respondents completed questionnaires ECRHS II and ISAAC (1596 at urban and 2237 at rural region). 1103 of them were subjected to medical examination directed towards allergy recognition, and had made a skin prick test (474 at the urban and 629 at rural region).

Results: 65,5% of persons from urban area revealed the positive skin results (for at least one allergen), while of the rural one the prevalence was significantly lower and equal to 34,5% ($p = 0.0001$). An frequency of positive cat prick test is significantly higher for urban region when compared to the rural region (18,4% vs. 10.8), and the intensity of the reaction measured as the average diameter of the observed skin prick test bubble is also significantly higher (4,01 vs. 3,63 mm).

It was found that a significant difference between urban and rural areas was observed for the cat frequency of presence at home 15% vs. 30%. This data can be also compared with the general frequency of the cat presence in the home and surrounding. For rural areas cat is present in about 59.8% of homes and surroundings, while in urban areas cat is present in 15.4% of homes and surroundings.

About 50% of people with positive skin tests for cat have cat at home. In urban area these values are significantly lower and equal to 19.5%.

Conclusion: For cat allergies one can observe that frequency of positive skin prick test is higher on the urban region than on the rural region, contrastively to the lower presence of the animal allergy sources. Additionally the frequency of cat presence in homes of sensitive persons is much higher in rural areas than in urban ones. This observation can lead to the conclusion of protective, immunotherapeutic role of the high environmental allergen concentration in rural region.

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Clinical characteristics of new-onset respiratory allergy in immigrants

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Background: Allergy is influenced in clinical characteristics by genetic and environmental factors. We assessed the clinical characteristics of new-onset respiratory allergy in immigrants in Brescia, Italy.

Methods: We collected the data of all the immigrants referred to our allergy service within the last two years, focusing those patients without family or clinical history of allergic diseases.

Results: Two hundred and 39 patients were evaluated (108 male, mean age 35 years). Their countries of origin were uniformy distributed among the 4 macroareas (Pacific southwest, Africa, South America, East Europe). None of them had previous symptoms of allergy and their family history was negative. The mean time of onset of symptoms after immigration to Italy was 5.21 years, and the first clinical presentation was rhinitis+asthma in 68%, whereas the remaining had rhinitis only. The percentage of monosensitized subjects was 25% and 20% had also sensitization to cockroaches. Only 16% and 6% had a concomitant positivity for foods or drugs respectively.

Conclusion: In immigrants to Italy the new-onset respiratory allergy displays a) long latence, b) preferential onset with rhinitis and asthma associated, c) poor occurrence of sensitization to non inhalant allergens, d) high prevalence of cockroach sensitization.

Keywords: immigrants, respiratory allergy.

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Reported pertussis infection and risk of atopy in 8–12 year old vaccinated and non-vaccinated children

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Background: Pertussis infection has been suspected to be a potential causal factor in the development of atopic disease because of the effect of pertussis immunization on specific IgE antibodies. Although several studies found a positive association between pertussis infection and atopic disorders, this relationship has not yet been studied in a population stratified by vaccination status. Objective of the present study was to assess the association between pertussis infection and atopic disorders in pertussis-unvaccinated children and in pertussis-vaccinated children.

Methods: Using data from a previously conducted study on the relationship between the diphtheria-tetanus-pertussis-(inactivated) poliomyelitis vaccination in the first year of life and atopic disorders, the study population of 1872 8–12 year old was divided into children pertussis-unvaccinated and children pertussis-vaccinated in the first year of life. Within each group the association between pertussis infection and atopic disorders (both as reported by the parents) was assessed.

Results: In the unvaccinated group there were no significant associations between pertussis infection and atopic disorders. In the vaccinated group all associations between pertussis infection and atopic disorders were positive, the associations with asthma (odds ratio (OR) = 2.24, 95% confidence interval (CI95%): 1.36–3.70), hay fever (OR = 2.35, CI95%: 1.46–3.77), and food allergy (OR = 2.68, CI95%: 1.48–4.85) being significant.

Conclusion: There was a positive association between pertussis infection and atopic disorders in the pertussis vaccinated group only. From the present study it can not be concluded whether this association is causal or due to reverse causation.

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Prevalence of allergies and asthma in Pakistan

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Background: Allergic diseases are fairly common in all parts of the world and involve all ethnic groups with bronchial asthma, allergic rhinitis, conjunctivitis and eczema being the commonest manifestations. Despite our efforts, their incidence is on an escalating path. There appear to be no authentic data on the prevalence of allergic diseases in children in Pakistan, therefore the need was felt to carry out the prevalence studies in the country.

Methods: To estimate the prevalence of allergic diseases in our country, this first ever study was carried out in the largest coastal city of Karachi which houses people from all ethnic groups and all socio-economic classes. As there were no statistics available for any age group in this regard, a Saudi Arabian Protocol using questionnaires for school children were used instead of ISAAC Protocol. Questionnaires were distributed to 1200 school children, age 6-15 years, in three different areas for proper proportional representation of high, middle, and low socioeconomic levels. A total of 700 (58.3%) questionnaires returned by the parents were found valid and thus included in the survey. The data was analyzed by using SPSS software.

Results: The result shows that the prevalence of allergic diseases is quite high in Pakistan. The frequency of wheezing was found to be 15.2%, while the diagnosed cases of asthma were 9.5%. The frequency of allergic rhinitis was found to be 34.3%. The frequency of those having allergic rhinitis as well as wheezing episodes was 8%. There was no statistically significant difference between asthmatics and non-asthmatics by sex (P-value:0.402). Socio-economic status was found to affect significantly (p value 0.001) as the prevalence of diagnosed asthma cases was 6.17% in high socioeconomic class, 13.11% in the middle-class and 2.4% in the low socioeconomic class. Family history of atopy was also found to be significantly higher in asthmatics.

Conclusion: Allergic diseases are quite common in Pakistan but they remain under-diagnosed. The prevalence data of allergic rhinitis shows an alarming situation. These diseases were found to affect middle-class the most, while the lower and lower-middle class remains the least affected. In addition to diagnosis and management, prevention and education may help the country to fight back against the disease. Our study is in progress to increase the sample size and compare with the data from other cities being collected under the same protocol.

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Allergic sensitization among Surabaya suburb school children and undergraduate students

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Objective: To identify major allergic sensitization among school children and undergraduate students in Surabaya, a cross-sectional epidemiologic study was done. Participants were recruited by multistage simple random sampling to represent students of Surabaya's suburb areas.

Methods: Only 499 out of 550 invited students were agreeing to be involved in this study in voluntary basis. They were 128 students of primary schools, 221 of secondary schools, and 150 of a university. Complete personal and family history of allergy, allergic symptoms or diseases, environmental or home exposure of various common allergens were obtained, physical examination and skin prick tests were done using a set of 36 different inhalant and ingested allergen extracts. Total serum IgE and specific IgE RAST for selected allergens were also measured.

Results: There was a trend of increasing skin prick test (SPT) positivity among study subjects, being 21.90% among primary school children, to 29.05% among secondary school children, and to 45.30% among undergraduate students. Cockroach (42.85%) and mould (42.85%) were the most common allergens in primary school children, followed by house dust mites (28.57%), grass pollen (17.86%), and crab (14.29%). The order of most common allergens for secondary school children were: house dust mites (63.16%), cockroach (37.85%), mould (28.04%), grass pollen (23.38%), and crab (22.77%). The order for undergraduate students were: house dust mites (58.82%), cockroach (47.05%), crab (41.17%), mould (36.76%), and grass pollen (20.59%). There were some multiple SPT positivity among them, toward 2 or 3 allergens in general. Urticaria and rhinitis were the commonest clinical manifestation of allergy. History of atopy in the family was positive for only 36.7% of respondents.

Conclusion: The prevalence of allergic sensitization among school children and undergraduate students in Surabaya suburb areas were increased, being above the previous estimates for Indonesia in 2003. House dust mites and cockroach were the most common allergen; while urticaria and rhinitis were the commonest allergic manifestation.

Keywords: allergy, sensitization, schoolchildren, skin prick test, epidemiologic study

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Anaphylaxis rates in New Zealand (NZ)

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Background: A high prevalence of atopic disease and asthma has been documented in NZ based primarily on studies in children. No prevalence data is available for anaphylaxis although anecdotally the incidence appears to be high. This study obtains baseline data for anaphylaxis rates in a well-defined adult population in Auckland, NZ.

Methods: A retrospective audit of adults admitted to Auckland City Hospital (ACH) was performed. Patient charts for the audit were identified using a broad range of diagnostic codes that may have been used in cases of anaphylaxis. Four major diagnostic categories were included in the chart retrieval: allergy/allergic reaction, anaphylactic shock or reaction, angioedema/urticaria, and insect sting. Two time periods were selected for the audit, 1st July 2000 to 30th June 2001 (A), and 1st July 2005 to 30th June 2006 (B).

Files were reviewed and data extracted almost exclusively by one author. A cohort of patients that satisfied the Olmstead County definition of anaphylaxis was identified for each time period. Where the diagnosis of anaphylaxis was uncertain the case was excluded.

Interval A included community-onset anaphylaxis only while B included all cases of anaphylaxis.

Detailed data on the population served by ACH is available for both time intervals. This was used to determine anaphylaxis rates in adults (older than 14 years). National population data will be used to determine age-standardised rates for New Zealand.

Results: 367 and 664 admissions were reviewed for intervals A and B respectively. 129 episodes of community-onset anaphylaxis were identified in 121 individuals in A and 113 episodes in 111 individuals in B. The number of individuals per 100,000 adults suffering at least one episode of anaphylaxis in the community per year was 41 (95% CI 34 – 48) in A and 34 (95% CI 28 – 40) in B.

An additional 37 episodes of anaphylaxis occurring in hospital were identified in B. The total rate of anaphylaxis in all settings in B is therefore 46 (95% CI 39-53) per 100,000 adults per year.

Conclusion: The rate of anaphylaxis in NZ adults is one of the highest in reported literature. However this is still an underestimate as it does not include cases managed solely in the community or those misclassified as asthma. In addition in this retrospective study some cases of anaphylaxis may have been excluded due to uncertainty. Despite this it provides us with an important assessment of the potential size of the problem for further studies.

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Knowledge and concerns of treatment and care among Thai allergy patients in Rayong Province, Thailand

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Patient education and health promotion are important to prevent and decrease morbidity and mortality in allergic diseases. Despite improvement of health care system, improper knowledge, attitude and incorrect beliefs on allergy have been worried. To evaluate the knowledge and concerns of treatment and care of allergy would give the information for health care personnel and physicians to develop a good campaign on prevention and care among allergy patients in the future. Self-assessment by questionnaire on knowledge and opinions about allergy and allergist was performed among 125 patients in Rayong Province, which is one of leading centers for tourist and machinery industry. 44.8% know that allergy is an important global health problem and 52.8% also think that it is not severe and life threatening. 39.2% agree to be treated only to relieve symptoms not for continuing treatment and 59.2% need continuing therapy. So 56.8% used the over-the-counter medicine from pharmacy and 4.8% had never used any drug to relieve their allergic symptoms. 80% prefer oral drugs to inhaled drugs but 85% of them will follow the physician advice of using inhaled drug. 53.6% still believe that allergy is permanently cured and about one-forth to third of the patients think that allergic symptoms can be alleviated by nutrient (24%) supplements and air cleaners (34.4%). Though 69.6% know the existence of allergy specialist but few (9.6%) have visited the special clinic and only 7 patients (5.6%) were tested and known their own specific allergens by allergen skin test. However, 89.6% indicated that allergen avoidance is a key role of treatment and 84% know that house dust mite is the most common allergen in Thai population. Only one-fifth (21.6%) satisfy an updated information of allergy and all of them give their concerns on insufficiency of the information provided to their community.

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Allergy and migraine - a significant comorbidity

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Background: Molecular mechanisms of peripheral sensitisation of nociceptors could be identical for painful and pruritic (allergic) diseases. Therefore therapeutic approaches for pain treatment may also be beneficial for allergy. Montelukast, cyproheptadine and ketorolac tromethamine are found to be effective in migraine and allergy. This study is to document the association between migraine and allergic disorders.

Methods: Study spanning 5 years. 10240 migraine patients were questioned about various allergic manifestations.

Results: 7995 (78%) were suffering from various allergic manifestations either at the time of consultation or sometime in the past. Nasal allergy was the commonest 6395 (80%), ocular allergy in 4797 (60%), bronchial allergyin 3198 (40%), and skin allergy in 640 (8%). 1439 were suffering from all 3 allergies(naso oculo bronchial).

Conclusion: Allergic disorders increased the risk for migraine and migraine increased the risk for allergy. This bidirectional association immensely helps the clinician to explain the causative molecular mechanisms and the genetic origin of these two conditions in the most simple way to their patients.

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Dust mite one of the risk factor of allergic asthmatics in developing country

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House dust are considered to be a source of potent allergen responsible for different allergic manifestations. A total of 54 species of mites belonging to 33 genera, 18 families under 3 orders have been recorded from house dust samples collected from asthmatic patients residing in and arround rural areas. All the allergenic mites viz. Dermatophagoides pteronyssinus, D. farinae, Austroglycyphagus sp., Blomia tropicalis., Acarus siro, Glycyphagus domesticus, G.destructor, Tyrophagus putriscentiae except Euroglyphus sp. have been recorded. Among them Dermatophagoides alone contituted more than 60% of the total mites isolated. Patients bed contained significantly higher mite population than the control subjects bed dust ($p<0.02$) and among two different habitats examined i.e. bed and corresponding bed room floor dust, the former contained higher ($p < 0.01$) mite density than the later. Analysis of skin prick test against dust related allergens revealed that 82.4% of the patients showed marked preference towards Dermatophagoides mite allergens. Estimation of total serum IgE level indicates that patients sera contained significantly higher level than the control sera ($p < 0.001$). RAST result further confirms the higher degree of sensitivity towards house dust mites (84.76%). The clinical history of the patients, the physical appearance of allergenic mites in higher concentration in the patients bed, positive skin test results with mite extract and identification of mite specific IgE antibodies in patients' sera by RAST technique established the role of dust mites on the aetiopathogenesis of bronchial asthma in developing country.

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Allergic reactivity to mites and Ascaris lumbricoides in a group of Venezuelan amerindian children

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Background: There was a strongly controversy between the protective or inductor effects of parasites infections and the risk of development allergic diseases in several countries. Moreover, It had been proposed that shared antigens between helminthes parasites and environmental allergens could modulated allergic immune responses. However, evidences for this process has not been consistently establish.

Methods: The aim of this study was to evaluate the allergic reactivity and helminthes parasites coexistence in a group of unselected 61 scholar Children, reflecting a population with a profound socioeconomic depression, a high rate of common childhood infectious diseases such as diarrheas (8344 x 100.00 inhabitants), acute respiratory infections (1528 x 100.00 inhabitants) and high prevalence of helminthic infections (53%). Additionally by ELISA, we detected the levels of th1 (INFγ), and th2 (IL-4) cytokines as well as the levels of total and specific IgE against Dermatophagoides pteronissynus, Blomia tropicalis and Ascaris Lumbricoides. We performed an inhibition specific IgE test between mites and Ascaris lunbricoides in order to establish the cross reactivity between these allergens. We also performed clinical evaluation, hematological studies and skin prick test with common food, mites and parasites allergens.

Results: Sixty seven percent of children (group 1) showed respiratory symptoms or skin manifestations. Fifty % had high leukocyte count and had high INFγ levels (4190-5320 UI/ml) than control group. Skin prick tests were positive in 43% of children to at least one of the common allergens evaluated: 77% were positive to Ascaris lumbricoides, 27% to Blo t and 27 % to D.pt. Fifty % had high total IgE levels, (3812 - >10000UI). Eighty one percent showed levels of specific IgE higher than 17.5 UI/ml to Ascaris and 67% to D.pt. Before the inhibition test the median levels of specific IgE to Ascaris diminished from 14.17UI/ml to 3,43UI/ml % and to D.pt from 12,94 to 3,37 UI/ml.

Conclusion: These results indicates that high allergic reactivity, (evidenced by clinical, skin prick test and specific IgE), and high levels of Th1 cytokines were both been present in sera of children evaluated. We also found that there were an inhibition in the level of specific IgE in the sera of these children, suggesting a cross reactivity between mite and Ascaris allergens.

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The approach to improvement diagnosis of lambliasis in patients with atopic bronchial asthma

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Backround: Since 1859 when has described first time *G. lamblia* as the activator of disease of a gastroenteric path, the huge material of epidemiology and pathogenesis of lambliasis and as effective enough ways of therapy of the given disease have been developing and collecting. But also at the moment, concerning this disease, there are the numerous questions expecting the decision. Most important of this group of problems, intensively discussed problem of diagnostics and prevalence *G. lamblia* among people is.

Methods: We use commercial ELISE-test and the immunochromatography methods revealing specific antibodies in whey of blood and antigen's in excrements. They possess more sensitivity (66,3 – 98,9 %) and high specificity (92,6 %) unlike routine microscopic methods in diagnosis of lambliasis. In connection with before be the aforesaid, till now the most widely applied test in laboratory diagnostics of lambliasis, such as a microscopy of sedimentary components of excrements or researching contents at duodenal sounding for the purpose of revealing cysts of *G. lamblia*. In the literature as the method enrichment formalin-radio is recommended. Unfortunately, the specified tests of laboratory diagnosis of lambliasis possess low sensitivity, subject to influence of some subjective and objective factors among which, such as changeable allocation cysts of *G. lamblia*, difficulty for microscopic identification.

Results: We used the combined method of research of excrements This method allowed to improve essentially delectability cysts *G. lamblia* in comparison with a routine method microscopy of fecal. We will especially notice, that at comparative research of tests in which activators lambliasis have been revealed and using three components a method and microscopy research of fecal in the first case the big degree invasion came to light. It has paramount value at definition of necessity of carrying out of therapy lambliasis.

Conclusion: As using combined method using three components for flotation of research of excrements, we managed to increase essentially delectability *G. lamblia* and as to reduce quantity of repeated researches necessary for the exact diagnosis of lambliasis. Accuracy of definition of degree invasion an organism this elementary has besides, increased, that has allowed to correct therapy.

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Titanium used for fabrication of obturators after maxillectomy -an alternative therapeutic option in cases of incompatibility of polymer based denture materials

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Background: Microbiological colonization and subsequent biofilm formation on denture materials play an important role in the development of denture stomatitis often triggering allergic incompatibilty of denture materials as the penetrability of the mucosa is changed. Despite the high standards in reconstructive surgery and even with the use of free vascularized flaps post-maxillectomy rehabilitation with obturator prosthesis is still the mainstay reconstructive procedure in several cases. With current use polymer based silcone- or methacrylate-obturators bear the risk of an increasing settlement and penetration of bacteria and thus the risk of local or even systemic infections. Titanium-obturators [TO] for oral rehabilitation offer favorable characteristics concerning growth of microbial biofilms. The aim of our study was to compare the prevalence of microbial contamination on titanium- and polymer based -obturators [PO] used for post-maxillectomy rehabilitation.

Materials and Methods: We investigated the obturators of 36 patients (17 TO, 19 PO), all had undergone maxillectomy due to malignancy in the period 1999-2204. Microbiological samples of the obturator surfaces, the adjacent mucous surfaces and of the interior of the PO (when new fabrication was planned) were collected with a sterile swab. Microbiological analysis was performed by conventional methods, including microscopy and culture on different agars. For statistical data analysis Fisher's exact test [FT] (P-value $P \leq .05$) and odds ratio [OR] (95% confidence interval) were used.

Results: PO showed a significantly higher quantitative contamination and a shift to non physiological bacteria compared to TO ($p = 4.944e-06$ [FT], 6.84 [OR]). There is a high statistical risk (factor 1.4) for penetration of bacteria to the interior of PO when the pathological bacteria are seeded on the surface of PO ($p = 0.01966$).

Conclusion: The use of titanium-obturators for the oral rehabilitaton of post-maxillectomy patients significantly reduces the risk of contamination with pathological oral bacteria and therefore prevents patients from local oral or systemic infections. Especially in cases of incompatibility of polymer based denture materials titanium-obturators offer an alternative therapeutic option.

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Atopy history in family associated with persistent allergic symptoms in Thai population

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Allergy has been estimated about 10–20% of Thai population. Most studies conducted in Bangkok and other regional centers showed a increase trend of allergy prevalence in a recent decade. This study is aimed to determine the prevalence of allergic diseases in an urban area, Rayong province, which has high growth of industry and oil/gas refineries. 524 Rayong residents, with mean age (± SD) of 26.26 (± 3.77) years old and male 59.4%, were asked about their history of allergy regarding rhinitis, asthma, conjunctivitis, dermatitis including urticaria, food allergy and drug allergy. Prevalence of overall allergy is 15.6 %. Persistent rhinitis, asthma and dermatitis are 12.3 %, 3% and 2.3%, respectively. About urticaria, 9.5% reported history of acute (78%) and chronic type (22%). 4.4% revealed history of food allergy of which the most common culprit was sea-food allergy (73.9%). 2.7% showed history of insect sting allergy in which 46.2% of them had large local reaction and 3 of 13 had anaphylactic reaction. Drug allergy is 2.3% with history of allergy to beta-lactams (25%), NSAIDS (25%) and paracetamol (16.7%). Family history of atopy is significantly associated with history of rhinitis ($P < 0.0001$), asthma ($P < 0.0001$), urticaria ($P = 0.001$), conjunctivitis ($P = 0.029$) and food allergy ($p < 0.0001$). There is no association between history of allergy and sex, age or occupation. There is also a close association among chronic rhinitis, asthma and food allergy ($P < 0.0001$). In conclusion, allergy is not uncommon even in an urban area like an eastern part of Thailand. History of atopy in family is useful to be a predisposing factor of allergic diseases.

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Effect of prenatal and postnatal exposure to low-level toluene and peptidoglycan on systemic Th1/Th2 immunity in infant mice

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Background: Toluene, a widely used aromatic organic solvent, has been well characterized as a environmental toxic chemical. Volatile organic compounds including toluene are the possible substances causing chemical sensitivity in human. Our laboratory has previously showed that the exposure of low-level toluene causes immunological and neurological modulation in adult mice. Recently, it has been suggest that TLR2/4 ligand such as peptidoglycan (PGN) or LPS modulates Th2-driven allergic immune responses in animal studies. The aim of the present study was to investigate the effect of low-level toluene exposure and PGN stimulation on systemic Th1/Th2 immunity in infant mice.

Methods: We exposed pregnant BALB/c mice to filtered air (0 ppm), 5 ppm, and 50 ppm toluene for 6 h/day on 5 days/week for 1 week (from day 14 of gestation until delivery) in the whole body exposure chamber. The offspring were further exposed to toluene of same schedule for 3 weeks (from the day of birth up to day 21). Some of the pregnant mice were repeatedly exposed to aerosolized PGN 200μg/10 ml (3 times per week) during the toluene exposure period. Offspring from the toluene and PGN exposed pregnant mice were further repeatedly administered PGN 100μg (5 times per 2 weeks) intraperitoneally from day 7 up to day 21 during the toluene exposure period. One day following the last toluene exposure, we collected blood, spleen, and lung from each mouse. Then, total IgE, IgG1, and IgG2a antibodies in plasma and the production of cytokines (IL-12, IFN-γ, IL-4, IL-5) in spleen and lung were assayed by ELISA method. Also, the expression of cytokine and transcription factor T-bet mRNAs in spleen was analyzed by real-time RT-PCR method.

Results: Exposure of mice to low-level (5 or 50 ppm) toluene resulted in (1) increased IgG1 and decreased IgG2a antibodies in the plasma; (2) decreased T-bet mRNA significantly in spleen; and (3) decreased IFN-γ and IL-12 mRNA in spleen but not statistically significant. Next, exposure of mice to low-level toluene by combination with PGN stimulation compared to toluene alone resulted in decreased IgG1 and IgG2a antibodies in the plasma and IL-12 mRNA in spleen.

Conclusion: These findings suggest that low-level toluene exposure from fetal stage to neonatal stage may enhance the systemic Th2 function in infant mice. However, the exposure to combination of low-level toluene with PGN stimulation shifted Th1/Th2 immunity to suppressive direction.

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Risk factors for recurrent wheezing in infancy

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Background: Although recurrent wheezing is frequent, there are few data regarding risk factors associated with this condition. The aim of study is to verify the risk factors for recurrent wheezing in infants, in the south of Brazil.

Methods: Cross-sectional study using a standardized and validated questionnaire with 45 questions about recurrent wheezing and familiar, environmental, socio-economic and respiratory infections risk factors in the first twelve months of life, from International Study of Wheezing in Infants (EISL, from spanish: Estudio Internacional de Sibilancias en Lactantes). Parents of infants that attended the Health Centers between August/2005 and December/2006 for regular immunization were interviewed. Among 107 Immnunization Centers 35 were randomly selected in order to maintain a homogeneous selection of population sample. Factors associated to recurrent wheezing in a univariate analysis were studied using multivariate regression of Poisson. Prevalence ratio and confidence intervals of 95% were calculated due to a high prevalence of wheezing infants and statistic was considered significant when $p < 0.05$.

Results: Three thousand and three infants, aged 12-15 months-old were studied. The prevalence of recurrent wheezing was 45.4%. There are associations between recurrent wheezing and male gender, mother, father and siblings asthma history, mother's instruction (secondary and advanced), others pets in domicile during pregnancy (birds), 6 colds episodes or more, personal history of dermatitis and molds in home. Updated immunization was a protection factor for recurrent wheezing.

Conclusion: Prognostic risk factors for recurrent wheezing in the first year of life were male, familiar history of asthma, high mother's instructions, other pets during pregnancy and molds in home, a high number of colds episodes, and personal history of dermatitis. Interventions acting on these factors may decrease the occurence of recurrent wheezing in infants in Curitiba, Brazil.

FACTORS	PR	95% CI	p-value
Male	1.14	1.05–1.24	0.002
Mother's asthma history	1.18	1.04–1.33	0.007
Father's asthma history	1.20	1.05–1.39	0.008
Sibling's asthma history	1.23	1.08–1.42	0.002
Mother's instruction (Secondary)	1.39	1.24–1.55	<0.0001
Mother's instruction (Advanced)	1.20	1.07–1.35	0.001
Other pets in domicile during pregnancy (birds)	1.28	1.07–1.54	0.007
≥6 colds episodes	1.32	1.21–1.44	<0.0001
Personal history of dermatitis	1.09	1.003–1.19	0.04
Molds in home	1.14	1.04–1.24	0.003
Updated immunization	0.79	0.63–0.98	0.03

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Monitoring of cytokines in breast milk from allergic and non allergic mothers: prospective birth cohort study

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Background: It is well documented that human milk contains several immunomodulatory components which are important during the newborn's immune system development. In this context, breastfeeding could provide the best possible nutrition. Its role in the development of allergies remains although controversial, due to possible individual differences and variations in the composition of the breast milk. Furthermore, other stimuli could interact with immune system during perinatal-neonatal period, interfering with breast feeding and hence with the immune system development. For these reasons, we are interested to investigate the different components (environmental, diet, familiarity etc.) that could be involved in the allergic diseases apperarence.

Aim: The aim of this prospective birth cohort study is to investigate the concentration of cytokines involved in allergic reactions, IgA antibody production, soluble (s) CD14 and fatty acids in cord blood, colostrum and mature milk, from allergic and non allergic mothers. Moreover, the aim is to investigate whether breastfeeding practice, correlated with immunological profile, is determinant for the increased risk of atopic disease development in children. 32 mothers (17 allergics, 15controls) and their full term newborns were included in the study, until now. We measured cytokines in colostrum samples (2 days after birth), and in transitional breast milk samples (5 days

after birth). Children underwent paediatrician evaluation till now at 6 months of life.Levels of cytokines (IL-4, IL-5, IL-6, IL-10, IL-13, TGFBeta1, TGFBeta2,TNF alfa and beta) were determinated by commercial ELISA kits.
Results: Different cytokines levels in colostrum and milk samples, were evidenced between allergic and non allergic mothers. In particular, our preliminary data showed that colostrum from allergic mothers has a significant increased level of IL-4, IL-13, IL-5, TGF beta 1 e 2 compared to samples from non allergic mother, while no differences in cytokines level were revealed in the milk samples from the same two groups. Follow-up is in progress, first round of clinical evaluation 6 months after birth, does not reveal yet any allergic symptoms in all the children.

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Kawasaki disease and allergic disease - a sibling control study

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Background: It has been shown that Kawasaki disease (KD) may increase the risk of developing allergic diseases. We investigated the relationship between KD and allergic diseases using a sibling control study design in order to minimize the impact of familial effect confounders.
Methods: Children with a history of KD along with their healthy siblings were recruited from the two major paediatric hospitals in Singapore. All subjects undertook the standardized modified ISAAC questionnaire for allergic diseases and underwent a detailed physical assessment as well as skin prick testing (SPT) with a standardized panel of allergens.
Results: Eighty-one KD-sibling pairs were studied. There was no statistical difference in gender and age distribution between the KD and the healthy sibling controls. KD patients had an overall increased risk of allergic diseases (Odds Ratio [OR] 3.29, CI 1.36–9.07, p = 0.005) particularly allergic rhinitis (OR 3.13, CI 1.37–8.02, p = 0.005). KD patients also had a higher risk of eczema (OR 1.38, CI 0.50–3.94, p = 0.648), asthma (OR 1.80, CI 0.54–6.84, p = 0.424) and allergic conjunctivitis (OR 1.33, CI 0.41–4.66, p = 0.791), although the differences were not statistically significant. The risk of sensitization against common allergens was also higher, although not statistically significant, in KD patients (OR 1.33, CI 0.62–2.82, p 0.500), with risk of positive SPT highest against cockroach allergen (OR 3.33, CI 0.86–18.85, p = 0.092) followed by Blomia Tropicalis (OR 1.41, CI 0.73–2.80, p 0.349). There was no significant difference in wheal size against histamine and allergens between KD patients and their sibling controls.
Conclusion: Our study suggests that KD may be an independent risk factor in the development of allergic diseases, particularly allergic rhinitis.

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Serum total IgE levels and type of sensitization may predict asthma onset in patients with allergic rhinitis

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Background: Recent epidemiological surveys suggest that allergic rhinitis (AR) and asthma are intimately linked while AR is considered as a risk factor for asthma development.
Objective: To investigate retrospectively the predicting value of total serum IgE (tsIgE) and type of sensitizations in the onset of asthma to individuals with allergic rhinitis (AR).
Patients and Methods: Serum total IgE analysis and Skin Prick Test (SPTs) to 9 different, representative for Mediterranean populations, aeroallergens (grasses, parietaria, olive, d. farinae, d. pteronyssinus, alternaria, cladosporiom and cat) were performed to 1439 adults (20 to 60 years old). The adult-sample was considered representative since it consisted of all staff of the Greek Air Forces who were examined (after written consent was obtain from all) during their obligatory annual medical check up. A mean wheal diameter of 3mm or more, in the absence of any reaction in the negative control SPT, was considered as a positive reaction. Multiple logistic regression was performed to reveal any statistical significant association between type of respiratory allergy (AR and/or asthma) and all known parameters (gender, age, type of sensitization, tsIgE levels). Thereafter the diagnostic properties of tsIgE and type of sensitization in discriminating between rhinitis and asthma were analyzed using the Receiver Operator Characteristic (ROC).
Results: Patients with allergic rhinitis sensitized at least to indoor aeroallergens have an increase risk of asthma onset 8.46 times [Confidence intervals (CI) 3.28–21.8, p < 0.0001], while this risk increases 1.36 times more for every increment of 100kU/lt of the tsIgE (p < 0.0001). These two factors provide an excellent discrimination between those with AR and AR and asthma (ROC Area Under the Curve: 0.8319, CI:0.7813–0.8825).
Conclusion: Type of sensitization and serum tsIgE seem to be promising significant predictors of asthma development in patients with AR, although they should be evaluated prospectively in future longitudinal studies.

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Allergic migraine

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Background: Molecules involved in allergy and migraine may be identical.This study is to document oculonasal allergies triggering migraine attacks.
Methods: 4 year study, 28 patients aged 15 to 50 years. Migraine diagnosed according to ICHD 2 diagnostic criteria.
Results: 22 patients reported severe allergic rhinitis and 6 with severe ocular itching precipitating headpain attacks resembling migraine 24 of them were also getting migraine attacks when exposed to common migraine triggers in this region. These common triggers are exposure to sunlight, travelling by bus, sleep disturbances and missing meals at the right time. Travelling by bus triggered allergic symptoms in 21 of these patients most probably due to exposure to dust and smoke.
Conclusion: Molecules involved in allergic inflammatory processes definitely trigger migraine episodes by exciting peripheral nociceptors.

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Familial allergy in Surabaya's suburb schoolchildren

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Objective: To evaluate the influence of parental allergy to their offspring, data of 358 schoolchildren were analyzed. They were parts of an epidemiologyc study of allergy in schoolchildren and undergraduate students done in Surabaya's suburb areas.
Materials and Methods: A questionnaire was filled by the children with the help of the researcher. Another questionnaire was filled by both of their parents. Complete personal and family history of allergy, allergic symptoms or diseases, environtmental or home expossures of various common allergens were obtained. Skin prick test (SPT) were done to the entire children and their accompanying parent(s), using a set of 36 different inhalant and ingested allergen extracts.
Results: Of 358 schoolchildren, only 163 were accompanied by both of their parents, so that the data could be analyzed. There were 60 children with SPT

(+), among them 22 mothers only (36.67%), 14 fathers only (23.33%), and 9 both parents (15%) had positive SPT, while 15 (25%) were both negative. Among 103 children with SPT (–), there were 23 mothers only (22.33%), 17 fathers only (16.50%), and 14 both parents (13.59%) had positive SPT, while 49 (47.57%) were both negative. Atopic status of the mother had stronger influence on their children's SPT results (Pearson chi-square = 4.058, p = 0.044), while atopic status of the father did not (Pearson chi-square = 1.057, p = 0.304).

Conclusion: Compared to the father, atopic status of the mother had signifycantly stronger influence on the schoolchildren SPT results.

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Degree and clinical relevance of sensitization to common aeroallergens in adult respiratory allergic population of Calcutta City

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Background: The aim of the study was to assess the types of allergic sensitization, risk factors and clinical impact of being sensitized in the adult respiratory allergic population of Calcutta City, India.

Methods: In the period of 2003–05, 1353 adult respiratory allergic subjects from Calcutta were tested (skin prick tests) for common aeroallergens including 29 pollen types, 19 mould spore types, house dust mites and animal dander. All subjects were interviewed on symptoms and diseases including respiratory irritants and childhood environments.

Results: The maximum sensitivity was recorded for *D. farinae* (94%), grass pollen (76.55%) and *Fusarium* spore (53.92%) among the three groups of allergens tested. The prevalence of at least one positive prick tests was 37.12%. A large difference by age was found: 58.9% were sensitized in the age group of 20–29 years, 42.8% in the age group of 40–49 years and 28% in the age group of 50–60 years ($p < 0.001$). Sensitization to multiple allergens was common in the age group of 20–29 years with 42.1% of the sensitized responding to at least four allergens, while this portion was only 12.8% of the sensitized among those aged 50–59 years. The prevalence of physician-diagnosed asthma, allergic rhinitis (AR), conjunctivitis and wheeze increased significantly with increasing number of positive skin responses. Having a family history of AR or conjunctivitis was a significant risk factor for allergic sensitization and for sensitization to any of the pollen grains. Further, urban living in childhood added an increased risk for pollen sensitization.

Conclusion: The prevalence of allergic sensitization was high in the urban adult population of Calcutta. More than half of those aged 20–29 years were sensitized and 42.1% was sensitized to at least four allergens. Sensitization to multiple aeroallergens was associated with a high prevalence of asthma, AR or conjunctivitis and wheeze.

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Association of HLAG gene polymorphism and house dust mite respiratory allergy in an Algerian patient group

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HLAG, a nonclassical MHC class Ib molecule, plays a significant role in immuno-tolerogenic processes. HLA-G expression through alternative splicing generates membrane-bound and soluble isoforms secreted by activated (by IL-10) peripheral blood CD14+ monocytes. Genetic diversity of HLA-G consists of sequence variations in promoter, exonic and 3'untranslated (3'UTR) regions, some of which have been shown to affect HLA-G mRNA stability. Polymorphisms in the HLAG promoter region had previously been associated with asthma. Of interest is also the fact that the gene encoding the effector IL-10 also exhibits genetic polymorphism in populations. Given the plausible effect one over the other, in this study, we investigated the potential role of a 14-bp insertion/deletion polymorphism in the HLAG-3'UTR region in conjunction with IL10 gene promoter polymorphisms (-1082 A/G, -819 C/T and -592 C/A) on respiratory allergy development. The association study involved 125 patients suffering from asthma and/or rhinitis, all monosensitized against Derp 1 and 74 matched-controls, all originating from Algeria. PCR-based techniques were used to assess the HLAG and IL-10 genotypes. While the IL-10 promoter allele or genotype frequency did not differ between patient and controls, the 3'UTR +14-bp insertion allele was significantly more frequent in the patient group than in the control group ($Pc = 0.01$) with the homozygous state of the 3'UTR -14-bp deletion allele being more prevalent in the healthy controls ($Pc = 0.04$). Our findings suggest that HLA G gene may participate in the pathogenesis of respiratory allergic diseases in our population. A possible mechanism that could be envisaged is that the diminished immunosuppressive activity, mediated by HLA-G, may contribute to initiation/persistence of chronic airway inflammation in asthma.

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Sensitization to ascaris spp. influences total and specific IgE levels to dermatophagoides pteronyssinus and blomia tropicalis

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Background: Although sensitization to *Ascaris* may not play a role in the severity of asthma, it may be implicated in total and specific IgE levels to common aeroallergens.

Objective: To determine sensitization to the mite species *D. pteronyssinus* and *B. tropicalis* in patients who have a positive, or negative specific IgE determination to Ascaris spp.

Methods: We analysed 852 consecutive patients (417 males and 435 females; mean age 18.5 years; ranging from 9 months to 82 years. 518 children (302 boys and 216 girls) with allergic asthma and/or rhinoconjunctivitis with or without eczema were included. Patients were separated into 2 groups. Group A consisted of *Ascaris* spp. positive patients and Group B of patients with a negative specific IgE determination (Phadia) to *Ascaris* spp.

Results: A total of 370 individuals had a positive specific IgE determination to *Ascaris* spp. (>0.35 kU/L), and 482 had a negative result. Specific IgE to *D. pteronyssinus* was positive in 598 patients and to *B. tropicalis* in 608. Mean total IgE level was 840.35 kU/L. In group A, mean total IgE level was 1490.31 versus 341.42 in group B ($p < 0.001$). In group A, 345 patients were positive to *D. pteronyssinus* with a mean specific IgE level of 45.0 kU/L. In group B, 253 patients were positive, with a mean specific IgE level of 26.41 kU/L ($p < 0.001$). In group A, 354 patients were positive to *B. tropicalis* and the mean specific IgE level was 42.28 kU/L. In group B, 254 patients were positive (Chi square $p < 0.001$), with a mean specific IgE level of 12.76 ($p < 0.001$).

Conclusion: Sensitisation to *Ascaris* spp. seems to play an important role in rising total, as well as specific IgE levels to *D. pteronyssinus* and *B. tropicalis*. Statistically, more patients were sensitized to mites in the *Ascaris* spp. positive than in the *Ascaris* spp. negative group. The clinical significance of these findings remains to be established.

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In vitro sensitization to Ascaris spp. in a population of allergic individuals in the caribbean island of Martinique

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Background: Sensitization to allergens of the nematode *Ascaris* spp. is common in the tropics, where a large proportion of the population has been in contact with this, and other intestinal parasites. It has been suggested that sensitization to *Ascaris* spp. allergens may influence the allergic response to other allergens.

Objective: To study specific IgE levels to *Ascaris* spp. in a large population of individuals consulting for allergic respiratory complaints in an Allergy Clinic in Fort de France, Martinique.

Materials and Methods: This study has a prospective, cross-sectional design and was conducted from January 2003 to March 2007. A total of 1,060 consecutive patients (507 males and 553 females; mean age 20.41 years; 6 months to 88 years), evaluated for allergic respiratory complaints were entered in the study. Specific IgE to *Ascaris* spp. was measured by the CAP method (Phadia). Results were stratified by age and sex.

Results: A total of 431 individuals; 239 men (47.14%) and 192 women (34.72%) were positive to *Ascaris* spp. When analysing differences by age, a total of 599 children with <15 years of age were included (349 boys and 250 females). In this population, 271 children (45.24%) were positive, including 169 boys (48.42%) and 102 girls (40.8%). In the group of patients with >15 years of age (461 individuals; 303 females and 158 males), 160 (34.92%) were positive, including 70 men (44.3%) and 90 females (29.7%). Overall, there was a steady decline in the percentage of positive results in males and females until the age of 44. After this age, however, there was a sharp increase in the prevalence of positive results in males.

Conclusion: This study demonstrates a high prevalence of sensitization to *Ascaris* spp. in the Caribbean island of Martinique and confirms that approximately 40% of patients seeking diagnosis, and or treatment, for allergic respiratory diseases are sensitised to *Ascaris* spp. allergens. In younger children, especially in boys, this prevalence is closer to 50%. Although antiparasitic treatment may be required in these patients, we have not confirmed if specific IgE to *Ascaris* spp. represents current infestation or the effect of previous antiparasitic treatments, since the majority of children are systematically treated against intestinal parasites in Martinique.

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Methylation status in the promoter region of IL-12 gene might be correlated with allergy

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Background: Helper T cells can be classified into Th1 and Th2 subtypes, which have been shown to contribute to cellular immunity and humoral immunity, respectively. It is currently thought that impairment of the balance between Th1 and Th2 cells may cause immune diseases, including allergic diseases. Th1 and Th2 cells are polarized from naive T cells following their contact with mature dendritic cells (DCs), which are antigen-presenting cells resident at the frontline of immunity. Only mature DCs can activate naive T cells. When the contacted DCs secrete interleukin (IL)-12, the naive T cells become Th1 cells. In contrast, when the contacted DCs secrete IL-4, the naive T cells become Th2 cells. Therefore, the type of cytokine secreted by the DCs is very important. DCs generated from cord blood were previously shown to secrete lower amounts of IL-12 than DCs from adult peripheral blood. The authors suggested that this finding was correlated with the immune status of infants, who have weak cellular immunity and develop allergic diseases easily.

Objective: We hypothesized that reduced secretion of IL-12 by DCs from both allergic patients and cord blood would be due to methylation of the IL-12 gene promoter region, and that the allergic status may be attributed to delayed demethylation of this region. To clarify this hypothesis, we examined the methylation status of the IL-12 gene promoter region in monocytes (DC precursors) prepared from peripheral blood from healthy adults (HA) and allergic adults (AA) and from cord blood from normal infants (NI).

Methods: Monocytes were purified from each of the three groups using a MACS system and an anti-CD14 antibody. Genomic DNA was extracted from the monocytes. The methylation status of the IL-12p35 gene promoter region was investigated using a bisulphate-sequencing analysis method. In addition, real-time RT-PCR amplifications were carried out to determine the IL-12p35 mRNA levels in cultured and lipopolysaccharide-stimulated monocytes from peripheral blood from HA and AA.

Results: Among the three groups, there were significant differences in the methylation ratios at 7 CpGs in IL-12p35 gene promoter region. Contrary to our expectations, the methylation ratios were significantly lower in AA and NI than in HA. The IL-12p35 mRNA levels in HA and AA did not differ significantly.

Conclusion: These results suggest that hypomethylation of the promoter region may be involved in the development of allergic diseases.

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The membrane-spanning 4-domains, subfamily A, member 2 (MS4A2) +6960A/G genetic polymorphism and the risk of allergic disorders

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Background: The atopic triad of asthma, allergic rhinitis, and atopic dermatitis are multifactorial diseases with a considerable genetic component. Several genes, through association and linkage analyses, have been found to serve as susceptibility factor of atopy. A polymorphism in the membrane-spanning 4-domains, subfamily A, member 2 (*MS4A2* or FCER1B) has been shown to be associated with increased IgE levels. The role of *MS4A2* +6960A/G (E237G) polymorphism in modulating atopic disorders in the Filipino population was evaluated in this study.

Methods: One-hundred two pairs of allergic individuals and controls were phenotyped for total serum IgE level using enzyme-linked immunosorbent assay (ELISA). Atopic status was defined by serum IgE concentration >100 IU/mL. DNA was extracted and genotyped for the *MS4A2* +6960A/G polymorphism by amplification refractory mutation system-polymerase chain reaction (ARMS-PCR). Allele A was identified after PCR by a 280 bp band on 4% agarose gel and G allele by a 238 bp band.

Results: The study population was in Hardy-Weinberg Equilibrium ($\times^2$ = 0.007521, df = 1, $p < 0.05$). There was no significant difference in the total serum IgE levels between subjects with AA, AG, and GG genotypes ($p>0.05$). Likewise, allele frequencies of +6960A/G polymorphism did not differ statistically between cases and controls ($\times^2$ = 2.624, df = 1, $p >0.05$). Frequency of the G allele was 0.103 in cases and 0.157 in controls (OR = 0.617, 95% CI = 0.342 – 1.111).

Conclusion: The lack of association between *MS4A2* +6960A/G polymorphism and total serum IgE levels among allergic individuals in the Filipino study population indicates that the single-nucleotide polymorphism may not directly be involved in the development of allergic disorders.

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Alpha-1-antitrypsin genotype and allergy

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Background: Alpha-1-antitrypsin deficiency (alpha-1-AT) is usually correlated to COPD or to hepatic disease. However alpha-1-AT deficiency can be a risk factor for allergy, co-existing or interacting with asthma or other allergic diseases.

Methods: 2 patients with confirmed alpha-1-AT deficiency and allergic diseases and complete identification of genotype have been studied. Genetic studies have been done in the Genetics section of J. Chaves Clinical Pathology Clinic.

Results: Case 1: C.J.J. 62 male heavy smoker during 30 years, complaints of cough, sputum from 7 years, more than 3 months each year. No diagnosis. Previously treated with fluticasone salmeterol + tiotropium objective examination respiratory sounds diminished. PFR 300 (normal 550±100). CT. Centro lobular emphysema mainly in superior lobes. Hyperinsuflation. Electrophoresis decrease of alpha-1 fraction (0,6% of 6,9) alpha-1 antitrypsin 64,5. Genetic study genotype Pi SZ mutations GLU 342 lys and GLU 264 val. Case 2: C.M.C. 40, female, non-smoker, story of rhinitis and urticaria without lower respiratory symptoms. Objective examination normal. On electrophoresis alpha-1 fraction was 0,6% of 7.45. Alpha-1 antitrypsin assay 25-Genotype Pi ZZ mutation GLU 342 lys. Thyroid auto antibodies increased TG 72,3 TPO, 1.673.
Conclusion: Alpha-1-AT Z alleles are more frequent in northern Europe and in Iberian Peninsula. The S variant gives alpha-1 antitrypsin levels of 60% and Z variant 10% of normal (M allele). Therefore a MS heterozygote must have levels of alpha-1-AT around 40% (30% from S allele and 10% from Z allele) and a homozygotic ZZ had levels of 10%. In our cases the patient with the slightest form of alpha-1-AT deficiency has respiratory symptoms and an emphysematous pattern in CT. The theoretical more severe form ZZ homozygotic presented only rhinitis and urticaria without any lower respiratory complaints. That discrepancy could be explained by the non-smoking habits of the homozygotic patient in contrast with a long history of smoking habits of the patient SZ. In patients even without lower respiratory complaints but presenting less than 1% of alpha-1 fraction in electrophoresis assay of alpha-1-antitrypsin must be done and in cases of low values a genetic study completed.

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Changes of β-casomorphins content in human milk during lactation

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Background: Milk is the best, complete food important for development and nourishment of neonate. Except nutrients milk contains biologically active opioid peptides derived from β-casein named β-casomorphins which can exert effects in neonatal gastrointestinal tract as wall as in all organism. It has been suggested that opioid peptides may play the role in regulation of infants' gastrointestinal functions and development and be involved in the process of immune maturation. The content of β-casomorphins in human milk during maturation phases has not been studied so far. Therefore, the aim of this study was to determine the content of β-casomorphin-5 (βC5) and -7 (βC7) in human milk in different phases of lactation.
Methods: There have been chosen thirty healthy newborns and healthy mothers who plan to breast feed. The milk samples were collected every two months starting from delivery. The peptides were extracted according to Halwarker and Elliot method, purified by SPE with the use of C18 columns and identified using HPLC.
Results: A significantly highest β-casomorphin concentration was found in colostrums βC5 = 5,03 ± 1.02 μg/ml; βC7 = 3.10 ± 0.89μg/ml) than in mature milk. The concentration of β-casomorphin in milk collected in the second month of lactation βC5 = 0,98 ± 0,35μg/ml; βC7 = 0.39 ± 0.07μg/ml) was similar to the level obtained from the fourth month of lactation (βC5 = 0,58 ± 0,21 μg/ml; βC7 = 0.33 ± 0,13μg/ml). The level of β-casomorphin-5 in individual phase of lactation was not significantly different than the level of β-casomorphin-7.
Conclusion: β-Casomorphins content in human milk is changing with the period of lactation. The level of opioid peptides may depend on the function of these peptides in neonate?s organism and change with the maturation process.

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The concenttation of β-casomorphines 5 and 7 in women with history of allergy and food hypersensitivity in their babies

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Background: Breast milk provides the optimal nutritional supply which is critical for the postnatal adaptation of newborns. Although human milk is considered a hypoallergenic food, the proportions of breast-fed children demonstrating symptoms of atopic dermatitis is increasing. The allergy preventing effect of breast - feeding remains controversial and there are no consistent data regarding the degree to which breastfeeding prevents, reduces, delays or increases the development of allergic disease. A number of studies reports suggested the differences between allergic and non-allergic mothers in relation to milk concentration of immunological agents involved in allergic reactions. β-casomorphine seems to be a milk ingredient associated with pseudo-allergic reactions.
Methods: Samples of human milk were collected from 12 volunteer breast-feeding women with a history of allergy and clinical manifestation of allergy to bovine milk proteins and from 30 healthy women. Milk samples were collected in two phases of lactation - first sample - colostrums, the second - one month after delivery. In the allergic group 9 of 12 women had asthma, 4 were diagnosed with allergy to bovine milk proteins; one women had both diseases. All infants mothers of allergic group have presented skin allergic symptoms i.e. atopic dermatitis. The peptides were extracted according to Halwarker and Elliot method, purified by SPE with the use of C18 columns and identified using HPLC.
Results: β-casomorphins concentration in colostrums in the allergic group was roughly similar (βC5 = 1,58 ± 0,40μg/ml; βC7 = 1,67 ± 0.42μg/ml) and there was no statistically significant difference in β-casomorphins content between phases of lactation for both investigated peptides (mature milk: βC5 = 1,63 ± 0,64μg/ml; βC7 = 1,26 ± 0.64μg/ml). Results in the control group were more diversified - the content of β-casomorphins ? 5 and -7 in colostrums (βC5 = 5,03 ± 1,02μg/ml; βC7 = 3,10 ± 0.89μg/ml) was significantly higher than in mature milk (βC5 = 0,98 ± 0,35μg/ml; βC7 = 0,39 ± 0,07μg/ml): $p < 0,001$ and $p < 0,05$ respectively).
Conclusion: The content of both investigated β-casomorhins: 5 and 7 in healthy breast-feeding women is decreasing during lactation but remains roughly constant in the group of mothers with allergy symptoms.

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The use of a (α1-3)-Fuc-core-modified N-glycans-containing glycoepitope from a seed glycoprotein for a novel approach to the diagnosis and therapy of vespids and honey bee-allergic patients

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Background: Mucuna pruriens seeds are an oral prophylactic for snake bite in humans. We demonstrated that GpMuc, a 29 kDa glycoprotein found in these seeds, is the immunogen of the antibody that cross-reacts with snake venom proteins. The immunogenic properties are postulated to

reside in its core (β1-2)-linked xylose- and core α(1–3)-linked fucose-modified N-glycan chains. To verify this hypothesis we investigated fragmented gpMuc for immunoreactivity. Since the same core modified N-glycans in honeybee and vespid venoms have often been considered immunogens responsible for venom allergies, we also investigated cross reactivity between gpMuc and Hymenoptera venom proteins.

Methods: 1 mg of gp-Muc was trypsin digested. The digested sample was fractionated by reverse-phase HPLC and an aliquot of each fraction was analyzed for immunoreactivity to anti-gpMuc IgG by ELISA and dot blot tests, and the positive glycopeptide analyzed by Maldi TOF MS.

Serum samples from patients with IgE positive to Hymenopera venoms were investigated by immunoblot and/or ELISA for IgE and IgG against the venom proteins, gpMuc and the corresponding glycopeptide. Anti-gpMuc IgG were analyzed for reactivity against the proteins of venoms.

Results: The anti-gpMuc epitope was found to be a 1760 Da glycopeptide characterized by N-glycans containing fucose. Anti gp-Muc IgG only cross reacted with a protein having an apparent MW of 29 kDa when the venom proteins of Vespula polistes, Vespa cabro, Vespula germanica were analyzed and with a protein of 20 kDa in the protein fraction of Apis mellifera venom. Twelve of eighteen patients with IgE positive to one or more Hymenoptera venoms showed IgE and IgG positive to the 29 kDa venom protein of vespids and/or to the 20 kDa protein of Apis venom and to the gpMuc and corresponding glycopeptide.

Conclusion: Anti-gpMuc glycoepitope is included in a 1760 Da peptide and the glycoepitope is shared by a similar venom protein in vespids and by a different one in Apis. Second finding: IgE and also IgG cross reaction between vespid and Apis venoms is restricted to one protein. The third finding was the existence of IgE and IgG cross reaction between the cross reacting protein of Hymenoptera venoms and gpMuc, specifically with the peptide containing the glycoepitope. This raises the possibility of using the gpMuc glycopeptide in place of whole venoms for the diagnosis and immunotherapy of Hymenoptera allergies, at considerable saving and with reduced risk.

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Insect sting allergies of emergency department visits in Al-ain Hospital, United Arab Emirates

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Background: Insect sting is a common public health hazard in the United Arab Emirates. However, nothing is known about the demographic, clinical manifestation and management of insect sting allergic patients.

Objective: To assess the clinical manifestation of insect sting allergies and its management in Al-Ain hospital.

Methods: In this retrospective observational study, we analyzed emergency department (ED) records of insect stung patients in Al-Ain hospital. A total of 783 patients (Mean age ± SD; 29.2 ± 12.6 yrs) were seen at the emergency unit of Al-Ain Hospital.

Results: Of the total sample, 425 (54.3%) were female. Only 39 (5%) patients of the total were UAE nationals and 744 (95%) were heterogeneous expatriate workers. The site of the sting of 130 (16.6 %) patients was on the leg, 70 (8.9 %) on the arm, 21 (2.7%) on the head, 21 patients (2.7 %) on the trunk, and 12 patients (1.5 %) on the neck. However, a significant majority of the patients 528 (67.4 %) there was no available data with regard to the site of sting. In this study 41% of the subjects had local reactions, 40.4% had urticaria, 4.3% had angioedema, 47.5% had dyspnoea or wheezing, while only 3 patients were presented with unconsciousness. According to Muller classification, 17.8% were of class I severity, 4.2% class II, 36.8% class III and 2.6 % class IV. At ED 68.2% of the patients received antihistamine, 90.5% systemic antihistamine, 53.3 % systemic corticosteroids and only 1% received epinephrine treatment.

Conclusion: Insect sting is common particularly among the expatriate community, often under documented, and of sever presentation. Since most of the insect sting was on the legs and arms, culturally covered clothing among the UAE nationals may have had significant protection impact. Increasing awareness, appropriate diagnosis, and adequate management of insect sting allergies is required.

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Jack jumper ant sting anaphylaxis in South Australia

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Background: In south eastern Australia the Jack Jumper Ant (JJA) is a dominant cause of sting anaphylaxis. We have described the epidemiology of JJA sting anaphylaxis on the island of Tasmania in a prospective study; approximately 12% of allergic subjects were stung each year, 70% of these experiencing further systemic reactions. Age over 35 years was a risk factor for severe reactions.

Aim: To examine the natural history of JJA sting allergy in South Australia (SA), 1000 km NW of Tasmania.

Methods: Subjects with a history of systemic reactions to ant stings were recruited through the Australian Ant Venom Allergy Study (AAVAS). Demographic details, blood samples for allergy testing, ant specimens and details of sting reactions were obtained. Subjects were followed prospectively for further sting reactions.

Results: In 116 subjects with a history of systemic reaction to JJA sting there have been 15 further JJA sting events in 12 subjects during a mean follow-up period of 10.8 months; theses stings led to a total of 12 systemic reactions in 9 subjects. Only 4 of the 12 who were stung were less than 35 years of age, all reporting a lessened (2) or local only (2) reaction. Four of 8 older subjects experienced reactions similar to their worst previous sting event.

Conclusion: Despite wide geographic separation the frequency of re-sting (10% of subjects per annum) the proportion of systemic reactions (~75% of subjects, 80% of sting events) and the tendency of those aged >35 to have further serious reactions bears a striking similarity to the Tasmanian data. Older subjects allergic to venom of this aggressive ant have a high risk of further systemic reaction to future stings.

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Reactions to hymenoptera sting in adult patients: experience in a clinical allergy/immunology service in Monterrey Mexico

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Introduction: The most commonly implicated insects in stings to humans are the honeybee, yellow jacket wasp and paper wasp. Stinging ants include the red and black fire ants. In Mexico, there are no data on the degree to which the general population is affected.

Methods: A retrospective study, in which a questionnaire was given to all the patients who came to our clinic for the first time for different allergic symptoms, from January 2001 to December 2001 the questionnaire, asks about personal history of hymenoptera sting. We found 72 patients with positive history. Results: The distribution by sex was male (26) 36.1%, female (46) 63.9%. The distribution by insect's sting was; Ant (58) 80.6%, honeybee (22) 30.6%, yellow jacket wasp (13) 18.15%, paper Wasp (2) 2.8%, none identified (1) 1.4%. The average age was 15 a 59 years SD (32.97 + 1.25) (63) 87.5% did not have work risk, number of sting trough life (51) 70.8% 1 a 3 stings episodes, (21) 15.1% 2 a 5, (19) 26.4% had an interval of 1–6 months between sting episodes, (16) 22.2 % had a 3–5 years interval between stings. (36) 50 % had itch or local pain, (6) 8.3%, had intense itching, (2) 2.8 % urticaria or angioedema (2) 2.8 % cardiovascular symptoms, (2) 2.8 % referred to have late reactions, (6) 8.3% had large local reactions. 69 (95.8%) did not seek for medical attention, 4 (5.6 %) New the severity of the problem, 3 (4.2 %) New what to do, (27) 37.7 % had allergic rhinitis and or allergic conjunctivitis, (16) 22.2% had urticaria and or angioedema, (15) (18.9%) had contact dermatitis, (7) 9.7% had asthma, (4) 5.6% have had at least one adverse reaction to drugs, (3) 4.2% had atopic dermatitis, (1) 1.4% had food allergy, (1) 1.4% present with anaphylaxis, (25) 34.7% had a family history of allergy.

Conclusion: About one third of those experiencing allergic sting reactions are atopic. Most insect stings are associated with local reactions, including pain, swelling and redness, which are self-limiting. However, systemic reactions can lead to potentially life-threatening manifestations in 0.4% to 0.8% of children and 3% of adults. Large local reactors have a 5%–10% risk of subsequently developing a systemic reaction if re-stung.

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Development of a murine model of cockroach allergy

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Background: An animal model resembling the human clinical manifestations of allergy is needed for various aspects of allergy research, *e.g.* for investigating mechanisms and pathway of immunopathology, measuring allergenicity of a protein, testing drug and vaccine therapeutic efficacy.

Objective: To develop a murine model of cockroach allergy.

Materials and Methods: Individual BALB/c mice were intraperitoneally (i.p.) injected with three doses, *i.e.* on days 0, 7, and 14, of alum adjuvanted-crude extract of American cockroach (CR), *Periplaneta americana*, which is the predominant CR species in Thailand. The animals were subsequently aerosolically challenged on days 21 and 22 with 10 ml crude CR extract (1.0% w/v in PBS), using a home-made air-pressure nembulizer for mice. The mice were bled and sacrificed on day 23. Their serum samples, broncho-alveolar lavage fluids (BALF) and lungs were collected. Control mice received only alum i.p. and aerosolic PBS via the nembulizer.

Results: All CR extract treated mice showed features of allergy. Their BALF contained inflammatory cells that out-numbered those of the non-allergic counterparts as determined by differential cell counts after cyto-spinning and hematoxylin-eosin staining. Most of the cells were eosinophils. Eosinophil infiltration was also pronounced in the lung tissues of the CR extract treated mice and was negligible in the tissue of the controls. Levels of serum IgE specific to the American CR extract rose significantly above those of the control levels.

Conclusion: A murine cockroach allergy model with features resembling the human allergic manifestations was successfully developed. The model can be used for study of various attributes of CR allergy, *e.g.* allergenicity of a protein of interest, immunopathology, and drug and vaccine therapeutic efficacy.

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Crude Forcipomyia taiwana (biting midge) extracts stimulate PBMC proliferation, IFN-α, IL-6 and IL-10 secretion from midge-allergic subjects

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Background: *Forcipomyia taiwana (F. taiwana)* is a tiny (1–1.5 mm) blood-sucking midge that is widely distributed in urban and suburban Taiwan as well as in southern China. *F. taiwana* attacks in groups to exposed parts of the human body during the day, causing intense pruritis and swelling in sensitive individuals. There are two types of reactions after midge bite: 1) immediate reaction: large local swelling develops within one hour of bites 2) delayed reactions: intense itching papules and vesicles occur 6–24 hours after bites. The delayed lesions may turn centrally necrotic and last for weeks or even months. We have previously reported that about 59.1% of the exposed subjects develop hypersensitivity reactions to F. taiwana bites. Among which, about 14% developed immediate reaction only, 43% developed an immediate reaction followed by delayed reactions and 43% developed solely delayed reaction. Our previous study suggested that the immediate large local reaction is IgE-mediated and the 24-, 35/36- and 64-kDa proteins are major IgE-binding allergens. However, from our previous studies, we also found that delay reactions to *F. taiwana* bites may not react through the IgE pathway. Delay itching skin papules or vesicles usually last for weeks or even months that causing an even greater impact on patient's quality of life than the rapidly resolved immediate reaction.Except some limited studies focused on the immunologic mechanisms to mosquito bites, there has been no report in the literature regarding the immunologic mechanisms to biting midges.

Materials and Methods: Peripheral mononuclear cells (PBMC) from midge-allergic patients with delayed reactions were cultured with media only, Con A or *F. taiwana* crude extract at different time points. Cell proliferation was measured by MTT assay. Change of cytokines in culture supernatants was measured using fluorescent bead immunoassay.

Results: In the present study, we demonstrate that peripheral mononuclear cells (PBMC) from delayed typed midge-allergic subjects proliferate and secret more interferon-α (IFN-α), interleukin-6 (IL-6) and interleukin-10 (IL-10) in response to crude *F. taiwana* extract. IL-4 and IL-5 were not detected in the culture supernatants.

Conclusion: The delayed reactions of *F. taiwana* bites may be mediated by a more Th1 type cells. The role of IL-10 in midge-bite allergy requires more investigation.

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Local allergic reaction on bone wax in a patient with bee venom allergy

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Introduction: A 67 year-old farmer was transferred to our clinic after an outpatient operation on his right middle ear (stapes reconstruction) that showed an intraoperative strong bleeding. During the exploration of the ear in our operation room a damaged raised bulb of the jugula vein could be detected. The lesion was closed by conventional bone wax, fibrin glue and perichondrium.

Follow Up: Ten days after the removal of the endaural tamponade the patient complained of chronical otorrhoea of the operated ear that showed resistance to any treatment. The examination showed a distinct inflammation of the external and middle right ear with a clear secretion. During the operative revision of the situs an extensively grown granulation tissue around last bits of the applied bone wax was seen. After its removal the defect was closed by cartilage of the tragus.

Diagnostic Investigation: After the second operation the patient mentioned a previously known bee venom allergy he had not mentioned in the first examination. Recurrent anaphylactoid reactions had happened on bee stings. We decided to get into contact with the producer of the bone wax. We experienced that the used common bone wax consists of 80% of bee wax and 20% of isopropylpalmitate.

The allergy diagnostic on the patient confirmed the bee venom allergy (prick test, RAST), furthermore the epicutaneous testing on the applied bone wax batch showed a positive result.

The epicutaneous testing of the single components of the bone wax (bee wax, isopropylpalmitate) was negative.

Conclusion: For the producer could not guarantee a 100% pureness of the product, we conclude that the applied charge of bone wax containing bee wax was probably contaminated with bee venom.

Bee wax represents a great part of the base of common ointment and creams. The patient never mentioned reactions on those substances. Therefore, the probabiltiy of an allergy against uncontaminated bee wax (allergic reaction type I on propolis) is very low.

In future, a detailed allergy anamnesis should be performed before using bone wax in ear or any other ent surgery. Alternatively, hypoallergenic substances should be used in patients with known bee wax allergy undergoing ent surgery.

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Carpenter bee anaphylaxis: description of two cases and identification of the responsable allergens

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Background: Carpenter Bee (Xylocopa violacea) is a solitary, fast flight, ubiquitous bee usually non aggressive and rarely stinging. Up to now, in medical literature Carpenter bee anaphylaxis has never been reported. Purpose: to describe, for the first time, Xylocopa violacea venom anaphylaxis in two patients and to determinate the relevant allergens of Xylocopa violacea venom.

Materials and Methods: Patients: we report the case history of two patients (L.M. – 26 y.o; B.S.M. – 39 y.o.) with anaphylactic systemic reaction after Xylocopa violacea sting. All the two patients were also stung by yellow jacket before and after anaphylaxis, well tolerating the stings. At the clinical visit we performed serum venom specific IgE antibodies and venom skin prick test and intradermal tests with yellow jacket, paper wasp, european hornet, honeybee and Polistes dominulus venoms. Five patients with negative allergenic history were used as negative controls. Xylocopa violacea extract: Xylocopa violacea extract was supplied by Anallergo (Florence, Italy).

SDS-PAGE and Immunoblotting: SDS-PAGE was performed with Xylocopa violacea and honeybee venom extract. The gel was blotted on a nitrocellulose membrane, cutted into strips and matched with the sera of the two patients. After incubation with anti-IgE antibodies, the allergenic bands were detected by a chemiluminescens revealing system.

Results: In the two allergic patients skin tests and serum IgE detection were positive only for yellow jacket extract. In vitro analysis did not show any IgE binding in the honeybee immunoblotting, while IgE reactivity was shown with the band at 25,2 kDa, 18,8 kDa and 6,5 kDa in the Xylocopa violacea extract. No response was found in control sera. The allergenic bands at higher molecular weight were specific of Xylocopa violacea venom, while the band at 6,5 kDa corresponds to mellitin.

Conclusion: This is the first report of two cases of anaphylaxis caused by Xylocopa violacea, a member of the Apidae family. Two allergenic bands (25,2 kDa, 18,8 kDa) are specific allergens of this venom extract, not shared with honeybee venom extract.

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Tryptase, mastocytosis and Hymenoptera venom allergy

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Background: Elevation of basal serum tryptase as a one of signs of systemic mastocytosis (SM) can be a risk factor for the reaction severity in patients allergic to Hymenoptera venom (HV). The exact pattern of the relationship between elevated basal tryptase and anaphylaxis is not completely understood. We focused on the relationship between serum basal tryptase and patient's clinical data, severity of systemic allergic reactions (SAR) and safety of venom immunotherapy.

Patients and Methods: We analyzed data from 69 consecutive patients with SAR due to HV allergy. 27 men and 45 women were included; mean age 48.26 years, most of them with sensitization to relevant venom allergen (prick tests or specific IgE positive). The IgE and tryptase level was measured by Phadia ImmunoCAP FEIA assay.

Results: We found significant differences between mean tryptase values in groups of different severity of SAR: Grade I (mean 4.77 ug/l), Grade II (5.56), Grade III (5.48) and Grade IV (12.67, p=0.035). 10 patients (14.5 %) had elevated basal tryptase (mean value 28.52 versus 5.28 ug/l, $p < 0.0001$). Patients with elevated tryptase were significantly older than those with normal value (61.3 and 46.2 years, p=0.0049). Two patients were diagnosed by systemic mastocytosis, one by non-Hodgkin lymphoma, in two patients SM was not confirmed. Venom immunotherapy was well tolerated.

Conclusion: The elevated basal serum tryptase level (not systemic mastocytosis only) can be a risk factor for more severe SAR caused by Hymenoptera venom allergy, particularly at the higher age.

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Hymenoptera venom allergy: clinical importance of tryptase values

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Background: Increased serum tryptase (ST) levels and Mastocytosis are described in patients with severe anaphylactic reactions after hymenoptera sting (from 7,3% to 10.5% depending on the study).

Aim of this study was to determine basal value of ST and its possible clinical relevance in hymenoptera venom allergy.

Methods: One hundred six patients with a history of venom anaphylaxis (79 yellow-jacket, 10 paper-wasp, 5 european hornet, 12 honey-bee) were included. The grade of allergic reactions was considered according to Müeller classification. ST levels were measured by UniCAP, Pharmacia-Uppsala, (normal range <11.4 μg/L) at baseline and in 25 patients after 1 year immunotherapy. Venom serum specific IgE levels (sIgE) were measured at baseline by CAP System, Pharmacia- Uppsala.

Results: Mean basal ST level were 3.86 (IQR 2.61–5.39). Only 4 patients (3.77) have elevated ST levels.

There were no significant difference in ST levels between HB and other Vespid patients (3.47 vs 3.91). Mean basal venom-specific IgE (sIgE) was higher in HB than in all the other venom allergic patients (19.4 vs 4.14).

In Vespid patients, but not in HB ones, higher ST levels were statistically correlated with a higher Müeller grade of reaction ($p = 0.049$), while in HB patients there is an inverse correlation between basal ST and sIgE ($p = 0.053$). No difference in ST levels was observed after 1 yr immunotherapy in respect to baseline values.

Conclusion: In our population cutaneous or systemic mastocytosis appear not to be so recurrent. The only patients with high ST levels were allergic to Vespid venoms. HB patients, despite a higher specific venom IgE levels, did not present statistically more severe reactions than Vespid patients. To note that HB patients lower ST levels correlate with higher sIgE levels. These data suggest that in HB patients severe anaphylactic reactions are due to the high sIgE level and not to a larger number of mast cells which a higher ST level would indicate.

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Hymenoptera venom allergy: type of occupation as a risk factor

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Background: Much is known about the risk factors involved in hymenoptera venom allergy (HVA) including the work occupation. On the other hand, little

is known about the quantitative weight of the different work activities. Aim of study was to evaluate the distribution of work habits among subjects with HVA.

Methods: Subjects referred to our unit for suspected HVA underwent the standard diagnostic workup. According to the modality of exposure to hymenoptera stings, they were classified: i) at high risk (subjects working usually outdoors or in places were hymenoptera live): farmers, gardeners, firemen, truck drivers, masons, beekeepers, dustmen; ii) occasionally at high risk (working sometimes outdoors): plumbers, electricians, railswaymen, at low risk (working indoors) students, housewives, clerks, professionals. The distribution of occupations in the general population was provided by the Central Office for the Work Insurance of Regione Veneto.

Results: Three hundred and 46 subjects (238 male, mean age 44 ± 15 yrs) seen at our unit between 2000 and 2007 were diagnosed as having HVA. All had a clinical history of systemic reactions to stings (20% bees, 17% polistes, 63% wasps), of various grades of severity (56% grade III-IV). Concerning exposure, 23,4% of patients were at high occupational risk (43 farmers, 15 masons, 14 truck-drivers, 4 gardeners, 3 beekeepers, 2 dustman), 8,38 % had occasionally high risk (3 warehousemen, 3 soldiers, 9 factory workers, 1 plumber, 5 electicians, 2 railwaymen 1 agronomist, 5 engineers) and 68,2 % were at low risk. In the general population, wokers not at risk were 91.3%, at occasional risk 0.9%, and at high risk 7,8%. Among the patients at high risk or occasionally high risk (110 patients), 71% were stung only during their work activity. Beekeepers, considered typically at high risk, in our database were less represented than, for instance, masons and truckdrivers. Conclusions. The risk of having HVA is higher than expected in some categories. Also taking into account the limit of such a survey, where the data of the general population may be imprecise, it is clear that some occupations are much more frequent in patients with HVA. Thus, for some categories (farmers, truckdrivers) allergy to hymenoptera venom could be considered an occupational disease.

Keywords: Hymenoptera venom allergy, work occupation, risk.

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Results of a modified ultrarush immunotherapy in hymenoptera venom allergy

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Background: Patients allergic to hymenoptera venom may develop local or generalized sting reaction or they may show systemic reactions of differing severity. If venom immunotherapy (VIT) is indicated, increase doses are usually given during hospitalisation, because severe adverse reactions may occure. There is a wide range of rush- and ultrarush protocols known. We have tested a 3-day ultrarush-VIT protocol according to its tolerance.

Methods: 20 patients were treated with VIT, 4 of them received bee-VIT, 16 vasp-VIT. We administered 15 injections over 3 days. Incidence and intensity of adverse reactions were evaluated. Besides the coherence between cardiovascular diseases (especially hypertension), severity of systemical reaction to the insect sting challenge, specific IgE-level and skin prick test and the intensity of adverse reactions during VIT were determined.

Results: In total 3 of 20 patients showed adverse systemic reactions, all of them got a vasp-VIT. 2 of these patients had a third grade anaphylactic reaction (according to Ring and Messmer) due to a hymenopteron sting in case history, 1 patient reported about a first grade anaphylactic reaction. Local reactions to the injections ranged from mild (<5cm) to moderately severe (up to 10cm). There was no obvious correlation between intensity of local reaction during VIT and degree of anaphylaxis in case history. 2 of the mentioned 3 patients showed positive reactions in skin prick test at 1µg-dose, 1 patient just reacted at 100µg-dose. The specific IgE-level also showed no correlation to local or systemic reactions.

Conclusion: The 3-day ultrarush protocol was well tolerated by our patients. We noticed 3 systemic reactions. This is in accordance with the data of studies in literature. Due to the small number of current participants we can find no connection between the incidende of systemic reactions during VIT and the other facts examined. The only noticeable fact was that 2 of 3 patients with systemic reaction during treatment reported about third-degree anaphylactic reaction by first sting challenge. Local reactions to the injections showed moderate intensity, there were no skin effects more than 10cm.

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Leishmanial antigen detecting by dot immunoblot assay in phlebotomus papatasi, in Iran

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Introduction: Leishmaniosis is a polymorphic disease of the skin and viscera caused by an intracellular Protozoan. Zoonotic Cutaneous Leishmaniosis (ZCL) is a major health problem in rural areas of Iran.

Methods: A simple and highly reproducible dot-immunoblot assay was developed to detect leishmanial antigen in Phlebotomus papatasi that were naturally infected with Leishmania major. The test was sensitive to as little as 10 ng of antigenic protein and also appeared to be specific, in that it gave a positive result with some P. papatasi (the primary vector of L. major in Iran) and L. major but not with P. sergenti or other pathogens. When used to investigate a large number of sandflies collected from areas of the Iran where cutaneous leishmaniasis is endemic

Discussion: The assay appeared sufficiently sensitive and specific to detect the naturally infected insects. The simplicity, reproducibility, high sensitivity and high specificity of the assay should make it useful for field studies.

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Cockroach sensitization in patients with allergic airways disease in reunion island

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Background: The objective of this study was to evaluate the prevalence of skin Prick sensitivity and RAST (UniCap-Pharmacia)to *Blattela germanica, Blatta orientalis,Periplaneta americana*in patients with rhinitis and / or asthma living in Reunion Island (Indian Ocean).

Methods: Patients were 51 male, 53 female, aged 5 to 65 years (mean age of 29.6 years) were evaluated in one private clinic and in one public hospital: 18.2% had asthma, 32.5% rhinitis and 49.3% rhinitis and asthma. They were considered sensitized if the prick test induced a papula with at least one half the area of papula induced by histamine, or , if specific IgE was superior to 0,70 KU A/L. Skin tests were conducted with extracts of *B. germanica, B. orientalis, P. americana* and a battery of common aeroallergens (Allerbio, ALK Abello, France).

Results: A positive skin test was recorded in 98 patients (94.5%), 87% of which were positive to mites, 15% to pollens, 9% to moulds, 4.8% to epithelia and 51.9% to at least one of the cockroach species tested; 24.9% of these patients was sensitized to all 3 cockroaches and 27% were sensitized to just one species.The prevalence of sensitization to the respective species was: 41.8% to *B. germanica*, 33.9% to *B. orientalis*, 39.8% to *P. americana*. Mean values of specific IgE for positive RAST were respectively: 34,9 ± 36,4;25,1 ± 31,0; 24,6 ± 32,6. In addition, a significant correlation between results of skin test to cockroach and mite was observed (p.0004).

Conclusion: The authors concluded that sensitization to cockroach is frequent in Reunion Island. Our results suggest the recommendation that cockroach extracts should be routinely used for evaluation of patients with asthma and rhinitis,or both in tropical countries.

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Effective treatment of mosquito bites with rupatadine; an experimental study

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Background: People frequently experience whealing and delayed papules from mosquito bites. Whealing is mediated by antisaliva IgE antibodies and histamine.

Rupatadine is a new antihistamine and PAF antagonist effective in allergic rhinitis and urticaria but the effect on mosquito-bite allergy is not known.

Methods: A double-blind, placebo-controlled, cross-over study was performed with rupatadine 10 mg and matched placebo in 30 mosquito-bite-sensitive adults. The mean age was 37 years and the subjects had suffered from harmful mosquito bites for a mean of 15 years. Either rupatadine or placebo was taken at 8 am for 4 days, followed by a 5 day wash out period and then alternative treatment was given for 4 days. On day 3, in the both drug periods the subjects received two *Aedes aegypti* mosquito-bites on the forearm. The size of 15 min bite lesions and intensity of pruritus (VAS) were measured.

Results: 27 subjects could be analysed for the efficacy. Rupatadine decreased the size of 15 min wheals by 43% compared to placebo. This effect was significant ($P = 0.0065$) whereas the effect on accompanying pruritus did not reach statistical significance although there was a favourable trend ($P = 0.0621$).

Conclusion: The present placebo-controlled study in mosquito-bite-sensitive adults shows that prophylactically given rupatadine 10 mg is an effective treatment for the mosquito-bite whealing.

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Staphylococcus species as potential virulence factors in atopic dermatitis

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Background: Atopic dermatitis represents a chronic inflammatory skin disease which results from complex interactions between genetic and environmental mechanisms. Referring to this the colonization and infection with staphylococci is a serious issue in skin disorders especially atopic dermatitis (AD). Human skin epithelium provides a mechanical barrier to invading bacteria and also participates in innate immune defence by producing cationic antimicrobial peptides, e.g. β-defensins (hBD) and the cathelicidin LL-37. The aim of our study was to determine the potential virulence of staphylococci from AD patients in bacterial skin infections and to examine the innate immune response to infection through expression of antimicrobial peptides.

Methods: Identification of the staphylococci was performed by using T-RFLP analysis of the gap gene. Pathogenicity profiles of Staphylococcus spp. isolated from AD patients were determined by screening genes for staphylococcal enterotoxins (sea-see, seg-sej), toxic shock syndrome toxin 1 (tst) and Panton-Valentine leukocidin (luk-PV) by PCR. A model of bacterial skin infection was used to test the pathophysiology of these isolates in a human keratinocyte cell line (HaCaT), including the induction of hBD-1, hBD-2, hBD-3, LL-37, IL-6 and IL-8 (real time PCR, ELISA) and cellular invasion (fluorescence staining, confocal laser scanning microscopy).

Results: Toxin genes with various pathogenicity profiles were studied for S. aureus. No superantigenic exotoxins by coagulase-negative staphylococci were detected. Exposure of keratinocytes to Staphylococcus spp. triggers the production of cytokines IL-6 and IL-8 and of β-defensins (hBD-1 mRNA (2-to 8-fold), hBD-2 mRNA (2-to 270-fold), hBD-3 mRNA (2-to 6-fold)) and the cathelicidin LL-37 (2-to 6-fold), whereas the levels of this activity were different for all clinical isolates. Adherence and invasion to keratinocytes was shown for clinical isolates of S. schleiferi and S. aureus, but not for S. hominis, S. epidermidis and S. capitis.

Conclusion: Staphylococci from AD patients showed various pathogenicity profiles, invaded keratinocytes, the production of cytokines and antimicrobial peptides. Thus, they are potential pathogens in skin infections, especially atopic dermatitis.

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Immunological study in patients of Pemphigus Vulgaris

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Background: Pemphigus Vulgaris (PV) is an autoimmune, intraepithelial, blistering disease affecting the skin and mucous membranes. It is mediated by circulating autoantibodies (IgG) directed against keratinocytes cell surfaces and potentially life-threatening. PV antibody binds to the keratinocyte cell surface molecules Desmoglein-1 and Desmoglein-3 (Dsg-1 and Dsg-3). In India the incidence of PV is 86% with the mean age 30–40 years while in Western countries is 40% only with the mean age 50–60 years.

Methods: So it is very important to identify the implicated antigens in PV patients and diagnosis the pattern of expression of Dsg1 and Dsg3 in patients suffering from bullous diseases. The levels of cytokines-TH1 and TH2 was also studied for the involvement of T and B cells as inflammatory.

Results: It has been seen that the mucocutaneous type of PV is more prevalent in Indian patients as two types of band pattern are observed during immunoblotting with patient sera against 160kD and 130kD antigens extracted from normal human patients. It was also found that cytokine profiling of the patient sera that the level of TH2 like cytokines for example IL-4 and IL-10 were significantly high during the active stages of PV whereas, no relative difference was seen for TH1 like cytokine for example IFN-g. Cytokine profiling for confirmed PV cases from sera showed direct evidences for the involvement of auto reactive T cells responses to Dsg-3, which may be critical for pathogenesis of PV. Among its other biological activities IL-10 also induces B-lymphocytes to proliferate and differentiate to produce antibodies, which may play a important role in production of auto antibodies for PV.

Conclusion: This study indicates that PV is an antibody-mediated disease with increase in TH2 cytokines and auto reactive T cells from PV patients produced TH2 cytokine preferentially in active stages of PV disease. This study will help to elucidate the cellular immune abnormalities leading to production of IgG autoantibodies in PV patients.

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Successful treatment in pulmonary alveolar proteinosis with the combination of intravenous immunoglobulin and pulse methylprednisolone: a case report

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Background: Pulmonary alveolar proteinosis (PAP) is a rare disease in children, characterized by intra-alveolar accumulation of lipoproteinaceous material

that positively stained with periodic acid-Schiff (PAS). The susceptibility to pulmonary infection is a salient feature of the disease which could be partly explained by defective alveolar macrophage function.

Methods: We report a case of PAP and chronic interstitial pneumonitis in a girl who presented with chronic respiratory distress since the age of 5 months. Her clinical condition progressively worsened with chest radiograph showing diffuse hazy opacities of both lungs. High resolution computerized tomography (HRCT) of the chest demonstrated crazy-paving pattern. Definite diagnosis of PAP was made by open lung biopsy showing marked interstitial lung inflammation with alveolar proteinosis pattern.

Results: Bronchoalveolar lavage were attempted twice without any clinical improvement. The respiratory clinical collapse with continued recurrent pneumonia was observed. After pathological diagnosis, a short tried of G-CSF was attempted again to no avail. Low dose methylprednisolone and hydroxychloroquine was then initiated without satisfactory improvement. It was decided to start intravenous immunoglobulin (IVIG) (500 mg/kg, every 4 weeks) with pulse methylprednisolone (30 mg/kg, every 4 weeks) to which she gradually responded. She no longer required oxygen supplement after 21 months of treatment. Currently she is asymptomatic at age 4 years and continues to receive IVIG and pulse methylprednisolone every 4 weeks.

Conclusion: Aside from therapeutic lung lavage and GM-CSF therapy, IVIG and pulse methylprednisolone may have potential role in successful treatment of PAP.

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Cytokine profile of children allergic diseases

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Inflammation process is participated by a cascade of cytokines, which predetermines development, direction and power of immune response and regulates cellular interaction. The character of changes in cytokine profile may be a distinguishing sign of a pathologic process.

The research objective was to investigate specific features of cytokine spectrum of nasal secretion, saliva and blood serum in the process of children allergic diseases development (allergic rhinitis, atopic dermatitis, bronchial asthma). IL-4, IL-13, IL-8, IFNγ, TNFα cytokine content, I IFNγ/IL-4 cytokine regulatory index and total IgE level were monitored within a year at different stages of the disease. Cytokine level in biological liquids was determined by immuno-enzymatic method, sandwich variant, with R and D Diagnostics Inc. (USA) reagents. Researchers examined 22 children in the age from 1 to 16 with allergic diseases, control group was made of 30 health children of the same age.

Control group examination results: serum cytokines IL-4 = 8,5 ± 1,1 pg/ml, TNFα = 12,1 ± 2,2 pg/ml, IL-13 = 8,01 ± 1,6 pg/ml, IFNγ = 50,6 ± 4,2 pg/ml, I IFNγ/IL-4 = 5,2 ± 1,1; saliva cytokines IL-4 = 0,3 ± 0,1 pg/ml, TNFα = 1,47 ± 0,4 pg/ml, IL-13 = 2,25 ± 0,4 pg/ml, IFNγ = 65,2 ± 3,4 pg/ml, I IFNγ/IL-4 = 80,5 ± 5,4 respectively; nasal secretion cytokines IL-4 = 1,88 ± 0,64 pg/ml, TNFα = 6,87 ± 1,08 pg/ml, IL-13 = 6,19 ± 1,14 pg/ml, IFNγ = 156,8 ± 4,8 pg/ml, I IFNγ/IL-4 = 75,4 ± 4,8.

High IL-4 and IL-13 levels at decreasing INFγ were recorded mostly in nasal secretion of children with allergic diseases in isolated allergic rhinitis (AR) cases (I IFNγ/IL-4=0,34 ± 0,06). High IL-4 content 87,62 ± 2,59 pg/ml in nasal secretion were observed in exacerbation period and in rhinitis and bronchial asthma (BA) combined cases, at the same time children with BA but without AR showed 15,04 ± 2,21 pg/ml and I IFNγ/IL-4 = 4,6 ± 1,2. In the period of AR clinic remission IFNγ content in nasal secretion was 1,1 ± 0,96 pg/ml, in exacerbation period – 9,36 ± 2,21 pg/ml. Cytokine levels observed in saliva in children dermatitis cases were as follows: IL-4 = 89,3 ± 0,64 pg/ml, IL-13 = 124,41 ± 5,9 pg/ml, IFNγ = 11,9 ± 1,7 pg/ml, blood serum IL-4 = 55,2 ± 4,2 pg/ml, IL-13 = 39,4 ± 4,9 pg/ml, IFNγ = 21,2 ± 2,0 pg/ml.

Investigation of local cytokine secretion and determination of their ration help to specify the localization and rate of allergic inflammation providing data for forecast and treatment effectiveness evaluation.

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Severe eczema associated with IgG2/G3 subclass and IgM deficiencies successfully treated with intravenous immunoglobulin

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Background: Patients with severe eczema and recurrent infections should be investigated for immunodeficiency. We report our experience in treating a patient with severe eczema, recurrent bacterial infections, chronic otitis media, IgG2/IgG3 subclass and IgM deficiency with monthly intravenous immunoglobulin (IVIG) infusion.

Methods: An 8 year-old boy was a case of long standing severe eczema with multiple food and drug allergies since 2 years of age. He had had chronic otitis media for the past 2 years. His treatment included elimination of suspected foods and drugs, topical corticosteroid, oral antihistamine and topical calcineurin inhibitor without satisfactory improvement. He was admitted several times due to bacterial skin infections with which he developed Acenitobacter septicemia on one occasion. His CBC did not show thrombocytopenia (platelet 300,000 /mm3) nor small platelets. Immunoglobulins were: IgG 1,480 mg/dl, IgG1 1,030 mg/dl, IgG2 115 mg/dl, IgG3 10.7 mg/dl, IgG4 18.3 mg/dl, IgA 142 mg/dl, IgM 18.3 mg/dl, and IgE 8,080 mg/dl. Specific IgE for egg white and cow's milk were positive. Immunophenotype revealed: CD3 692 cells/mm3, CD4 284 cells/mm3, CD8 351 cells/mm3, CD19 378 cells/mm3 and NK cell 193 cells/mm3. Lymphocyte proliferative response to PHA was below normal. The antibody response to pneumococcal vaccine increased 8.1 folds after receiving conjugated pneumococcal vaccine.

Results: IVIG infusions at 400 mg/kg were given every four weeks. The patient's skin lesions were gradually improved after the 4th dose of IVIG infusion.

Conclusion: The patient who presented with severe eczema and recurrent infections should be evaluated for immunodeficiency. The mechanism of beneficial effect of IVIG therapy in this patient was unknown but could be due to prevention of infection and its anti-inflammatory effects.

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Long-term effects of treatment on the cytokine production in scleroderma patients

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Background: Scleroderma, a debilitating acquired connective tissue disease, is characterized by fibrosis, particularly of the skin and lungs. The cytokine disbalance is one of the main manifestation of scleroderma. The treatment of this pathology is very difficult problem for therapy.

Objective: To compare the long-term effects of infusion of alprostadil with those of oral pentoxyphyllin on the production of cytokines in patients with systemic sclerosis (SSc), and evaluate their relationship with effects of the two treatments on clinical parameters.

Methods: The production of cytokines by alloactivated circulating mononucleated cells was assessed before and after one year of treatment in a subset of 50 patients enrolled in a one-year study. Twenty five patients were treated with a 10-day (100 mkg) infusion of alprostadil - synthetic analog of prostaglandin E2. Another 25 patients were treated with an oral slow-release formulation of pentoxyphyllin, 600 mg twice daily. Quantitative determinations of interleukin-1 beta (IL-1beta), interleukin-6 (IL-6),

tumor necrosis factor-alpha (TNF-alpha) in the serum of patients were performed with a ELISA.
Results: The production of IL-1beta was significantly lower in the alprostadil group than in the pentoxyphyllin group. Both the cutaneous fibrosis and the capillaroscopic patterns were better in patients treated with alprostadil. There was significant positive covariance between IL-1beta and TNF-alpha changes and the changes in both the skin score and the capillaroscopic score.
Conclusion: There are several mechanisms by which alprostadil could exert its clinical efficacy. Vasodilatation and inhibition of platelet aggregation are certainly important, but they are transient. We suggest that the long-lasting modulation of the cytokine network observed in the present study could be another potential mechanism responsible for the persistent efficacy of alprostadil despite its administration.

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Endothelial dysfunction and inflammatory markers at the chronic heart failure with hypertension

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The chronic heart failure and arterial hypertension has a lot of biochemical manifestation. The ideas of our research work were: 1) to compare high sensitive C-reactive protein (hsCRP) and soluble receptors TNF alpha (sTNF-Rs) with coronary heart disease (CHD) and the chronic heart failure (CHF); in the control group (C group); 2) to compare ultrasound markers of endothelium-dependent vasodilatation (EDV) and endothelium-independent vasodilatation (EIV) in hypertensive (HT) patients in both groups.
Patients and Methods: 105 hypertensive pts (mean age 61.3 yrs.), with CHD+ CHF were investigated. In the NYHA classes I/ II/ III/ IV were 20/31/46/8 pts. The C group was represented by 20 healthy volunteers. Ultrasound assessment of flow mediated dilation (FMD) of brachial artery was used for the measurement of EDV and the response to isosorbide dinitrate (ISDN) for the measurement of EIV. sTNF-Rs and hsCRP were examined by the ELISA method.
Results: FMD was significant diminished in hypertensive pts. In 60th sec of reactive brachial artery hyperemia the luminal size has not reached the basal parameter (HT pts mean FMD: 92.0 ± 9.9%, C group mean FMD: 109.5 ± 7.8%, $p < 0.001$). EIV was significant reduced in HT pts comparing with C group (HT pts mean EIV: 105.2 ± 18.3%, C group mean EIV: 118.6 ± 10.5%, $p = 0.01$). In HT group significant elevated levels of hsCRP and sTNF-RI were found (HT pts mean hsCRP: 8.14 ± 4.05 mg/l, C group mean hsCRP: 3.81 ± 3.02 mg/l, $p < 0.01$; HT group mean sTNF-RI: 1161.6 ± 168.1 pg/ml, C group mean sTNF-RI: 638.5 ± 37.5pg/ml, $p < 0.001$). No significant difference in sTNF-RII levels between the groups was found. Correlation between sTNF-RI and rest luminal size of brachial artery was observed ($r = 0.25$, $p = 0.05$). No other correlation between EDVa EIV and inflammatory markers was found.
Conclusion: Significant reduced endo thelium-dependent and endo thelium-independent vasodilation of brachial artery in HT pts was found. Significant elevated levels of hsCRP and sTNF-RI in these pts were measured, too. No significant correlation between ultrasound markers of endothelial dysfunction and inflammatory parameters was observed. Ultrasound assessment of endothelial dysfunction and inflammatory markers may give us an information concerning the severity and progression of disease.

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Pharmacogenetic study for predicting cyclophosphamide side effect in thai systemic lupus erythematosus

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Background: Systemic lupus erythematosus (SLE) is the serious autoimmune disease that primarily affect young women. Although cyclophosphamide improves outcome of SLE, frequent ovarian toxicity is a major side effect of this therapy. Cytochrome P450 (CYP), CYP2C19 polymorphism has been shown to significantly influence the pharmacokinetics as well as the ovarian toxicity of cyclophosphamide in Caucasian SLE patients.
Objective: To assess association between CYP2C19 polymorphism and ovarian toxicity in Thai SLE patients who have been treated with cyclophosphamide.
Patients and Methods: Female patients who were diagnosed as SLE according to ACR criteria and treated with cyclophosphamide were genotyped for CYP2C19 polymorphism. Ovarian toxicity was defined as sustained amenorrhea for more than 12 months occurring before 50 years of age or lack of menstruation for more than 4 months during treatment. Chi-square test was used for assessing association. Logistic regression was applied.
Results: Seventy-one patients were enrolled for analysis. The mean age at diagnosis and starting cyclophosphamide was 26.42 ± 8.35 and 29.27 ± 7.82 years. The mean cumulative cyclophosphamide dosage was 23.36 ± 20.81 grams. Thirty-six (59.7%) patients developed ovarian toxicity. CYP2C19*2 allele frequencies were 27.8% and 21.4% in ovarian and non-ovarian toxicity groups. Genotype frequencies in non-ovarian toxicity group were complied with Hardy-Weinberg equilibrium ($p = 0.304$). ROC analysis was applied to calibration cut-off of cyclophosphamide dosage and found that dosage of 23.75 grams yielded likelihood ratio for developing ovarian toxicity of 3.5. Logistic regression analysis was applied to assess interaction effect between cyclophosphamide dosage and CYP2C19 allele. We found that among CYP2C19*1 allele, patients received cyclophosphamide 23.75 grams or higher were 12.6 (95% CI = 3.3–46.8) times more likely to have ovarian toxicity than patients received cyclophosphamide < 23.75 grams. This risk was decreased to e 5.1 (95% CI = 1.1–22.7) times if patients had CYP2C19*2 allele.
Conclusion: Cyclophosphamide dosage of 23.75 grams or higher carries a high risk of inducing ovarian toxicity. However, the risk can be decreased if patients have CYP2C19*2 allele.

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15-epi-lipoxin is a key mediator of Th1 adverse reaction induced by aspirin and theophylline co-treatment

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Background: Aspirin-induced asthma (AIA) is a distinct clinical entity and appears to be a specific subtype of asthma. There are lots of efforts to uncover AIA pathogenesis, but adverse reaction by aspirin is still remains unclear. Aspirin is a well known non-steroidal anti-inflammatory drug. The anti-inflammatory action of aspirin generally has been attributed to direct inhibition of cyclooxygenases (COX-1 and COX-2). AIA takes part in 10% of patients with severe asthma. Theophylline has been demonstrated to have an anti-inflammatory effects as an long-term medication for severe asthma and COPD. It works as a PDE inhibitor and a nonspecific adenosine receptor antagonist.
Objective: To assess the therapeutic effects of aspirin, theophylline, and combination of two drugs in murine model of asthma.
Methods: To generate a murine model of asthma induced by LPS with OVA, 6-week-old C57BL/6 mice were sensitized intranasally with 75 ug of OVA on days 0, 1, 2, and 7 with or without LPS, and then challenged intranasally with 50 ug of OVA on days 14, 15, 21, and 22. Whenever mice were challenged, aspirin (10mg/kg), theophylline (10mg/kg), and both were injected intraperitoneally. Asthmatic phenotypes such as lung inflammation using

bronchoalveolar lavage (BAL) cellularity, cytokine production, and lung histology were evaluated on day 24.
Results: Total cell number of BAL fluids in mice treated with each of Aspirin and theophylline was significantly decreased compared with non-treated group of mice. However, inflammatory cells in BAL fluids were increased in mice treated with aspirin and theophylline combination. And similar results were represented in histology. IP-10 protein, downstream of IFN-gamma, was decreased in each aspirin and theophylline treated mice compare with each of aspirin and theophylline non-treated group of mice. However, aspirin and theophylline combination treated mice were represented increase of IP-10 level in BALF. Moreover, only aspirin and theophylline combination treated mice were revealed specially increased level of 15-epi-LXA, which generated from lipid metabolite by aspirin.
Conclusion: In murine model of asthma enhanced by LPS and OVA, each of aspirin and theophylline treatment is effective, but combinative treatment resulted in adverse reaction.

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Serum levels of immunoglobulin and complement factors in beta-thalassemia major patients in Southern Iran

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Background: Beta-thalassemia major is one of the major health problems in our country. Many studies have confirmed the fact that, these patients have an increased susceptibility to bacterial infections.
Objective: In this study, we have assessed the humoral immune system in 68 thalassemic patients by measuring their serum concentration of Immunoglobulin G (IgG), IgM, IgA, C3 and C4 in order to find out a responsible immune defect.
Methods: Sixty eight beta-thalassemia major patients were enrolled randomly from referrals to Dastgheib clinic of thalassemia. The same number of case controls with matched age and sex were selected from healthy-people without any history of recent or recurrent infections. Serum IgG, IgM, IgA, C3 and C4 levels were assessed using Single Radial Immunodiffusion (SRID).
Results: Serum levels of IgG, IgM and IgA were significantly higher ($P < 0.01$) and those of C3 and C4 were significantly lower ($P < 0.01$) in thalassemic patients than the controls. Considering the result of analytic tests, it was revealed that, thalassemia patients show much more increase in serum immunoglobulin levels as they get older. Splenectomized patients had higher serum IgG and IgA levels than non-splenectomized patients but had no difference in serum IgM, C3, and C4. Serum ferritin level had no correlation with the changes of humoral immunity; however, patients with serum ferritin level > 2500 ng/ml had higher serum IgM level.
Conclusion: These results can be due to continuous exposure to antigens, repeated infections, chronic liver disease and splenectomy but not iron overload. The only probable cause of humoral immune deficiency found in these patients is a defect in serum complement levels.
Keywords: Thalassemia Major, IgG, IgA, IgM, C3 and C4

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A rare cause of acute eosinophilic pneumonia

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Introduction: Acute eosinophilic pneumonia (AEP) is a rare disease described as follows: an acute febrile illness with severe hypoxemia, diffuse pulmonary infiltrates, an increase in bronchoalveolar lavage (BAL) eosinophils, no evidence of infection, asthma or atopic illness and complete resolution after treatment with corticosteroids. It is suggested that AEP is an acute hypersensitivity reaction to an unidentified inhaled antigen. We report a case of AEP that developed after Narghile water pipe smoking (WPS).
Case report: An 18-year-old woman was admitted with a cough and high fever for the last 4 days. She had no previous history of asthma or atopic disease. There was no history of smoking, recent traveling or using medications. During the week before her admission she smoked a narghile water pipe. The patient was in moderate general condition, dyspneic, RR 20, Sat O2- 90% in room air, T-38°C. The physical examination was unremarkable besides bilateral inspiratory rales. Chest X-ray revealed bilateral ground glass opacities. Peripheral WBC count was 14370 cell/mm^3 with 1465 eosinophils/mm^3 (10 %). IV antibiotics, oxygen support and inhaled bronchodilators were administered. Blood cultures were negative. The eosinophil count rose to 3700 eosinophils. Total IgE level was 943 IU/ml (normal range 0–100). A chest CT scan revealed bilateral patchy ground glass opacities with no lymph node enlargement. Fiberoptic bronchoscopy was performed and BAL yielded 80% eosinophils. IV corticosteroid therapy was given with remarkable improvement. The peripheral blood eosinophil count returned to normal (1.7%) in two days. The patient was discharged in good condition and 3 months later she was asymptomatic.
Discussion: Our case met the 6 diagnostic criteria of AEP: an acute illness of short duration, high fever, severe hypoxemia, bilateral infiltrates on chest radiographs, eosinophilia more than 25% on BAL, and no history of drug hypersensitivity or evidence of infection. There are reports of AEP in new cigarette smokers. Narghile tobacco smoke contains tar, nicotine, carbon monoxide and heavy metals and it is higher than in cigarette smoke. Water-pipe smokers may absorb higher concentrations of these substances due to the mode of smoking (higher frequency of puffing, depth of inhalation).

A search in PUBMED revealed no case reports of AEP that evolved after narghile water pipe smoking. In our case it was the only etiologic factor that has emerged through detailed history.

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Serum concentrations of eosinophil cationic protein and eosinophils of patients with Kimura's disease

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To clarify the role of eosinophils in the pathogenesis of Kimura's disease and the values of measuring serum levels of eosionophil cationic protein (ECP) for monitoring disease activity, a total 14 serum and 7 tissue samples from patients with Kimura's disease were studied. The concentrations of ECP and cytokines (interleukin-4 (IL-4), granulocyte-macrophage colony-stimulating factor (GM-CSF), and interleukin 5 (IL-5)) in sera from patients with Kimura's disease were measured by enzyme-linked immunosorbent assay (ELISA). The density eosinophils and the degree of activation of eosinophils in the tissue were also studied immunohistochemically. The concentration of ECP in sera from patients with Kimura's disease was significantly higher than that in the control group ($p < 0.05$). At the time of the remission, a significant decrease of ECP was observed. In interfollicular areas, most infiltrated eosinophils were positive for EG2 antibody (64.0–94.0%) and the mean percentage of EG2-positive eosinophils was 75.7%. The concentrations of IL-4, GM-CSF, and IL-5 in sera from patients with Kimura's disease were within normal ranges or below the detectable level in all sera examined. Our findings suggest that eosinophils play an important role in the pathogenesis of Kimura's disease and ECP may be used as an additional parameter of disease activity.

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The evaluation of osteocalcin and osteoprotegerin, osteogenic markers of bone remodeling in rheumatoid arthritis

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The purpose of this study is to evaluate some osteogenic markers involved in bone remodeling, such as the estimation of osteocalcin (OC) and osteoprotegerin (OPG/RANK) concentrations in the sinovial fluid as well as in the serum of rheumatoid arthritis (RA) patients. These measurements were performed in the initial stage of the disease, without using any anti-inflammatory non-steroidal therapy for at least 1 month before our study began. The concentration of OPG in the sinovial fluid represented 14.25 ± 0.75 pmol/l while in the serum the concentration was 1.69 ± 0.80 pmol/l, which was significantly increased ($p < 0.001$) compared to the values determined in the serum of healthy controls (1.19 ± 0.30 pmol/l). The increase of OC concentration evolves in the same manner in both sites of our measurements, the values being of 6.43 ± 4.80ng/ml and 19.9 ± 4.8ng/ml respectively, compared to the values determined in healthy controls, in the sinovial liquid (< 0.5ng/ml) and in the serum (3.1 ± 0.1ng/ml).

Key words: rheumatoid arthritis, osteocalcin, osteoprotegerin, bone remodeling, bone metabolism

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The effect of probiotics on immune markers of undernourished preschool Filipino children

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Background: Undernutrition in children is associated with increased susceptibility to infections and depressed effect on cell-mediated and humoral immunity. Probiotics can enhance immune responses which can lead to illness prevention or decrease duration and severity.

Objective: The study aims (1) to determine baseline immune markers (CD3,CD4, CD8, CD19, CD 20, CD56) of undernourished Filipino children aged 3–5 years old (2) to determine whether probiotics can enhance immunity by (a) changes in immune markers, (b) changes in weight and (c) decreased incidence and duration of common infections (e.g., colds, pneumonia and diarrhea).

Methodology: Undernourished children were randomized into control and experimental group. Probiotics (mixture of Lactobacillus bulgaricus, L casei subsp rhamnosus, L fermentum, L plantarum, Bifidobacteria bifidus, B breve, B infantis, B longum, and B lactis.) were given 2 times a day for eight weeks. Weekly weight, incidence and duration of illnesses were recorded. Blood for immune markers was drawn before and after the study.

Results: 29 children were enrolled, 14 in the control and 15 in experimental group. The basal levels CD3, CD4, CD8, CD19, CD 20, CD56 were normal in both groups. The control group showed an increase in CD4, CD8 levels compared to the probiotics group but both groups' levels stayed within normal limits. The probiotics group gained significant weight by 1 kg ($p < 0.003$). The incidence of illnesses (diarrhea and colds) was the same for both, but there was significant shorter duration for the probiotics group. The median was 2 and 14 days for the probiotics and control group respectively ($p < 0.001$).

Conclusion: Probiotics lead to significant weight gain. They also lead to shorter duration of infections.

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Life expectancy of patients with leukocyte adhesion deficiency in Iran

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Background: Leukocyte Adhesion Deficiency (LAD) is a rare congenital immunodeficiency with fewer than 200 affected individuals in the world which is characterized by sever recurrent infections. The purpose of this study was to describe the survival time of Iranian patients with LAD over a period of 16 years.

Methods: Twenty seven patients with definite diagnosis of LAD (based on standard criteria and laboratory tests), attending Allergy, and Immunology clinic in Children Medical Center Hospital-from 1991 to 2006 ranging in age from 3 months to 24 Years old, were studied and followed. Severity of disease was defined as a presentation of CD18 marker on the surface of leukocyte (less than 1% of normal expression). Also a questionnaire was filled for recording all necessary information.

Results: All patients (63% male, 37% female) was LAD type I. The prevalence of consanguineous marriage was 81.5%. The mean age at the time of disease onset was 0.78 ± 2.03 months and the mean age of diagnosis was 1.33 ± 0.55 months. Overall 33.3% of patients (9 patients) had severe form of disease. The overall 5 year survival rate was 67% and mortality rate was 22.2%. The mean of survival time for alive patients was 73 ± 27 months (CI:21–125). The main leading cause of death was bacterial infections.

Conclusion: Mortality rate in other studies mentioned about 4% which is very lower than our study (20.2%). Diagnostic delay can result in high mortality and more complications in untreated patients and failure to provide therapy can be a cause of low survival rate. Therefore early diagnosis, good control and appropriate treatment will improve the survival rate.

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Acute myocarditis after steroid withdrawal in an asthmatic patient

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A 64-year-old Caucasian man presented with febrile dyspnoea one month after having withdrawn steroidal therapy previously prescribed to treat asthma of recent appearance associated to a marked increase of serum IgE (2493 U/L). The finding of increased troponin and myoglobin levels on hospital admission, seemed to point to a cardiac ischemia, although ECG showed only sinusal tachycardia and despite of a leucocytosis with 60% eosinophils, Thus, he was directly referred to the cardiac intensive care unit, where he underwent a cardiac catheterization and a stent was inserted in the left anterior descending coronary artery. Three days later, cardiac enzymes started to increase again (troponin up to 64 μg/L) and the clinical picture worsened with severe dyspnoea and fatigue. For this reason, he had a further catheterization done, that showed no problem with the stent. In the meantime, paresthesias and neurological pain, slightly present on admission, worsened to a severe stage. The temperature was still high and cutaneous signs of vasculitis appeared. At this point a cardiac magnetic resonance was performed which revealed a diffuse sub-endocardic necrosis. Finally, thinking of a Churg-Strauss Syndrome (CSS), a myocardial biopsy was obtained and ANCA were tested, both confirming the working hypothesis. Moreover, EMG showed a severe mixed multi-neuropathy. High-dose-methylprednisolone was started, with prompt clinical improvement and normalization of cardiac enzymes in 10 days of therapy. CSS is a vasculitis characterized by asthma, hypereosinophilia and involvement of other organs. Being CSS more prevalent in the fifth decade, when a novel asthma presents in an individual aged 60 or more, it is common to think of a previously latent atopy, made overt by the concomitant intervention of precipitating factors, or of a cardio-pulmonary disease. In the present case report, the steroid withdrawal in a novel asthma of an aged patient caused the burst of a severe carditis in an undiagnosed CSS.

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Does breast feeding protect against asthma and other atopic diseases?

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Rationale: Breast feeding is associated with the lowest asthma and allergic disorders rates and it significantly reduces the severity of respiratory illnesses during the first year of life.

Methods: we had included questions related to breast feeding in the ISAAC questionnaires (the international study of asthma and allergies in childhood) to see the outcome of allergic diseases in both groups: breast feeding and non breast feeding.

Results: Mean (S.D) age: 9 years, weight: 30 kg and height: 135.6 cm. Children breastfed more than 8 months had significantly lower prevalence rates of persistent cough in the past year, ever rhinitis, rhinitis in the past year, and itchy-watery eyes in the past year than those who were not breastfed (18% vs 25%; 22% vs 32%; 15% vs 23%; and 6.5% vs 14.2%, respectively). Incidency was lower of asthma and wheeze in breastfed children than those who were not breastfed (asthma: 2.5y vs 4.5 y; 2.2y vs 3.9y, respectively). Significantly higher proportion of children who breastfed more than 8 months were resolved from asthma, wheeze and eczema than those who breastfed less than 8 months (89% vs 76%; 90.7% vs 77.2%, and 97.4% vs 82.2%, respectively).

Conclusion: Our Study shows the impact of breast feeding for at least 8 months in protecting and reducing the prevalence of allergic disorders.

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Transient IgM monoclonal gammopathy in an asthmatic child

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Background: Monoclonal gammopathies are extremely rare in children either primary or secondary. A case of a transient monoclonal gammopathy in a child is reported and the possible causes dicussed.

Methods: Patient JPA male, 5 years, consulting by Asthma and rhinitis perenneal worsening in fall and winter and a previous history of atopic eczema at 2 years. Objective examination was normal and prick skin tests only positive to house dust mites. Blood analysis were requested. Meanwhile an acute respiratory illness treated symptomatically by G.P. On analysis PBC count was normal, IgG 891 mg/dl IgA 105 mg/dl IgM 155 mg/dl. In electrophoresis a peak was detected which leads to an immunofixation showing an IgMK monoclonal gammopathy. Total IgE was a 160 Ul/ml. Specific IgE 1,5 KU/l for D.pteronyssinus and 2,23 KU/l for D.farinae. A battery of antibodies to Virus, Mycoplasma and Leishmania which is slightly endemic in south of Tagus river was requested. One week after new immunofixation showed a marked decrease of monoclonal peak and IgM decreased to 113mg/dl. No light chains in urine. 2 weeks after antibodies to EBV, CMV, Chlamydia pneumoniae Leishmania were negative. Antibodies to Mycoplasma pneumoniae positive: IgG 2,54, IgM 3,3 (normal<0,9). 4 weeks after new immunofixation was done with absence of the monoclonal peak.

Conclusion: Secondary gammophaties can be included in the broader group of monoclonal gammopathies of indetermined signification (MGUS). In general and with the exception of Leishmania infection a clear monoclonal peak is not detected after an acute infection. IgM is the immunoglobulin implied in primary response to infections. In children the occurrence of monoclonal gammopathies is very rare and mainly described for CMV and EBV. Transient monoclonal gammopathy is characterized by a fast decrease of the monoclonal peak after the infection. In our case a recent Mycoplasma pneumoniae infection was shown by positive serology for IgG as well for IgM. This young patient has a slight increased concentrations of total IgE and the values of specific IgE for house dust mites were not very high. The fast decrease and disappearance of the monoclonal IgM peak points out to a transient monoclonal gammopathy in an atopic child triggered by a Mycoplasma infection.

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Cystic adenomatoid transformation (CAT) of lung in a child with hyper IgE syndrome

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Hyper IgE syndrome with recurrent infections is a rare immunodeficiency characterized by recurrent skin infection and pulmonary abcess and extremely elevated levels of IgE in serum, associated facial and skeletal features have been recognized. Their frequency and genetic basis is poorly understood. The patient is a seven-year old girl presented with two years history of productive cough, superinfected facial eczema from infancy and two prior hospitalization for pneumonia and perianal abcess. She had coarse face, crackles on both lungs, clubbing of fingers, laxicity of elbow joints. Immunological work up revealed eosinophilia and markedly raised IgE levels. In view of the clinical features and other finding diagnosis of hyper IgE syndrome was suggested. Chest X-ray revealed multiple large cystic lesions in left total lung which confirmed by spiral CT-scan. Left thoracotomy and pneumanectomy was done and removed severe adhesions. Histological report revealed cystic adenomatoid transformation. CAT is Characterized by the presence of variously sized cysts lined by epithelium. Probably, this is the first report of CAT in a child with hyper IgE syndrome due to repeated infections. Early diagnosis and prophylactic antibiotic therapy is effective in management of these patients.

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Pulmonary complications and some changes in immune reactivity on the course of acute heroin intoxication

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Objective: To examine the complications in pulmonary system and changes in immune reactivity (some parameters of humoral and cell mediated immunity) on the course of acute heroin intoxication.

Materials and Methods: The study includes 16 patients, with acute heroin and mixed with other psychoactive drugs intoxications at average age, 21,5 ± 5.04 years (12 men and 4 woman), hospitalized in the Clinic of Toxicology, MHATEM "N.I.Pirogov", Sofia. We have used clinical, clinico-laboratory, immunological, chimicotoxicological, instrumental methods.
Results: Results from the study shows that in severity intoxications with heroin and other psychoactive drugs we observed spectrum of complications in pulmonary system - pneumonia, aspiration of gastric contents, pulmonary edema, and ARDS. Death is registered in 3 persons. It was found some changes of the immune reactivity: statistically significant lower mean levels of IgG, IgA, IgM and tendency to lower mean levels of complement components - C3 and C4 in the studied patients in comparison with the values in healthy people. The changes were more demonstrative in the group with pulmonary complications compared to the group without pulmonary complications. We observed that the CD4 lymphocytes are significantly less in the studied patients and is determined lower level of CD56 bearing lymphocytes - natural killer /NK/ cells, compared to healthy controls.
Conclusion: The acute heroin and mixed with other psychoactive drugs intoxications leads to complications in pulmonary system, and changes of some parameters of cell mediated and humoral immunity.
Key words: intoxication, heroin, psychoactive drugs, immune reactivity, noncardiogenic pulmonary edema, pneumonia, ARDS

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Atypical presentation of polyarteritis nodosa: a case report

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Polyarteritis nodosa is a systemic vasculitis that affects median sized muscular arteries, characterized by multi organ involvement. Presenting GI manifestations include: abdominal pain, nausea, vomiting, diarrhea, acute abdomen and, rarely, GI bleeding in the form of hematemesis, melena and occult bleeding. A 14 year old boy presented five years ago with hemetochezia, 5 to 7 episodes per day. he presented severe anemia and he was hospitalized to investigate mieloproliferative disease or systemic lupus erythematosus He was treated with IVIG and methylprednisolone with clinical and hematic biometry improvement. A colonoscopy showed suggestive data of CUCI with diffuse inflammation, pseudopolyps, adhered fibrin to mucosa, inflammatory lesions involving descendent colon and sigmoides, estenotic parts of the colon and bleeding. Hystologic examination described ulcer and inflammatory infiltrate with polimorphonuclears, limphocytes, plasmatic cells in lamina propia and submucosa, mild fibrosis and eosinophils. He received treatment with methylprednisolone, mesalazine, omeprazole, and azathioprine and he was referred. Physical examination revealed severe emation 45.4 kg, hepatomegaly, splenomegaly, bilateral pulmonar hipoventilation with bilateral crackles. The diagnosis of severe desnutrition, CUCI, and communitary pneumoniae were made, and treatment with cefuroxime and claritromicine was started. He presented fever, rectorragia with hemodinamic compromise that required transfusion of packed red blood cells, and platelets, and involvement of liver, kidney, gut and persistent fever, pancytopenia and elevated ferritin. He required orotraqueal intubation because ventilatory compromise, in the neurology area, he had hypotonic proximal extremities, hiporreflexia, and generalized seizures, which were controlled with phenytoin. The laboratory findings revealed negative, antinuclear antibody, perinuclear antineutrophilic cytoplasmic antibody, hepatitis B surface antigen, and anti hepatitis C virus were negative. Central nervous computed tomography showed isquemic temporal areas. Magnetic resonance and angiography revealed microaneurysm in both renal arteries, and vasculitis in central nervous system, hepatic and intestinal arteries. These symptoms and laboratory findings were compatible with diagnostic criteria for childhood classic polyarteritis nodosa, treatment of IVIG and various methylprednisolone pulses let to clinical improvement.

IMMUNOTHERAPHY

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The evidence on the efficacy of Specific Immunotherapy in the treatment of respiratory allergy: A randomized controlled trials database

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Background: According to Evidence Based Medicine, conclusions from meta-analyses and RCT represent the most solid evidence source to assess the efficacy of an intervention. Specific immunotherapy (SIT) is currently considered as the only allergen-oriented biological response modifier, and it is regarded as an essential part of the therapeutic approach for allergic diseases.
Objective: To provide a descriptive review of all available randomized, double-blind and placebo-controlled trials (RCT BD PC) published which evaluated the efficacy of SIT in the treatment of respiratory allergy.
Study Design: Systematic review.
Methods: A comprehensive search of the EMBASE and MEDLINE databases up to April 30th, 2007 was carried out. The search strategy retrieved citations containing the subject heading specific immunotherapy limited to RCT DB PC combined with asthma, allergic rhinitis or both. Variables recorded were the study design, journal impact factor, patient's and immunotherapy characteristics, clinical and laboratory outcomes and online full-text papers' availability.
Results: Two hundred ninety-six articles were identified and reviewed; 179 (61%) were RCT DB placebo-controlled. Of them, 116 evaluated efficacy of subcutaneous IT (SCIT) and 63 sublingual IT (SLIT). Twenty-six percent of studies (n= 46) had a minimum duration of 24 months (21% vs. 30% for SCIT and SLIT, respectively). Characteristics of studies are described in

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	SCIT			SLIT		
	Rhinitis	**Asthma**	**Both**	**Rhinitis**	**Asthma**	**Both**
DB Placebo RCT	37 (32%)	32 (28%)	47 (40%)	30 (48%)	6 (10%)	27 (42%)
Sample Size +	40 (33-53)	28 (20-55)	35 (27-60)	56 (30-83)	30 (21-99)	37 (24-79)
Grass/mites (n) #	13/6	5/15	10/10	15/4	1/3	8/6
Time endpoint +*	12 (5-24)	12 (12-24)	12 (10-15)	10 (6-24)	10 (5-24)	12 (5-18)
Journal Impact factor **	5.3	5.2	4.1	3.5	2.6	3.1
Mechanisms (n) @	26	28	37	20	5	21
Full text Online	60%	50%	60%	73%	100%	70%

Table. (+ Median-Interquartilic rank; # Number of studies found; * Months; ** Mean; @ At least one immunological mechanism studied)

Conclusion: One hundred seventy-nine Ib evidence-level studies on efficacy of SIT in the treatment of respiratory allergy were identified in two electronic databases. Both selection and search bias must be ruled out.

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The effect of antihistamine pretreatment on side effects during specific immunotherapy

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During one year of our study in a group of 47 patients we have conducted the specific immunotherapy (SIT) with House Dust Mite (HDM) Novo Helisen Depot extracts and pollen Allergovit extracts. 493 shots were applied and 32 local reactions occurred (6,49%). All of them were > 5 cm and all of them occurred before the maintenance dose was reached. No systemic reaction or life-threatening reactions were observed. In a group of HDM treated patients 5,21% of injections resulted in large local reaction versus 7,45% in a group of patients with pollens. There was no statistical difference between those two groups p=0,31. Reductions in IT-schedule were performed due to severe local reactions. Only 6 patients experienced more than ones a large local reaction. Four of them were on pollen immunotherapy and two were on HDM immunotherapy. All of these patients have had skin reactivity on skin prick test (SPT) more than 3+. We have decided to use loratadine 10mg as antihistaminic premedication in these patients. After the administration of the pretreatment antihistaminic drug, patients did not experience any severe local reaction.

We can conclude that in a very limited number of patients who underwent SIT (especially in those with very extensive cutaneous reaction to SPT) it could be useful to use antihistaminic premedication to reduce/secure the number and severity of cutaneous reactions.

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Shorter dosing intervals of sublingual immunotherapy lead to more efficacious treatment in a mouse model of rhinitis

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Background: Sublingual immunotherapy (SLIT) with allergen extract has been shown to be efficacious in treating allergic rhinitis and asthma. Clinically, SLIT is performed with varying doses of allergen extracts and with varying intervals of dosing, and although a dose-response relationship has been established in SLIT, the importance of dosing intervals remains to be documented. The objective of this study was to explore the importance of the dosing intervals in SLIT. For this purpose we used a mouse model of allergic rhinitis, induced by a clinically relevant allergen, timothy grass pollen extract.

Methods: Mice were sensitized to Phleum pratense (Phl p) by intraperitoneal injections of alum-adsorbed allergen extract. Sensitized mice were SLIT-treated with different intervals and with different doses of Phl p extract. In order to study the importance of dosing intervals, doses of 25 kSQ, 58 kSQ or 175 kSQ were given sublingually to mice 7, 3 or 1 day per week, respectively, resulting in a weekly cumulative dose of 175 kSQ for all groups. Two additional groups receiving the lower dose (25kSQ) of Phl p one or three days per week were also included. Following nine weeks of SLIT, all mice were challenged intra-nasally with Phl p. Antibody levels, T cell responses and numbers of inflammatory cells in bronco-alveolar lavage (BAL) were used as read-outs for the efficacy of SLIT in the different treatment groups.

Results: Increases in the Phl p specific IgA antibody levels showed a strict positive correlation with increased dosing frequency. The levels of Phl p specific IgE were reduced to the same extent in all groups, but both the systemic- (spleen) and local (draining lymph nodes) T cell responses decreased with increased dosing intervals, indicating a positive effect of higher dosing frequency on T-cell tolerance. In addition, the maximal reduction in allergen-induced inflammation in the lungs (BAL) was seen in the group receiving SLIT for 7 days per week.

Conclusion: The group receiving SLIT for 7 days per week was the only group with demonstrated positive effects of all immunological parameters measured. Our data demonstrate that although the effect of SLIT is dose-dependent when using the same allergen extract, a once daily regimen is more efficacious than regimens where SLIT, with the same weekly cumulative allergen dose, is administered with longer intervals.

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Faster reach to maintainance pleteau by delayed rush immunotherapy

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Introduction: Intermittent Rush immunotherapy was tried on some patients to evolve some faster and affordable immunotherapy modality to make the patient achieve the maintenance pleatu within a very short time span which does not also involves hospital stay. Conventional method of immunotherapy is administered with long durations, rush immunotherapy is super fast methodology in attaining the maintenance/boosting module, which requires hospitalization and other precautionary methods, and multiple allergen vaccines to be administered within short span of time. But in this method which was tried it was found that within a duration of around 15-20 days the relief of the immunotherapy was reached. Place of study: 1) Belle Vue Clinic, kolkata, 2) Hope Super Speciality Hospital, Purnea.

Materials and Methods: 62 patients out of which 46 with urticaria allergy and 16 with allergic Rhinitis. Were subjected for Intermittent Rush Immunotherapy schedules. The therapy consists of administration of four vials of allergen extracts, 1st vial : 1:25,000 dil, 2nd vial 1:2,500 dil, 3rd vial 1:250 dil, 4th vial 1:50 dil. The 1st & the 2nd concs were administered in daily schedules in a graphically rising manner. The patients had been given pre-medication before the study. Follow-up of the patients included blood examination IgG & IgE level estimation, before & after 8 weeks after the procedure.

Results: Some of the patients showed local skin reactions which subsided on duration of time no drugs were required and 7 patients developed systemic reactions which were managed by Deriphylline & Decdan injections. There was substantial decrease in IgE level & increased IgG level was marked, significant & marked satisfactory relief was observed in the patients symptomatology, thus the procedure may be graded as a very fast & affordable immunotherapy.

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A two year double blind placebo controlled study on immunotherapy with single and mixed allergen preparation

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Background: Studies on immunotherapy with single allergen extract have been performed to determine the efficacy in asthma and rhinitis patients. However, very few studies have been performed comparing single and mixed allergen immunotherapy in a double blind manner. Here, we have carried out a double blind placebo control trial to assess the efficacy of single and mixed allergen immunotherapy based on various clinico-immunlogic parameters.

Methods: One-hundred allergic patients were screened by history, skin tests and specific IgE levels. Sixty patients fulfilling the criteria of the study were

divided randomly into two groups – (a) active- single allergen, (b) active-multiple allergen and (c) - placebo containing 20 patients each. Single allergen immunotherapy was given with housefly extract. In mixed allergen immunotherapy, insect, pollen and fungal allergen extract were used in vaccines. In vivo parameters such as skin test and airway reactivity were assessed at baseline and after 2 year's of immunotherapy. In vitro parameters such as allergen specific IgE, IgG1 and IgG4 were also assessed. Apart from these, symptom/drug score cards were filled by the patient and analyzed every 3 month. Efficacy of the treatment was analyzed in terms of clinical (skin test, symptom score, and airway reactivity) and immunological parameters (IgE, IgG4, IgE/IgG4, and IgG1/IgG4).

Results: Patients getting active immunotherapy in both single as well as multiple allergen group showed significant improvement ($p<0.01$) in disease state in comparison to placebo patients. Skin tests and IgE values however showed non-significant reduction compared to baseline. There was a significant improvement ($p<0.01$) in airway hyperreactivity, symptom score and IgG4 levels after 2 year of immunotherapy in active group. Also good correlation was observed between symptoms reduction and increased IgG4 levels. All the above parameters were non-significant in placebo group. In the group treated with the single extract, fewer patients suffered with asthmatic symptoms after 2 years of IT and required less medication than in the group treated with the multiple extract. However, these differences, as well as differences in other clinical and immunological parameters were not statistically significant.

Conclusion: Both single and mixed allergen immunotherapy for 2 years have shown improvement in clinical and immunologic parameters of asthma and rhinitis patients.

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A role of immunotherapy in allergic conjunjuctivites

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Introduction: Allergic conjunctivitis is many times an ophthalmologist's nightmare. Antiallergic eye drops which often contain corticosteroids, may temporarily bring relief of symptoms but may bring an early cataract, among other side effects. Allergy testing and immunotherapy based on the test results can cure a significant number of these patients and provide relief to others.

Methods: 24 patients of allergic conjunctivitis were subjected to modified skin allergy testing. Out of these 10 patients also had associated allergic rhinitis and asthma. Common allergens found in these patients were: Dust, Dustmites, Pollen, Fungi & Food. Based on test results, vaccines were prepared for individual patients and immunotherapy given. The patients who did not significantly respond to conventional medicines and vaccine, we reviewed the history to discover any missed out allergen. A specially designed indigenous "personal volumetric air sampler" with safranine stained glycerin coated slides inside, was kept in patients home, office and surrounding atmosphere.

Results: Out of 24, 18 patients (75%) started improving within 3 months of starting immunotherapy. After 6 months their eye drops and other medicines were stopped. Restricted food items started one by one after 6 monthes. They are symptoms free for last 1½ to 2 years. For 6 patients who did not respond to above therapy, specially designed "air sampler" brought great surprises. The fungi and pollens, which were not tested, previously were found. Also three new pollens were discovered. Appropriate immunotherapy made 5 out of 6 patients symptoms free.

Conclusion: Allergy testing and immunotherapy can cure allergic conjunctivitis. Personal volumetric air sampler is a very useful tool to find out additional allergens in case of resistant cases.

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Quality of Life and Symptoms assessment in sublingual immunotherapy for patients with house-dust mite related perennial rhinitis: definition of a responder profile

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Background: The efficacy and safety of sublingual immunotherapy (SLIT) in well-established in seasonal allergic rhinitis. However fewer data were available regarding perennial allergic rhinitis. The aim of this study was to assess the relationship between the effect of SLIT on symptoms and Quality of Life changes in patients with perennial rhinitis.

Methods: This exploratory phase IIIb, double-blind, placebo-controlled trial was conducted during years 2003 to 2005 in the Cape-Town area (South Africa). Sixty patients having house-dust mite-induced allergic rhinitis were enrolled. Patients were randomized to receive, during 2 years, Dermatophagoides pteronyssinus SLIT solution (Stallergenes, Antony, France), with a maintenance dose of 300 IR (n = 39), or placebo (n = 21). The primary efficacy endpoint was the mean T5SS [Total Symptom Score for five symptoms: sneezing, runny nose, nasal congestion; ocular redness/itching/tearing; itchy nose/throat/ears. The limit percentage of improvement was fixed at 60% for the "responders". Rescue medication intake, individual symptom scores and quality of life (QoL-RQLQ) were assessed as secondary endpoints.

Results: The intention-to-treat (ITT) population included 55 patients (mean age: 32.93 yrs ± 11.31). The mean T5SS change was lower for the 300 IR [-6.57] group compared to placebo [-5.02], but the difference did not reach significance. The mean percentage of days with rescue medication, as well as each individual symptom score were lower for the 300 IR group compared to placebo. The percentage of good responders is significantly higher in the active group ($p=0.0405$). For the QoL, the percentage of good responders the ocular score is higher in the active group ($p=0.0512$), reaching significance. The association between good responders to QoL and good responders to T5SS gives an odds ratio = 15 ($p<0.0001$) for all patients (OR = 8.75, $p=0.0061$ in active group) showing a good correlation. The most frequent adverse events were oral pruritus and throat irritation. No related SAE occurred throughout the study.

Conclusion: This exploratory study gives promising results with a two-year treatment using Dermatophagoides pteronyssinus SLIT solution at a maintenance dose of 300 IR three times a week. A population of "responders" may be defined as patients with an improvement of 60% or more of the clinical symptoms. QoL is strongly correlated to clinical symptoms.

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Measuring quality of life in the treatment of allergic rhinitis with specific immunotherapy - identifying the best suitable instrument

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Background: Quality of Life (QoL) is becoming an increasingly important clinical outcome parameter, also in treatment of allergic rhinitis (AR). In AR QoL is often assessed with Rhinoconjunctivitis QoL Questionnaire (RQLQ) - a disease specific instrument. For pharmacoeconomic evaluation a generic QoL instrument must be used. RQLQ only detects symptoms in the upper airways. Due to a strong epidemiologic relationship between AR and asthma suggesting that both are manifestations of the same disease, an instrument also detecting symptoms in the lower airways seems more appropriate. To show the full impairment of QoL caused by AR and to perform pharmacoeconomic evaluation it is important to identify the right generic instrument to be used in clinical trials with specific immunotherapy (SIT).

Methods: QoL gain with the grass allergen tablet (Grazax, ALK-Abelló) against AR was assessed in a randomised, parallel-group, double-blind, placebo-controlled trial with 634 subjects using the generic EQ-5D questionnaire. EQ-5D contains 5 dimensions: mobility, personal care, usual activities, pain/discomfort and anxiety/depression, with 3 levels within each dimension. For each of the 5 dimensions the average score was calculated. It was then tested if the difference in scores between active and placebo was significantly different (t-test).

Results: The treatment with the grass allergen tablet significantly increased QoL compared to placebo ($p<0.001$). The difference between groups was mainly due to differences within the dimensions: Usual activities and Pain/discomfort. Mobility did also produce a borderline significant difference (Table 1).

	Mobility	Personal care	Usual activities	Pain/ discomfort	Anxiety/ depression
Grass allergen tablet	1.0472	1.0121	1.0967	1.1332	1.0780
Placebo	1.0716	1.0185	1.1498	1.2169	1.0864
p-value (t-test)	0.095	0.468	0.003	<0.0001	0.666

Conclusion: Although there is a significant increase in QoL for subjects receiving treatment with the grass allergen tablet, the increase was only justified in 2 of the 5 dimensions. EQ-5D is a very crude instrument and might not be optimally suited to detect QoL impairment in a disease like AR. This implies an underestimation of the QoL result with the grass allergen tablet. An instrument with a more detailed focus on usual activities could be more relevant to use within SIT and AR in future studies. Examples of potentially well suited instruments are SF-36 and EQ-15D.

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Long-term comparison of the efficacy of adding sublingual immunotherapy or montelukast in birch-induced asthma

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Background: There are no studies comparing the effects of immunotherapy and antileukotrienes. The main limit is the long duration required to appreciate the clinical effects of immunotherapy. We compared the effect of montelukast or SLIT, added to standard therapy in moderate persistent asthma up to 4 years.

Methods: Open controlled randomized trial. Patients with moderate asthma (and rhinitis) solely due to birch pollen were randomized to receive either monteukast MK (20 mg/day) or birch-SLIT (Anallergo, Florence, Italy) in the pollen seasons, in addition to formoterol 25 mcg+fluticasone 500 mcg twice daily. All patients received also salbutamol and cetirizien as rescue medications. Asthma and rhinitis symptoms were evaluated by diary card February-may at baseline and after 2 and 4 years of study. In-season nasal eosinophils and outseason bronchial hyperresponsiveness were evaluated as well.

Results: Thirty-three adult patients were enrolled and 29 completed. The groups were homogeneous at baseline. In the SLIT vs MK e group the total asthma symptom score at baseline, 2 and 4 years were: 178 vs 180 (NS), 76 vs 183 (<0.001) and 42 vs 167 (< 0.001). At the same timepoints the bronchial scores were 80 vs 92 (NS), 48 vs 87 (0.01) and 25 vs 81 (<0.01). In both group there was a significant decrease in the use of bronchodilators but in the SLIT group there was a significant decrease of nasal eosinophils and an increase of outseason bronchial hyperresponsiveness as compared to baseline and to MK group.

Conclusion: In patients with grass-pollen induced moderate asthma and rhinitis the addition of SLIT provides a greater clinical benefit compared to MK.

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SLIT response in children with HDM and cockroach

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Background: Recent times the allergy disorders is increased and most of the therapeutic approach is incomplete and yet not promising. In recent time the SLIT emerged with patient friendly Immunotherapy. This became a good boon among the children who were depend only on the symtpmatic drugs. This SLIT became an eye opener to Physicians and to parents to control the allergy symptoms. SLIT is introduce in Indian continent and made it possible at affordable price and showed good response in preventing the allergies.

Methods: A total 112 children aged about 2 to 14 yrs from various parts of India who were previously diagnosed with Allergic Rhinitis and Allergic Asthma and reported receiving allergy and asthma medication took part in this study. These children were tested with aeroallergens by SPT and selected for SLIT who were sensitive to House dust mite (body and fecal matter) sensitivity and among insects who were sensitive to Cockroach.

Results: Most of the patients' parents reported with good response and the pharmacotherapy drugs relatively reduced over a period of time and the symptoms drastically came down with increase PEFR. The drop outs were more fascinated to the alternative medicine and they continue to search for the relief from one system to another with continued suffering. Some reported frankly and some said they feel better.

Conclusion: The results of this study provide insight into how the SLIT greatly influence the therapy and increase the quality of these children and the normal growth pattern which is appreciated with most of the parents.

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Long-term comparison of the efficacy of the efficacy of sublingual immunotherapy vs inhaled budesonide in grass-induced asthma

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Background: There are few study comparing the effects of immunotherapy and inhaled steroids. The main limit is the long duration required to fully appreciate the clinical effects of immunotherapy. We compared the effect of inhaled budesonide vs SLIT in mild persistent asthma up to 4 years.

Methods: Open controlled randomized trial. Patients with mild persistent asthma and rhinitis solely due to grass pollen were randomized, after a run-in season, to receive either budesonide 800 mcg/day in the pollen season or grass-SLIT (Anallergo, Florence, Italy) continuously for 4 years. Rescue medications were prescribed to all patients. Asthma and rhinitis symptoms were evaluated by diary card may through july at baseline and after 2 and 4 years of study. In-season nasal eosinophils and outseason bronchial hyperresponsiveness were evaluated as well.

Results: Fifty-one patients were enrolled and 46 completed. The groups were homogeneous at baseline. In the SLIT vs budesonide group the total nasal symptom score at baseline, 2 and 4 years were: 120 vs 92 (NS), 56 vs 99 (<0.01) and 36 vs 108 (<0.001). At the same timepoints the bronchial scores were 180 vs 161 (NS), 85 vs 138 (NS) and 56 vs 122 (<0.01). In both group there was a significant decrease in the use of bronchodilators and no change in FEV1. In the SLIT group there was a significant decrease of nasal eosinophils vs the budesonide group at 2 and

4 years ($p<0.01$). The outseason bronchial hyperresponsiveness improved significantly only in the SLIT group.

Conclusion: In patients with grass-pollen induced asthma and rhinitis SLIT was not inferior to inhaled budesonide concerning bronchial symptoms. On the other hand, SLIT provided additional benefit on rhinitis symptoms and outseason bronchial hyperresponsiveness

Key words: SLIT, Budesonide, Asthma

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Safety and compliance of sublingual immunotherapy in spray

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Background: Immunotherapy by sublingual route (SLIT) is nowadays largely employed. Currently by drops or tablets. Sublingual spray has been used for other therapies. This route offers a great potential for immunotherapy increasing the area of mucosal contact with allergen and the rate of absorption. A first trial of SLIT-spray has been done in order to validate safety and patients compliance.

Methods: 40 patients with respiratory allergy, allergic rhinitis, asthma or both have been included in the trial. Informed consent has been obtained from all patients. SLIT spray has been given in a standardized schedule from 4 vials in progressive concentrations A 1/125, B 1/25, C 1/5, D 1/1. For mites the vaccine was standardized at 5 mcg/ml for Derp 1+Derp2 and Derf1+Derf2. Cummulative dose at the end of induction was for Derp 1 around 490 mcg (DIATER Laboratories, Spain). The composition of SLIT-spray was choosen in each case by history, skin tests and specific IgE. The procedure has been carefully explained at all the patients. Daily application for VIALS A,B,C 1 to 4 puffs. D manutention 1 puff daily continued 2 months more. A booklet with instructions was given to the patients and a visual scale from 0 to 5 points marked in a second appointment: convenience, practicability, clarity of instructions and general evaluation. Side effects were registered for each dosage of each vial. Symptomatic therapy was allowed during SLIT-spray.

Results: 40 patients, 30 adults (16–45) (18 M, 12 F) and 10 children (5 M 5 F) (9–15). Allergens used, Dpteronyssinus 28 cases, D.farinae 26, Lepidoglyphus 11, Tyrophagus 2, Blomia 1, cat 1, dog 1, grass pollens mix 6, Parietaria judaica 3, Olea 1, Plantago 3.

Adverse effects in 8 patients only one withdrawal. adverse events were reported. Adverse events reported: urticaria 1, bad taste 1, tongue swelling 2, nausea 1 (withdrawal), sneezing 1, fever 1, wheezing 2, rhinorrea 1,(20%). Adverse events disappear with prosecution of immunotherapy. Evaluation by patients: convenience, practicability, comprehension of instructions and general scores of 4,3–4,2–4,0 and 3,5 in a 0–5 scale.

Conclusion: SLIT-spray is a new safe and convenient form of SLIT. High dosage is quickly attained, side-effects relatively rare. The evaluation by patients give good level of appreciation. Larger studies must be done in order to confirm these results.

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Role of SLIT with D.farinae in allergic conjunctivitis

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Background: Perennial conjunctivitis due to House Dust Mite is the most frequent form of allergic conjunctiviis in urban environments. However, its diagnosos remains difficult for the ophthalmologists. In India, very little work is done in this field. Keeping this in mind, we have tried to present a case report with Sublingual Oral Immunotherapy.

Methods: A male patient, Mr. Pravin J. Patel, 34 yrs,residing at Ahmedabad was suffering from Perennial Allergic Conjunctivitis for over 4 years. Routine therapy including local antiinflammatory drops, antihistaminics, etc didn't give him long term relief. The antigenspecific conjunctival provocation test, skin prick tests and ImmunoCAP RAST were carried out with D.pteronyssinus and D.farinae mites. CPT was positive at low dilution allergen of D. farinae and negative for D.pteronyssinus. Skin Prick Tests showed two plus sensitivity to D.farinae. ImmunoCAP RAST revealed Class VI (Very High) sensitivity to D.farinae. Skin prick tests showed one plus positivity and ImmunoCAP RAST showed Class I response to D.pteronyssinus. The extract to be given for SLIT containing D.farinae was tested for Conjunctival Provocation Test and Skin Prick Test to confirm the positivity. The SLIT was given as twice a day schedule,initially in diluted form and later on with increasing concentration. The maintenance dose was then continued. Clinical assessment, Skin Prick Tests and ImmunoCAP RAST were carried out at 6 months and 12 months interval. The patient is still under SLIT therapy.

Results: It was observed that the patient was symptomfree at the end of six months and the redness and swelling had totally disappeared. Skin Prick Tests revealed one plus sensitivity to D.farinae at 6 and 12 months interval. ImmunoCAP RAST showed 46.6 kUa/l (grade IV) and 16.5 kUa/l(grade III) at the end of 6 and 12 months respectively.

Conclusion: Sublingual ORAL Immunotherapy has shown excellent clinical response with significant improvement in skin prick test and ImmunoCAP RAST test results in case of Perennial Allergic Conjunctivitis due to D.farinae.This is an isolated case and much more cases need to be studied.

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Safety, tolerability and efficacy of sublingual allergoid immunotherapy starting with a 4-day build-up scheme. Rationale and study design of an ongoing multicentre Portuguese study in allergic patients

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Rationale: The good efficacy, safety and tolerability of the sublingual monomeric allergoid (allergoid SLIT, Lais®, Lofarma S.p.A., Milan, Italy), either in tablets or drops, has already been shown in many studies. Yet, so far, only one study on allergoid SLIT starting with a 4-day build-up phase to achieve in a shorter time the maximum suggested dose for the maintenance therapy was performed in 39 allergic patients with promising results. The objective of the present study, still ongoing, is thus to confirm, in a broader population, if this approach is really useful and safe.

Study design: One hundred and thirty patients, gathered from the normal daily clinical practice of 16 Portuguese doctors, with an history of oculorhinitis and/or rhinitis with or without mild-moderate asthma, were enrolled during the last 24 months. The build-up rush scheme involved the administration of daily increasing doses of the allergoid SLIT: 500 AU the first day, 1000 AU the second, 1500 AU the third and 2000 AU the fourth. The cumulative dose administered in 4 days was 5000 AU. Also during the maintenance period the treatment dose was kept higher than one normally used. Main endpoints of the study were both tolerability and safety, as well as treatment efficacy. For the evaluation of this one, patient's subjective perception has been taken into consideration as well as the variations in the use of additional drugs for symptom control during SLIT.

Results: The final results will be presented at the end of the study in a subsequent poster.

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Sublingual immunotherapy for paediatric allergic rhinitis

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Background: No immunotherapy programme existed in KK Hospital prior to 2006. Work commenced in late 2005, to start sublingual immunotherpy (SLIT) as a new clinical service.

Methods: Paediatric AR patients, with moderate persistent symptoms that are not responding to conventional pharmacotherapy, were enrolled. Other inclusion criteria include house dust mite sensitisation, children 5 years and above, with committed and motivated parents/patients. Doctor visits are scheduled at baseline, 1st, 2nd months and 3 monthly intervals thereafter. An ENT review is performed to exclude any anatomical lesions. Allergy symptom scores were completed for 2 weeks prior to each visit and the mean score calculated. Mean rhinitis quality of life (QOL) questionaires were collected at 6 monthly intervals using a computerised database at the Allergy laboratory. Medication usage and adverse events were documented in a file diary provided. House dust mites allergen extracts (Dermatophagoides pteronyssinus and/or Blomia Tropicalis) from Stallergenes (Staloral®) were used, and immunotherapy was initiated as per recommended protocol.\

Results: 12 patients, 58% boys, were recruited over one year. The mean age was 12 years old (range 5.5–19 years). All had moderate persistent allergic rhinitis, 91.7% had concomitant allergic conjunctivitis, 33.3% had asthma, and 33.3% had eczema. Most (91.7%) have sensitisation to 2 different species of house dust mites, and were prescirbed separate HDM allergens for SLIT. There is an improvement in the mean allergy symptom scores in most patients during the first 6 months ranging between 28–53%. There is also a reduced mean QOL scores from 40.4 to 22.7, though not reaching statistical significance. There were no serious adverse events or anaphylaxis during SLIT treatment. The most common adverse reaction is sublingual itch 66.7%, which is transient and easily treated. In addition, there were complaints of mild abdominal discomfort and nausea in 2 patients; urticaria rash in 1 patient. One patient had deterioration of pulmonary function and cough 6 months after SLIT, requiring addition of inhaled corticosteroids for asthma.

Conclusion: SLIT is a safe and effective treatment for paediatric persistent AR. Good patient-medical team rapport is essential to the success of such programmes.

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Follow up study in patiens with pollinosis after sublingual immunotherapy (SLIT) compared to subcutaneous shorttime-immunotherapy (SCIT)

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The purpose of this study was to evaluate clinical efficacy and safety of sublingual immunotherapy (SLIT) in comparison to subcutaneous immunotherapy (SCIT) in patients with allergic rhinitis.

Alltogether we evaluated immunotherapy in 150 patients via inquiry scedule. 101 patients were treated with SLIT and 49 patients were treated with SCIT.

Patients treated with SLIT experienced a greater reduction of allergic symptoms (80%) than patients treated with SCIT (75%). In both therapies a reduction of antiallergic medication and an increase of quality of life was shown. The main adverse reactions in SLIT were burning and swelling of the mouth base mucosa especially during dose intensification. In SCIT a local swelling and induration at the injection sight was shown which disappeared completely within 4 to 6 weeks after the last injection.

Due to the high compliance of patients and the good results in both strategies the decision for the right therapy for each patient should be made in regard of the patients individual needs and ailments.

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Tolerance of high dose sublingual immunotherapy with pollen extracts administered under rush and ultra-rush schedules

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Background: High dose sublingual immunotherapy (SLIT) has been demonstrated as an effective alternative to subcutaneous immunotherapy (SCIT). However high doses could lead to adverse reactions, thus the tolerance of every type of extract should be ascertained. Parietaria and grass pollens are two of the most prevalent aeroallergens in southern European countries. In this work, we tried to asses the tolerance of P. judaica and grass high dose SLIT under two induction phases.

Methods: Biologically standardized extracts of P. judaica and grass pollen were used to prepare vaccines at a concentration which reached monthly accumulated doses of 20 and 50 fold, respectively, regarding conventional SCIT. Par j 1, Par j 2 and grass group 1, were controlled in the final preparations. Thirty-two subjects received grass SLIT and twenty P. judaica SLIT. Rush schedule consisted in the administration of one drop (50 µl each) the first day, two the second and thus successively until reaching five drops the fifth day. Ultra-rush consisted in the same volumes given all in the same day within 30 min intervals. Adverse events were registered and graded according to EAACI.

Results: There was no systemic reaction nor in the patients treated with grass vaccine neither in the patients treated with Parietaria. Only four out of the 22 patients treated with grass vaccine administered by an ultrarush schedule, showed local reactions in the induction phase (3.6% of doses). Over the whole period (6 months), eleven patients experienced some kind of local reaction (itching and/or swelling), that is 0.38% of the doses. There were three withdrawals due to local reactions. In the 30 patients under the rush schedule, 8 local reactions appeared in the build up, that is 5.3% of the doses. In the whole period, local reactions rose to nine (0.25% of the doses). There was only one withdrawal in the Parietaria group of the study due to repeated local reactions.

Conclusion: High dose SLIT vaccines with pollen extracts showed an acceptable safety profile administered by accelerated schedules.

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Short term efficacy of sub-lingual spray immunotherapy

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Background: In the last few years sublingual immunotherapy (SLIT) has gained increasing popularity at least in Europe and all allergen producers laboratories have launched sublingual drops. Sublingual sprays have a larger mucosal contact than drops or tablets increasing also the rate of absorption. Therefore this method of dispensing drugs and allergens can also be applied to SLIT. The efficacy of this technique must be assayed.

Methods: 40 patients with respiratory allergy, allergic rhinitis, asthma or allergic rhinitis asthma syndrome have been included in the trial. Informed consent has been obtained from all patients. SLIT spray has been given in

a standardized schedule from 4 vials in progressive concentrations A-1/125, B 1/25, C 1/5, D 1/1. For mites the vaccine was a standardized at 5 mcg/ml for Derp 1+Derp2 and 5 mcg/ml, for Derf1+Derf2 and Derf1. Cummulative dose at the end of induction was for Derp 1 around 490 mcg (DIATER Laboratories, Spain). The composition of SLIT-spray was chosen in each case by clinical history, skin tests and specific IgE. In children less than 10 years half-concentration of SLIT-spray was employed. The procedure has been carefully explained at all the patients. Daily application for VIALS A,B,C from 1 to 4 puffs each day. Vial manutention dosage 1 puff daily. This dosage being attained only after 13 days and continued 2 months more. A booklet with instructions was given to the patients and a visual scale from 0 to 5 points registered in a second appointment to evaluate facility, convenience, practicability, clarity of instructions and general evaluation of treatment. Side effects were registered for all the patients for each dosage. Symptomatic therapy was allowed during trial with anti-histamines nasal steroids, and formoterol-budesonide when necessary.

Results: Short-term efficacy of immunotherapy was good in all patients either for house dust mites allergy or pollens. Evaluation by the patients give on a visual scale an average of 3,5 on 5 points (70%). Medical evaluation was of good (12) or very good (24) results in the patients even in the patients treated during pollen season. Only 1 patient withdrawed from the trial due to the tongue swelling and nauseous state. SLIT spray is effective generally well tolerated and very convenient for the patients. Larger series of SLIT-spray must be evaluated in order to confirm these preliminary results.

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Sublingual immunotherapy with canova, a homeopathic formulation

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Rationale: Clinical efficacy of sublingual immunotherapy has been established using allergenic extracts, but there are very few studies using plant extracts. Canova a homeopathic formulation consisting of aqueous extract of Aconitum nepellus, Arsenicum album, Bryonia alba and Thuja occidentalis has been used as immunomodulator along with subcutaneous immunotherapy in patients suffering from different allergic disorders.

Methods: In a multi centre study, 30 subjects were selected, who had sensitivity to various allergens, confirmed by Skin Prick Testing, and were undergoing allergen specific immunotherapy course. These subjects were advised for sublingual immunotherapy with Canova along with subcutaneous immunotherapy (SIT). An other group was administered only SIT to compare the results.

Results: No relevant side effects were observed with the subjects who had undergone with sublingual immunotherapy with Canova. However, there was a significant reduction of clinical scores for nasal obstruction, sneezing, nasal itching and cough in this group in comparison to those subjects who were on SIT.

Conclusion: Canova a homeopathic formulation could be used as an adjuvant with allergen immunotherapy for the treatment of allergic rhinitis, allergic asthma etc. Administration of SIT together with an immunomodulator (Canova) may facilitate safe and effective immunotherapy.

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Treatment of patients with seasonal allergic rhinitis using modified allergen vaccines intranasal and sublingual immunotherapy

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Background: Type I allergy represents an important health problem that is currently affecting approximately 25% of the population in Western countries. SIT, the only causative treatment of Type I allergy, is currently performed with crude allergen extracts. (Niederberger V, Valenta R. Exp.RevVaccines.2006Feb;5(1):103-10.)

With the aim evaluate the efficacy of SIT in patients with SAR we selected 25 patients with ragweed sensitization. The patients were divided into two groups. 10 patients of 1 group were treated with modified SIT by Na-DNA salt and 15 patients of the 2 group - SIT with the same allergen and placebo (dilutant).

Methods: Microscopy of nasal smears, ELISA, blood tests, skin prick test, intradermal tests, immunologic examinations and tests: (CD3,CD4,CD8,

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Parameter	Result relative group 1 value M±m	Result absolute group 1 value M±m	Result relative group 1 value M±m	Result absolute group 1 value M±m
CD3	57.5±3.2	1.01±0.13•109	56.6±2.49	0.94±0,17•109
CD4	34.3±1.55	0.52±0.05•109	33.7±1.1	0.57±0.02•109
CD8	20.4±0.1*	0.35±0.01•109	18.6±0.2*	0.32±0.03•109
CD16	5.2±3.41*	0.14±0.12•109	4.47±1.15*	0.08±0.06•109
CD3, CD25	3.6±0.85	0.06±0.02•109	2.7±0.72	0.04±0.01•109
CD3, CD95	5.2±1.2	0.08±0.03•109	7.2±0.9	0.09±0.02•109
CD3, HLA-DR	3.7±1.1	0.06±0.07•109	6.2±1.7	0.10±0.03•109
CD20	8.9±3.8*	0.15±0.15•109*	13.1±2.7*	0.17±0.10•109*
IgA		1.5±0,12		1.03±0.15
IgM		1±0,2		0.91±0.1
IgG		11.6±0,04*		8.26±0.34*
IgE total		346.32±97.5*		668±112.1*
NST		1.93±0.24*		1.8±0.31
Th1		7.10±0.09*		5.06±1.01*
Th2		0.8±0.19*		1.44±0.18*
CIC		74.1±15.8		87.5±10.7

*p<0,05

CD16,CD20,CD3;CD95,CD3; HLADR,NST,IgA,IgMIgG,IgE,Th1,Th2). All groups were treated with combinations of sublingual and intranasal mucosal SIT using allergens dilutions beginning from 1:1,000,000 then 1:000,000 then 1:10,000 in doses of 0.1-0.2-0.4-0.8ml daily then 1:1000 given at the same doses 2 times per week; followed by 1:000 and 1:10 in doses of 0.1-0.2-0.3-0.4-0.5-0.6-0.7-0.8-0.9ml one a week until a dose of 0.1 of pure allergen was reached as the maintain dose given one a month.

Results: We stopped treatment for the period of seasonal manifestations and examined patients again. After comparing results in turned out that the patients overcame the therapy better in the 1 group. The cases of nasal congestion and ocular irritation during treatment were in the 1 group in 0.1% and in 1.2% of patients in the 2 group. No cases of systemic reactions were fixed. The average number of eosinophilic cells in nasal smears in the period of seasonal manifestations was less in the patients of the 1 group (68% in the 1 group and 95% in the 2 one). Patients of the 1 group noted that severity of symptoms and need in additional medication was less then during previous years and patients in the second group did not noticed major changes. The immune system tests showed changes towards Th1 response.

Conclusion: We can speculate that these positive changes in immune system of the patients of the 1 group could be the response of lymphoid tissue to the modified allergen stimulation during SIT and the anti-inflammatory effect that lead to better overcoming of the treatment. This shows the benefit of using of modified allergen SIT with Na-DNA salt.

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Metanalisis of the efficacy of sublingual immunotherapy in asthma in pediatric patients, 3 to 18 years of age

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Background: Recent studies have documented the efficacy and safety of sublingual immunotherapy (SLIT) in patients with rhinitis, but the value of this treatment in asthma is still debated. We evaluated the efficacy of SLIT in the treatment of allergic asthma in children, by a meta-analysis of randomized, double-blind and placebo-controlled (DBPC) clinical trials.

Methods: Electronic databases were searched up to May 31st, 2006, for randomized, DBPC trials assessing SLIT in pediatric asthma. Effects on primary outcomes (symptom scores and concomitant use of rescue medication) were calculated with standardized mean differences (SMD) using a random-effect model. RevMan 4.2.8 program was used to perform the metaanalysis and we followed the recommendations of the Cochrane Collaboration and QUORUM statement.

Results: Seventy three articles were identified and reviewed. Nine studies, all published after 1990, fulfilled the selection criteria. 441 patients had a final assessment and were included in the analysis. Two hundred thirty two patients received SLIT and 209 placebo. The results of the present analysis demonstrated a relevant heterogeneity due to widely differing scoring systems. Overall, there was a significant reduction in both symptoms (SMD – 1.14, 95%CI – 2.10 to – 0.18; p = 0.02) and medication use (SMD – 1.63, 95%CI – 2.83 to – 0.44; p = 0.007) following SLIT.

Conclusion: SLIT with standardized extracts reduces both symptom scores and rescue medication use in children with allergic asthma compared to placebo.

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Local nasal immunotherapy in allergic rhinitis

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Background: To assess the clinical efficacy & safety of local nasal immunotherapy &comparing it with other routes of immunotherapy in allergic rhinitis patients.

Methodology: 1000 patients from 6 egyptian governorates were included in this study.

Results: Skin tests showed significant improvement after local nasal immunotherapy as well as blood, nasal& sputum eosinophilia were all improved also totel ige & nasal symptoms before & after local nasal immunotherapy.

Conclusion: Local nasal immunotherapy showed very good efficacy, safety & a more convenient allergen delivery in pts. With allergic rhinitis also simplified, self adminstrable, method with reduction of local nasal symptoms.

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Efficacy and safety of sublingual immunotherapy with high dose blomia tropicalis extract in children with allergic respiratory disease: comparison study with drugs

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Background: Sublingual immunotherapy (SLIT) might become a viable alternative to drug in the treatment of airway allergies. The aim of this study was to evaluate,in a pediatric population monosensitized to Blomia Tropicalis mites, the clinical efficacy and safety of high dose SLIT in comparison with drug treatment (antihistamines plus β 2 agonists).

Methods: 97 children:52 males and 45 females with a mean age of 9.4 years, range 7 to 13 years, were included in the study. Of them 49 were treated with

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Study	N. patients	Asthma symptoms score SLIT(SD)	N. patients	Asthma symptoms score PLACEBO(SD)	Weight %	SMD (95% CI)
Caffarelli	23	2.39 (0.50)	20	4.60 (0.73)	10.64	-3.51 (-4.50;-2.53)
Ippoliti	17	1.29 (0.77)	39	3.15 (0.66)	11.47	-2.57 (-3.14;-1.99)
Tari	30	6.00 (1.62)	28	9.44 (0.96)	11.25	-2.53 (-3.23;-1.03)
Niu	49	0.04 (0.01)	48	0.06 (0.02)	11.68	-1.26 (-1.70;-0.82)
Pajno	12	2.50 (0.14)	9	6.60 (4.88)	10.69	-1.24 (-2.20;-0.28)
Hirsch	11	0.17 (0.30)	10	0.42 (0.58)	10.89	-0.53 (-1.40;0.35)
Rolinck-Wernighaus	20	2.54 (5.00)	19	2.50 (5.06)	11.38	0.01 (-0.62;0.64)
Bachecilier	7	0.42 (0.45)	7	0.29 (0.26)	10.46	0.33 (-0.73;1.39)
Vourdas	33	0.11 (0.08)	29	0.05 (0.02)	11.55	0.99 (0.46;1.52)
Chi2=144.10,df=8 (p<0.00001);I2=94.4%;z=2.32(p=0.02)	232		209		100	-1.14 (-2.10;-0.18)

SLIT while 48 with the above mentioned drugs. Allergen extracts Blomia Tropicalis from STALLERGENES namely STALORAL T.M were used. Patients with severe persistent asthma, poliallergies and vernal conjunctivitis were excluded. After the 3 week-induction period, the patients were kept on a daily extract dosage of 20 drops of 300 IR/ml for another 21 weeks. The cumulative dose came to about 41654 IR. Clinical efficacy was assessed using symptom and medication scores.

Results: A gradual reduction in daily rhinitis/asthma scores was observed in SLIT group ($P < 0.001$). In only 3 patients,out of 49, it was necessary to employ antihistamines and in 7 out of 49 salbutamol. Also the symptoms score was statistically lower ($P < 0.01$). The clinical efficacy was higher in patients with mild/moderate persistent asthma. No patient showed local or systemic side effects of any relevance. Local adverse reactions were oral pruritus, gingivitis and aphthas.

Conclusion: In this study on patients with single sensitization to Blomia Tropicalis allergen suffering from allergic respiratory disease, the SLIT improves the allergic symptoms to a greater extent than drug treatment. SLIT even at ultrahigh doses was safe and well tolerated.

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Immunological characterization of recombinant lactobacillus plantarum expressing Bt5 major allergen protein

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Background: Sensitivity to Blomia tropicalis (Bt) accounts for more than 50% of the incidence of mite allergy in the tropical regions and Bt5 is a major Bt allergen. In recent years, numerous studies have suggested that recombinant probiotics could be effective in primary prevention and management of allergic diseases. The long term objective of this study is to develop recombinant lactobacilli expressing dust mite allergen for clinical application.

Methods: Bt5 gene was inserted into an expression vector pSIP412 containing a strong inducible promoter $P_{orf\chi}$. Lactobacillus plantarum was electrotransformed with pSIP412 vector carrying Bt5 gene and designated as Lp-Bt5. Murine bone marrow-derived dendritic cells (BM-DCs) were pulsed with heat-killed Lp-Bt5 overnight. The cell surface molecule expression and cytokine production of the pulsed-DCs were examined by flow cytometry and ELISA. The antigen presenting function of the pulsed BM-DCs were analysed by co-culturing with Bt5-specific Th2 cell lines. In vivo study of Lp-Bt5 was performed in C57BL/6 mice fed with the live Lp-Bt5 for three consecutive days per week over three weeks duration. Sera and splenic culture supernatants were collected for antibodies and cytokines analysis by ELISA.

Results: High level of recombinant Bt5 expression was achieved in Lp-Bt5 upon induction using 50ng/ml sakacin P inducing peptide. After 4 hours induction, approximately 40μg of Bt5 protein was expressed by 10^9 cfu live Lp-Bt5. Lp-Bt5-pulsed BM-DCs showed upregulation of CD86 and MHCII with production of IL-10, IL-12 and TNF-alpha. Lp-Bt5-pulsed DCs were capable of inducing cell proliferation but suppressing Th2 cytokines production by the Bt5-specific Th2 cells. In contrast the wildtype-Lp-pulsed DCs, in the presence of exogenous Bt 5 protein, induced both cell proliferation and Th2 cytokine production by the Bt5-specific Th2 cells. In vivo studies showed that the splenocytes of the mice fed with live LAB-Bt5 produced IL-5, IL-6, IL-10, INF-gamma and TNF-alpha upon stimulation with Bt5 protein. 25% of the fed mice produced detectable levels of Bt5-specific IgG1.

Conclusion: Recombinant lactobacilli expressing Bt5 allergen is potentially useful for the modulation of allergen-specific T cells. Further studies are required to assess its application in the prevention and treatment of allergic diseases.

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Role of allergen specific Immunotherapy in allergic bronchial asthma due to Sorghum Vulgare pollens

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Introduction: Immunotherapy for specific allergy is extensively used as definitive therapy for bronchial asthma and it is suppose to be only disease modifying treatment. In our area, Sorghum Vulgare crop is extensively cultivated and lot many patients are having allergic manifestation during pollination.

Aim: To find out role of sorghum Vulgare pollens in causation of bronchial asthma with reference to pulmonary function test and also Study effect of allergen specific Immunotherapy in comparison with pharmacotherapy. Methodology: Prospective, comparative, Cohort study was planned.

Sorghum Vulgare pollens are collected and analyzed. For SV pollens SDS page electrophoresis and ELISA_ inhibition assay done and 14–100 kDa proteins are separated. The SV extract was highly immune reactive and require approximately 7 ng of proteins for 50% inhibition of specific IgE binding in asthma. Immunoblot identified 8 allergenic brands of which 5 proteins (MWs 94,70, 40, 35, and 14 kDa) recognized by more than 90% of serum samples of SV pollen sensitive patients. Skin test were done with Modified skin prick test technique. Histamine diphosphate is used as positive control while buffered saline is used as negative control. Testing is performed on forearm. Immunotherapy of above extracts was given by subcutaneous route. Effect of Immunotherapy was studied for 659 patients.

Results: Short term (4mth) improvement in clinical symptoms and pulmonary function test (FEV1,PEFT,FVC,FEF25– 75) is observed. Eleven percent patient (n=4) showed improvement with immunotherapy and 86% (n=13) with Pharmacotherapy. But long term (3yrs) improvement observed in 47% (n=17) with immunotherapy and 20% with pharmacotherapy.

Conclusion: MWs 94, 70, 40, 35, and 14 kDa proteins are recognized in sorghum vulgare sensitive patients. Allergen specific immunotherapy is helpful in long term management allergic bronchial asthma due to sorghum Vulgare pollens.

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Antigenicity prediction in melittin: approach for fragment based peptide drug development from Apis dorsata

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The bee venom is used for treating a wide variety of conditions from acute tendonitis to chronic back pain to rheumatoid arthritis (RA). In this assay we found peptide nonamers from melittin protein of Apis dorsata, which can be used to select for use in rational vaccine design and to increase the understanding of roles of the immune system in allergic reactions. For development of MHC peptide binder's prediction, an elegant machine learning technique support vector machine (SVM) method has been used. Antigenic epitopes on melittin protein of A. dorsata are important determinants of protection against Rheumatoid Arthritis. As our knowledge of the immune responses to a protein antigen progressed, it became clear that the whole protein is not necessary for raising the immune response, but small peptide fragments 4-AILKVLSTGLPALIS-18 of protein called the antigenic determinants or the epitopes are sufficient for eliciting the desired immune response. We also found peptide nonamers, which are from a set of aligned peptides known to bind to a given major histocompatibility complex (MHC) molecule as the predictor of MHC-peptide binding. MHCII molecules bind peptides in similar yet different modes and alignments of MHCII-ligands were obtained to be consistent with the binding mode of the peptides to their MHC class, this means the increase in affinity of MHC binding peptides may result in enhancement of immunogenicity of melittin protein. Predictions of binding affinity antigen peptides to MHC class I & II molecules are important in vaccine development from A. dorsata.

Key words: Melittin, SVM, MHC, Secondary structure, Hydrophobicity, Hydrophilicity.

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Study of MHC nonamers and TAP activity of tertiapin from Apis mellifera

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Tertiapin is a neurotoxin from the honeybee venom. It interacts specifically with calmodulin in the presence of Ca2+. Antigenic epitopes on tertiapin protein of Apis mellifera (honey bee) is important determinants sites for protection against disorders. This assay we found peptide nonamers from tertiapin protein, which can be used to select for use in rational vaccine design and to increase the understanding of roles of the immune system in infectious diseases. For development of MHC peptide binder's prediction, an elegant machine learning technique support vector machine (SVM) method has been used. SVM has been trained on the binary input of single amino acid sequence. In this analysis, we predicted the binding affinity of tertiapin protein having 21 amino acids. We found the MHCII-IAb peptide regions 7-RIIIPHMCW, 13-MCWKKCGKK, 10-IPHMCWKKC, 11-PHMCWKKCG, (optimal score is 0.318); MHCII-IAd peptide regions 3-CNCNRIIIP, 4-NCNRIIIPH, 5-CNRIIIPHM, 6-NRIIIPHMC, (optimal score is 0.202); MHCII-IAg7 peptide regions 7-RIIIPHMCW, 9-IIPHMCWKK, 4-NCNRIIIPH, 8-IIIPHMCWK, (optimal score is 0.805); and MHCII- RT1.B peptide regions 1-ALCNCNRII, 13-MCWKKCGKK, 9-IIPHMCWKK, 11-PHMCWKKCG, (optimal score is −0.730) which represented predicted binders from tertiapin from Apis mellifera protein. These peptide nonamers are from a set of aligned peptides known to bind to a given major histocompatibility complex (MHC) molecule as the predictor of MHC-peptide binding. MHCII molecules bind peptides

in similar yet different modes and alignments of MHCII-ligands were obtained to be consistent with the binding mode of the peptides to their MHC class, this means the increase in affinity of MHC binding peptides may result in enhancement of immunogenicity of tertiapin protein. Predictions of binding affinity antigen peptides to MHC class I & II molecules are important in vaccine development from Apis mellifera.

Key words: Tertiapin, MHC, SVM, immune response.

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Systemic adverse events during rush Hymenoptera venom immunotherapy (VIT) in relation to pretreatment methods

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Background: Impact of H-receptors blockers on the safety profile during rush Hymenoptera VIT is still relevant issue.

Objective: Evaluation of the frequency of systemic adverse events during rush Hymenoptera VIT in patients pretreated either with solely H1-receptors blockers or with combination of H1 and H2 - receptor blockers.

Methods: The initial phase was conducted according to 5-day rush protocol with the use of standardized venom allergens of wasp or honeybee. All patients were pretreated with either H1-receptor blocker (30mg cetirizine) or combination of H1 and H2 - receptor blockers (30mg cetirizine + 300mg ranitidine).

Results: The study group comprised 145 patients including 112 patients receiving wasp venom and 33 - bee venom. 100 patients were pretreated with H1-receptor blockers, 45 - with the combination of H1 and H2 - receptor blockers During rush Hymenoptera VIT systemic adverse events occured in 22 patients (15,1%), more frequently in the group receiving bee than wasp venom (24,2% vs. 12,5%), however the difference was not significant. Higher percentage of systemic adverse events was observed in the subjects pretreated with both H1 - and H2-receptor antagonists (24%) than in those pretreated with H1-blocker only (11%), $p<0,05$.

Conclusion: Considerable number of systemic adverse events, especially in the group receiving H2 blockers requires particular attention during rush Hymenoptera VIT.

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Hymenoptera venom specific immunotherapy in patients with systemic mastocytosis

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Background: The indication to specific vaccination in patients with hymenoptera allergy and concomitant mastocytosis is still a matter of debate, since severe adverse reactions have been described.

Methods: We treated with immunotherapy 16 patients with systemic mastocytosis. All had positive bone marrow biopsy and elevated serum tryptase and 3 of them had also positive skin biopsy for urticaria pigmentosa. All the patients (12 male, mean age 51.8 years) have had severe adverse reactions following hymenoptera stinging (Grade IV according to Mueller). Of them, one was allergic to bee, 7 to polistes dominulus and 8 to jellow-jacket. They were vaccinated with a slow-induction protocol (7-week induction phase).

Results: Only one patient had systemic urticaria in the updosing phase, that was easily controlled slowing the escalation and giving antihistamine premedication. No adverse event was recorded in the maintenance phases with 100 mcg venom. Nine patients were re-stung while on maintenance. Five had only a mild local reaction, two were stung by a different insect without reaction and two had a grade III reaction. In these subjects the maintenance was escalated to 200 mcg and after further re-sting they had no more reaction.

Conclusion: Despite the relative rarity of systemic mastocytosis, hymenoptera venom hypersensitivity can provoke life threatening reactions in those patients. Based on our experience, specific immunotherapy with a slow induction is safe and can lead to an relevant, although not complete, clinical improvement

Key words: Systemic mastocytosis, hymenoptera allergy, specific immunotherapy

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A modified latex hevein (Hev b 6.02) retains lymphocyte proliferation response

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Rationale: We designed a hypoallergenic Hev b 6.02 peptide by changing all cysteine residues to serine residues with the goal to produce a standardized specific immunotherapy reagent.

Methods: Sera of 24 latex allergic patients with confirmed IgE reactivity to MBP-rHev b 6.02 were tested for their IgE binding capacity to the modified hevein peptide by ELISA. BALB/c mice were vaccinated either with this modified Hev b 6.02 peptide alone or coupled to tetanus toxoid. The antibody responses of these mice to natural Hev b 6.02 were analyzed by immunoblot and to MBP-rHev b 6.02 by ELISA. Cytokine production was tested in spleen cell cultures of immunized BALB/c mice.

Results: The Hev b 6.02 peptide showed markedly decreased IgE binding (9/24) of latex allergic patients' sera. Immunization with each of the modified Hev b 6.02 peptide formulations induced Abs that cross-reacted with the natural and the recombinant Hev b 6.02. Uncoupled Hev b 6.02 peptide induced lymphocyte proliferation and Th1 (IL-2, INF-γ) cytokine production in spleen cell cultures indicating that this peptide was able to induce a Th1 biased T-cell response. Tetanus toxoid coupled modified Hev b 6.02 peptide revealed high titers of IgG, mainly of the IgG2a subclass, to MBP-rHev b 6.02.

Conclusion: The ablated allergenicity but retained lymphozyte proliferation capacity of the Hev b 6.02 peptide showed that such a reagent can induce a Th1-biased immune response that is also directed against the major latex allergen Hev b 6.02.

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Clinical efficacy and CXCR3 - CCR3 modulation by parietaria immunotherapy

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Background: It is well known that allergen immunotherapy modifies the peripheral immunoglobulin profile down-regulating the Th2 cytokines. Notably CCR5 and CXCR3 are found on Th1 cells, CCR3 and CXCR4 on Th2 cells. Aim of the present study was to identify an immunological marker that correlates with the clinical effects among CXCR3, CCR3, CXCR4, CCR5 expression on PBMC.

Materials and Methods: 18 patients were enrolled according to the following criteria: moderate/severe rhinoconjunctivitis, moderate asthma since at least 2 years, sensitization to Parietaria only, age between 18 and 55 years, no previous immunotherapy. After a one-week run-in period where an optimal asthma therapy was prescribed, patients were received either SLIT to parietaria (SLIT-ONE Alk-abellò) or drug treatment for rhinitis. Control visits were carried out at 1, 2, 6, 10 and 16 weeks (follow-up visit). Quality of life was evaluated with a validated questionnaire (Rhinasthma).

The immunological parameters assessed were: PBMC proliferation with specific allergen Par-j1, detection of CD4, CD8, CD3 and chemokines receptors CCR3, CXCR3, CXCR4, CCR5 before and after stimulation with Par-j1

Results: The symptom scores decreased in the control group since the first week of study whereas in the SLIT group a significant improvement versus baseline was seen after 6 weeks of treatment. Starting from 6 weeks there was no more difference in total symptom scores between the two groups. Quality of life significantly improved in the SLIT group if compared with controls from week 6. After 6 weeks SLIT group significantly decreased in drug doses for asthma treatment whereas no statistical difference in therapy dosage was seen in the control group until week 6 the SLIT group had a significant higher use of rescue medication if compared with control group after week 6 the uptake of rescue medications had an inverse trend with statistically significant higher intake in the control group. In the same period there was a decrease in CCR3 and an increase of CXCR3 in treated group these variations became significant at the end of the study from the baseline. No difference in CD4, CD8, CD3, CXCR4, CCR5 expression.

Conclusions: This study demonstrates efficacy SLIT when compared to conventional therapy for persistent asthma and rhinitis, correlations between clinical scores and both CXCR3 and CCR3 seen after 6 weeks of immunotherapy in treated patients demonstrate an immunological shift towards Th1 lymphocyte pattern.

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Prediction of antigenic MHC binders of neurotoxin M14 protein from Buthus eupeus

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The new paradigm in vaccine design is emerging, following essential discoveries in immunology and development of new major histocompatibility complex (MHC) Class-I binding peptides prediction tools. In this we analyzed MHC binders of neurotoxin M14 protein from Buthus eupeus [Lesser Asian scorpion] are important determinant of protection against cardiovascular disorder. Buthus venom was tested in vivo in anaesthetized rats and in vitro on isolated cardiac and skeletal muscle preparations. In vivo, the venom caused marked rhythmical fluctuations in blood pressure preceding cardiovascular collapse and death. Scorpion toxins constitute a family of homologous proteins that exert potent pharmacological effects on potassium or sodium ion channels. In this analysis, we predicted the binding affinity of B. eupeus protein neurotoxin M14 having 66 amino acids. We found the fragments peptide regions 8-DDRNCVYTCALNPYCDS-24 of protein called the antigenic determinant or the epitope is sufficient for eliciting the desired immune response. The predicted binding affinity is normalized by the 1% fractil. The MHC peptide binding is predicted using neural networks trained on C terminals of known epitopes. In analysis predicted MHC/peptide binding is a log-transformed value related to the IC50 values in nM units. Predicted MHC binding regions acts like red flags for antigen specific and generate immune response against the neurotoxin M14 antigen. So a small fragment of antigen can induce immune response against whole antigen. This theme is implemented in designing subunit and synthetic peptide vaccines. The sequence analysis method is allows potential drug targets to identify active sites against venom action. The method integrates prediction of peptide MHC class I binding; proteosomal C terminal cleavage and TAP transport efficiency.

Key words: Neurotoxin-M14, MHC, TAP, hydrophobicity, hydrophilicity

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Birch pollen allergic patients undergoing allergen specific immunotherapy also respond to minor allergen Bet v 2

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Background: Levels of allergen specific IgE are relatively unaffected by allergen specific immunotherapy (SIT), whereas levels of allergen specific IgG is markedly increased. The effect on antibodies to minor as compared to major allergens is not clear. In this study antibodies towards Bet v 1 and Bet v 2 of *Betula verrucosa* were investigated in sera of SIT treated patients.

Methods: Specific IgE was bound to monoclonal anti-IgE coupled to paramagnetic beads and measured by the binding of ^{125}I labelled purified natural Bet v 2 and recombinant Bet v 1.2801, respectively. IgE depleted serum was tested for specific IgG and following incubation with the labelled allergen, free and bound allergen were separated by protein G affinity chromatography. Sera were tested in immunoblotting with or without absorption using 1 mg birch pollen extract per serum sample.

Results: All sera (n=50) were positive for IgG and IgE against Bet v 1 before and after SIT. The relative binding activities of IgG compared to IgE were up to 30 times higher after SIT. In contrast, 22% of non-treated patients were positive for IgE anti-Bet v 2 and 48% for IgG anti-Bet v 2. At 1.5 years of SIT, the prevalence of IgG- and IgE-anti-Bet v 2 was unchanged, however, after 5 years of treatment, the prevalence of IgG anti-Bet v 2 was 91% but the prevalence of IgE anti-Bet v 2 was unchanged when analysed by quantitative immunoassay. Contrary to this observation, 3 sera became IgE anti Bet v 2 positive after 5 years of treatment when measured by immunoblotting.

Conclusion: The dominant binding activity against Bet v 1 and Bet v 2 in sera from birch pollen allergic patients both before and after SIT resides in the IgG fraction. Prolonged SIT treatment induced IgG against minor allergen Bet v 2 in most individuals. New IgE reactivities to Bet v 2 were observed after SIT in 3 patients by immunoblotting, however, this result was not confirmed by quantitative solid phase immunoassay and may possibly indicate differences in assay sensitivity with respect to denaturation insensitive epitopes.

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Development and characterization of allergen-specific monoclonal antibodies and their inhibitory effects on allergic patients' IgE binding to the major birch pollen allergen Bet v 1

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The induction of allergen-specific antibodies which inhibit the IgE recognition of allergens is a major mechanism underlying allergen-specific immunotherapy. In order to develop therapeutic antibodies for the treatment of allergy to birch and related allergen sources, we immunized mice with surface-exposed peptides derived from the major birch pollen allergen Bet v 1 and established 14 hybridomas secreting peptide-specific mouse monoclonal IgG1 antibodies. When tested for interference with allergic patients' IgE by ELISA competition experiments, Bet v 1-specific monoclonal antibodies inhibited polyclonal serum IgE reactivity of allergic patients (n=44) to Bet v 1 and Bet v 1-homologous allergens between 60–70%. After humanization, the described Bet v 1-specific monoclonal antibodies may be used for passive vaccination or local therapy of birch pollen allergy.

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Differences in specific cutaneous sensitivity between allergic patients treated with dermatophagoides pteronyssinus depigmented and glutaraldehyde polymerised extracts, non-chemically modifed, and treated only with pharmacotherapy

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Background: The evolution of skin sensitivity is one of the objective tolols to evaluate the efficacy of specific immunotherapy.

Objective: To compare the evolution of the cutaneous sensitivity of allergic patients with rhinoconjunctivitis with or without asthma due to hypersensitivity against D. pteronyssinus, treated with vaccines containing standardized extracts of this mite (alum adsorbed depigmented and polymeryzed -modified- or non-modified extracts -retard-) or only with pharmacological treatment.

Materials and Methods: It was conducted an open and controlled clinical trial with 3 groups of patients. These patients received at random the modified (n=21), the retard (n=21) or only pharmacological treatment (n=7). After an initial wash-out period, and before receiving inmunotherapy, there were skin prick testeds in duplicate, on the volar surface of the forearm using the same batch of native D. pteronyssinus extract at 0.002, 0.02, 0.2 and 2 mg/ml. The vial of maximum concentration had a potency of 100 HEP/ml and contained 40.70 mg of Der p 1/ml and 17.22 of Der p 2. These tests were repeated after 6 and 12 months of treatment (end of study). The results were expressed as the 10 HEP value (quantity of allergen extract that produces a wheal of the same size as the wheal produced by histamine HCl 10mg/ml).

Results: The patients treated with the modified preparation needed to obtain the 10 HEP value 0.233 mg at baseline, 0.508 mg after 6 months of treatment, and 1.1 mg at the end of the study (P=0.008). In the group of patients treated with the retard preparation the values were 0.328, 0.436, 0.880 mg, respectively (p=0.037). The control group experienced an increase on the cutaneous sensitivity (0.220, 0.188 y 0.115 mg, respectively), however the difference was not significant (P=0.513).

Conclusion: This open and controlled clinical study demonstrates that the patients sensitized against D. pteronyssinus and treated with specific inmunotherapy experienced a highly significant reduction on cutaneous sensitivity, being the group treated with the vaccine containing the modified extract the first in presenting improvement (after 6 months of treatment), whereas the patients treated with the retard experienced improvement after 12 months. The patients not treated with inmunotherapy did not show reduction on the specific cutaneous sensitivity.

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Safety evaluation of a fast build-up schedule of immunotherapy using therapeutic vaccines containing depigmented and glutaraldehyde polymerised allergen extracts

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Background: Various administration schedules of immunotherapy have been reported to be beneficial for the treatment of inhalant allergy: conventional, cluster and rush. The use of therapeutic vaccines containing modified allergen extracts (depigmented and glutaraldehyde polymerised), in which the allergenicity is decreased, and the immunogenicity maintained, allows to reach the maintenance dose using conventional schedules with few administrations in a very short period of time. The objective was to evaluate the safety of a very short build-up phase using these hypoallergenic vaccines containing standardised modified allergen extracts.

Materials and Methods: One hundred and nine patients (56 female and 53 male, mean age 29.7 years) with rhinoconjunctivitis and/or asthma, sensitised to mites and/or pollens were included in an observational cohort study. All of them received an individualised therapeutic vaccine containing depigmented and glutaraldehyde polymerised allergen extract. The patients reached the maximum dose after 2 injections of the vial of maximum concentration (0.2 as the first dose the first day and 0.5 ml as the maintenance dose after 1 week, followed by 0.5 ml after 1 month). The tolerance was assessed by recording all side reactions related to immunotherapy, which were classified according to the criteria of the EAACI.

Results: The total number of injections was 327. All patients reached the maximum recommended dose the first week. No premedication with drugs was used before the administration of the immunotherapy. Eight reactions (2.44 % of the total injections) were recorded, 7 local and 1 systemic. The 7 local reactions consisted of 3 immediate (diameter > 5 cm) and 4 delayed (diameter > 10 cm). One systemic reaction of grade 2 was reported. This reaction consisted in a slight urticaria that resolved with treatment with antihistamines (ebastine).

Conclusion: The therapeutic vaccines containing chemically modified extracts can be safely administered using a fast schedule in an immunotherapy unit, reaching the maximum dose the first week with 2 injections and without the need of premedication and/or hospitalisation.

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The influence of different application forms of allergen immunotherapy on the level of allergen specific IgG antibodies in patients with grass pollen allergy

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Background: Allergen immunotherapy (AIT) is the only therapeutic approach in the treatment of IgE mediated allergies which is targeting immunopathogenetic process. Allergen could be administered in several application forms preferentially subcutaneously or sublingually. It is difficult task to follow the efficacy of this immunomodulation. The aim of study is comparison of influence of two different routes of application of immunotherapy on the serum level of allergen - specific IgG and IgG4 antibodies.

Methods: Together 62 patients (31 male, 31 female) with grass pollen allergy were enrolled. Allergen immunotherapy by subcutaneous injection (Phostal, Stallergenes) was administered to 21 patients. Allergen immunotherapy in the form of sublingual capsules (Staloral, Stallergenes) was administered to 18 patients. 23 patients were treated symptomatically during pollen season. The blood samples were collected before the start of therapy (January, 2003), and after 12, 24 and 36 months of treatment. The serum level of specific IgG and

IgG4 antibodies against immunodominant allergen (timothy grass) was evaluated by diagnostic kit (Dr.Fooke, Germany).
Results: No significant differences were found between groups of patients before start of allergen immunotherapy in any parameter tested. There is statistically significant increase in allergen specific IgG in both groups of patients after one year therapy (both $p<0.0001$) and after 3 years of therapy (both $p<0.0001$). Statistically significant increase was found in allergen specific IgG4 in both groups of patients after 3 years of therapy (subcutaneous group $p=0.0007$, sublingual group $p=0.042$). There is statistically significantly higher level of allergen specific IgG and IgG4 (both $p<0.0001$) after 3 years of therapy in patients treated by subcutaneous form of allergen immunotherapy in comparison with patients treated by sublingual form of immunotherapy. The level of allergen specific IgG and IgG4 in control group remains without any significant change.
Conclusion: The subcutaneous application of allergen immunotherapy results in strong increase of allergen specific IgG and IgG4 antibodies in comparison with sublingual administration of immunotherapy.

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Two different application forms of grass pollen immunotherapy - the changes in the levels of cytokines IL-4 and IFNγ after 36 months of immunotherapy

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Background: Allergen immunotherapy (AIT) is the only therapeutic approach in the treatment of IgE mediated allergies which is able to target immunopathogenetic process. Allergen can be administered in several application forms preferentially by subcutaneous or sublingual route. Subcutaneous and sublingual forms are differentially interacting with immune system. Data concerning the comparison of these two forms of application favour studies with clinical efficacy over laboratory studies.
Aim: Of this study is the comparison of the influence of two different types of AIT on the serum levels of cytokines IL-4 and IFNγ.
Methods: Together 62 patients with grass pollen allergy were enrolled. Subcutaneous AIT (Phostal) was administered to 21 patients. Sublingual AIT (Staloral, 100 IR/ml)) was administered to 18 patients, 23 patients were treated symptomatically. Blood samples were collected before the start of study (January, 2003), after 12, 24 and 36 months. The serum level of cytokines IL-4 and IFNγ was evaluated by diagnostic kit (Quantikine, RDS).
Results: No significant differences were found between the groups before the study. There was significant increase of IFNγ only in the group of subcutaneous AIT after 12 months of therapy ($p= 0.016$) and after 36 months ($p= 0.0294$). The IL-4 demonstrated significant decrease in the group of subcutaneous AIT after 12 months of therapy ($p= 0.022$) and in both groups of AIT after 36 months ($p< 0.0001$), but comparing to control group only subcutaneous AIT group reached significance.
Conclusion: The subcutaneous and sublingual applications of AIT after 36 months resulted in the significant decrease of IL-4 in both groups, but decrease of IL-4 in the subcutaneous group was manifested earlier - after 12 months. Expecting increase of IFN γ was significant only in group of subcutaneous AIT. Immunomodulation by subcutaneous AIT seems stronger than sublingual AIT, especially in the first year of the study. For continual sublingual AIT are needed higher concentrations of allergens then 100 IR/ml as maintenance dosage.

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Double blind placebo controlled trial of sublingual monomeric allergoid in grass pollen allergy

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Background: Double blind placebo controlled trials of allergy vaccines are considered the better method to confirm his efficacy. An assay of a sublingual monomeric allergoid in tablets was done in patients allergic to grass pollens during 2 years. This monomeric allergoid was obtained by carbamylation of amino groups (lysine) of allergenic proteins.
Methods: 33 rhinitic patients allergic to grass pollens as confirmed by clinical history, skin prick tests and specific IgE were included in the trial. Patients were randomized between placebo and active vaccine both given as sublingual tablets administered by the sublingual swallow method (LAIS, Lofarma, Milano, Italy). A traditional posological scheme was used starting with a very low dose increased progressively in the course of 14 weeks before grass pollen season. Enrolment of the patients was preseasonal followed by a seasonal visit in May and a end of the year visit in October. Patients were followed during 2 consecutive years. Nasal provocation tests with the allergens included in the vaccine was done before treatment and after 1 and 2 years of treatment and symptom scores registered after each challenge. The use of nasal steroids as rescue was authorized for rhinorrea, sneezing and conjunctivitis. Symptom scores for eventual side effects and pharmaceutical scores were registered for each patient. Informed consent was obtained from all the patients.
Results: Results were evaluated after 1 and 2 years of treatment. Between vaccines and placebo the score for rhinorrea ($p<0,03$) sneezing ($p<0,03$) and conjunctivitis ($p<0,02$) was significantly decreased for the second wear of treatment. Between the first and second years of treatment the scores for placebo were not changed but for active vaccine, rhinorrea ($p<0,03$), sneezing ($p<0,03$) and conjunctivitis ($p<0,02$) were significantly decreased. Symptomatic score after nasal challenge was decreased ($p<0.03$) after 2 years treatment. Nasal steroid consumption were significantly decreased in the active group during May and June both years of treatment ($p<0,02$). Only two light local adverse events were reported in the active group and none with placebo.
Conclusion: Preseasonal treatment with a sublingual monomeric grass pollen allergoid in tablets improved rhinorrea, sneezing and conjunctivitis and reduced the steroid consumption. Tolerability was optimal.

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Phleum pratense alone is sufficient for allergen specific immunotherapy against allergy to Pooideae grass pollens

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Background: The grass family Poales contains more than 10.000 species which can be divided into subfamilies based on their taxonomy. One of the larger subfamilies is the Pooideae subfamily also denominated as the temperature pasture grasses. In spite of the large number of grass species, grass pollen allergic patients' IgE show a high degree of cross-reactivity between Pooideae grass species. Currently, immunotherapy for grass pollen allergy is often performed with mixtures of grass species. However, high clinical efficacy of grass pollen immunotherapy (SIT) using *Phleum pratense* (Phl p) alone has been demonstrated in several studies. The aim of the study was to investigate the antibody cross-reactivity towards Phl p and grass pollen extract mixes.
Methods: The amount sIgE reacting with Phl p pollen extract and four different grass pollen extract mixes was determined in sera from grass pollen allergic patients. The capacity of Phl p to inhibit the reaction of grass pollen allergic patients' IgE towards the four grass pollen extract mixes was performed using excess inhibitory concentration of non-labeled Phl p extract. The four mixes each comprised between 5 and 10 Pooideae grass pollen extract species. Phl p SIT induced IgG4 was measured to Phl p pollen extract, extracts of nine individual Pooideae grass species and the four grass pollen extract mixes. All allergen specific antibody assays was performed using an ADVIA Centaur solid phase immunoassay.

Results: The correlation between the reaction of IgE towards Phl p and the four grass pollen extract mixes, irrespective of the number included in the mixes, is statistical significant (0.96–0.98, Spearman). Phl p has the capacity to inhibit the reaction of the patients specific IgE to the four grass pollen extract mixes indicating a high degree of cross-reactivity of the patients IgE. SIT with Phleum pratense induces IgG4 which exhibit a statistical significant correlation (0.92–0.99, Spearman) in their reaction with the individual nine Pooideae grass pollen extracts and the four grass pollen extract mixes. Thus, IgG4 induced by SIT with Phl p cross-react extensively with both single and mixed grass pollen extracts.

Conclusion: Immunochemical and clinical data together support the conclusion that Phl p alone is as effective for allergen specific immunotherapy against allergy to Pooideae grass pollens as any mix or single specie fro the Pooideae family.

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Safety of subcutaneous immunotherapy for allergic respiratory diseases: a prospective study

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Background: The clinical efficacy of subcutaneous immunotherapy (SIT) in the treatment of allergic respiratory diseases is well documented, but its main drawback of is the risk of systemic reactions (SRs). This 3-years prospective study evaluated the safety of SIT with standardized extracts in real life in patients with allergic rhinitis and/or asthma.

Methods: Between January 2003 and June 2006 a total of 248 patients (140 male, mean age 31.4 years, range 7–65) referred to our Allergy Unit were prescribed SIT for their respiratory diseases. 176 of the patients had seasonal/perennial rhinitis and 72 asthma with or without rhinitis. Allergen extracts were all standardized by biological methods (ALK-Abellò and Stallergenes). 204 patients were treated according to a traditional (12 weeks) schedule, whereas 44 underwent a cluster protocol (7 weeks). Patients were treated for grass (35%), birch (13%), parietaria (31%), mite (21%). Systemic reactions (SRs) were categorized into immediate SRs (occurring within 30 minutes) and late (>30 minutes). They were graded in four levels of severity according to EEACI Immunotherapy Position Paper (0= No symptoms; 1= Non specific symptoms probably non IgE-mediated; 2= rhinitis/asthma responding adequately to treatment; 3= non life-threatening SRs, e.g. urticaria, angioedema or severe asthma; 4= Anaphylactic shock).

Results: A total of 9,650 injections were administered in the 248 patients. SRs registered are shown in table 1 and divided according to the two phases of the schedule (build-up and maintenance). Results. There were 18 SRs (7.3% patients and 0.20% injections). 13 occurred in the build-up phase and 5 in thye maintenance. SRs were more frequent in females (66%). Grass pollens were most frequently responsible of SRs (65%) than other allergens (parietaria 18%, tree 5%, mite 10%) SRs were Grade 1= 8 cases; Grade 2 = 7 cases; Grade 3 = 1 case, Grade 4 = 2 cases (Epinephrine administered). Both severe reactions occurred within 30 minutes after injection. Late reactions were less common (7 SRs).

Conclusion: If properly used, SIT has a low and acceptable rate of side effects. In our population female gender and build-up phase were the two main risk factors. Keywords. Specific immunotherapy, injection, systemic reaction.

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Adjuvant effects of heat-killed wildtype Lactobacillus casei on allergen immunotherapy in an allergy mouse model

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Background and Aim: Probiotics, like lactobacilli have been used as a management strategy for allergy diseases. Its effects on specific immunotherapy have been previously studied in humans. The aim was to evaluate the possible adjuvant effects of probiotics on allergen-specific immunotherapy in an allergy mouse model.

Methods: C57BL/6 mice were pre-sensitized by epicutaneous patching with recombinant Der p 2, and subsequently orally fed with either heat-killed wildtype *L. casei* (Lc) $NaHCO_3$ buffer for five weeks (n=6 per group). All mice then received two subcutaneous (s.c.) immunizations of Der p 2 to mimic allergen immunotherapy, followed by aerosol challenge with Der p 2 a week later.

Results: The Lc fed Der p 2 sensitized mice showed significantly lower Der p 2 specific-IgE and IgG1 after s.c. immunizations and airway challenge compared to the $NaHCO_3$ buffer control group. Spleen T-cells and mesenteric lymph node (MLN) cells of Lc fed mice showed suppression of Th-2 (IL-5, IL-13, IL-10, IL-4) and pro-inflammatory (TNF-α cytokines), with an increase in TGF-β production in MLN cells. In addition, these Lc-fed mice but not the controls showed attenuated lung inflammation as demonstrated by the significantly reduced lung histopathology and total cell count in BALF. These results suggest that combined administration of the heat-killed L casei and allergen could effectively down-regulate the pre-existing Th-2 allergic responses.

Conclusion: Probiotics such as *L casei* have the intrinsic adjuvancity and immunomodulatory properties that could potentially be exploited to improve the efficacy of allergen-specific immunotherapy for allergic respiratory diseases.

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Vaccinations with recombinant ProDer p 1 produced in E.coli prevent the development of house dust mite allergy

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Our recent data indicated that recombinant ProDer p 1, an enzymatically inactive Der p 1 precursor form, produced in *E.coli* displayed very low IgE binding capacity but retained its T-cell reactivity. To confirm this hypoallergenic character, the present study investigated the prophylactic potential of ProDer p 1 produced in *E.coli* in a Der p 1 sensitization murine model.

Gel fitration chromatography as well as FTIR spectroscopy demonstrated that ProDer p 1 is produced in *E.coli* as aggregates containing an higher β-sheet content than ProDer p 1 produced in wild-type *P.pastoris* or in wild-type CHO cells. These conformational changes could likely explain the drastic reduction of its *in vitro* IgE binding activity towards human allergic sera. Compared with ProDer p 1 produced in CHO, vaccination of naïve mice with ProDer p 1 *coli* adjuvanted with alum induced a mixed Th1-Th2 immune response characterized by the weak production of specific IgG2a, IgG1 antibodies and the absence of the specific IgE titers. Unpredictably, the drastic reduction of specific IgE titers was maintained after mice sensitizations with natural Der p 1/alum and subsequent challenges with aerosolized house dust mite extracts. Moreover, the Th1-Th2 bias was confirmed as vaccination with ProDer p 1 *coli* induced the secretion of IFNγ and IL-5 whereas the control allergic group secreted

only IL-5. ProDer p 1 *coli* prevented the development of airway eosinophilia following house dust mite extract challenges of immunized mice. Furthermore, the increase in airways sensitivity to inhaled methacholine was reduced by the prophylactic vaccination.

Taken together, our results indicated that ProDer p 1 *coli* could represent an hypoallergen suitable for the prevention of house dust mite allergy.

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Tolerance during dose-increase phase of specific immunotherapy with standardized house-dust mite extract in asthmatic children

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Background: To determine the tolerance during dose-increase phase of specific immunotherapy (SIT) with standardized house-dust mite extract in asthmatic children.

Methods: Subcutaneous SIT with standardized house-dust mite extract was administrated to 24 asthmatic children with allergy to house-dust mite (Dermatophagoides pteronyssinus). We monitored the immediate and late side-effects after each allergen injection during dose-increase phase of immunotherapy treatment. Lung function test was done before and 30 minutes after each injection for every patient.

Results: All patients completed a total of 370 injections during dose-increase period. 27 injections (7.30% of all injections) caused immediate side-effects within less than 30 minutes after injection in 14 patiens (58.33% of all patients), in which 26 side-effects (96.3% of all early reactions) were mild local reactions (<5cm in diameter) and 1 episode of mild asthma occurred. 21 patiens(87.50% of all patients) reported 38 late side-effects(10.27% of all injections) 30 minutes after injection, 9 late side effects occurred in 7 patiens were mild systemic reactions which included asthma, allergic rhinitis, allergic conjunctivitis and allergic dermatitis, all those systemic reactions responded well to ordinary treatment. Others late side effects were 28 mild local reactions and 1 moderate local reaction (5cm in diameter). The change rate of lung function parameters after each injection showed a wide range of variance such as FEV1(−18.9%15.8%), PEF(−35.6%33.3%), FEF_{25}(−29.5%51.7%), FEF_{50}(−38.648.4%), FEF_{75}(−49.1%249.2%), MMEF(−38.2%101.7%), but there was no significant difference among all injections during dose-increase period.

Conclusion: Subcutaneous SIT with standardized house-dust mite extract showed a good tolerance in asthmatic children. A majority of local reactions and all systemic reactions were mild and responded well to ordinary treatment and adjustment of dose-increase schedule. No significant change of lung function was observed during dose-increase phase of SIT.

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House dust mite (D.farine.pteronyssinus) immunotherapy in 45 patients of perenial and steroid-dependent bronchial asthma

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45 patients of simple perenial bronchial asthma and steroid-dependent bronchial asthma were studied before and after Immunotherapy with house dust mite (*D.farine.pteronyssinus*) alleregn vaccine. The three years course of specific Immunotherapy (SIT) in 45 patients with perenial bronchial asthma and steroid- dependant asthma showed significant reduction in amount of drug intake and attacks during the treatment. The purpose of study was to investigate the long term effect on the severity and of attacks in worst months as well as the amount of drug intake (I.C.S and oral steroids) in perenial and steroid-dependent asthma even after discontinuation of dust mite immunotherapy. The inclusion criteria during the study was patients suffering from perenial bronchial asthma sensitive to house dust mite (3+/4+) by skin prick test, eosiniphilia, and specific IgE, while 10 patients were on standard control with symtomatic medications.

A total of 55 patients were followed up for three years during Immunotherapy and three years after immuntherapy, SIT treated group had significantly less intake of ICS and BD while patients without Immunotherapy were found to have attacks in perenial bronchial asthma, while in steroid dependant asthma severity of the disease was found to decrease only during immunotherapy.

The result of the study revealed that specific Immunotherapy for three years with standardised allergen extracts of house dust mite results in decrease in severityand intensity of attack in both perenial; and steroid-dependent bronchial asthma, while on discontinuation of SIT, steroid-depenent astma patients reverted back.

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Effects of immunotherapy on symptoms, PEFR and spiromery in patients with allergic asthma to house dust mite (HDM) on inhaled steroid therapy

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The present study was designed to investigate the effects of immunotherapy (IT) with an extract of Dermatophagoides pteronyssinus (Alk-Abello, Spain and Credisol, India) during a 36 months period in patients with allergic asthma to house dust mites. We included 50 patients (mean age 18 years) treated with a combination of IT and inhaled corticosteroids (ICS) in comparison to another 50 (mean age 20 years) treated with ICS alone. We evaluated symptom scores, Salbutamol use, peak expiratory flow rates (PEFR) and spirometry during 24 months of therapy with ICS and in the 12 months after ICS interruption. The two kinds of treatment were efficient and comparable in relation to symptom score, salbutamol use, morning PEFR, FVC and FEV1, but patients treated with IT and ICS had a faster improvement of symptom score and PEFR variability. The interruption of ICS after 24 months of therapy was linked to an impairment of all end points which were more pronounced in patients previously treated only with ICS. These findings suggest that in selected asthmatic patients allergic to house dust mites, the association of IT and ICS is more effective than therapy with this ICS alone and to a lower rate of relapse after the interruption of therapy with ICS.

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Antihistaminic therapy during house dust mite immunotherapy (our experience)

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The incidence of patients of allergic diseases has shown steady increase recently. The mites, especially of the family dermatophagoides

pteronyssinus are the main sources of the house dust allergens and an important etiological factor in allergic respiratory diseases. Specific allergen vaccination is an important tool to reduce allergic symptoms and prevent more severe respiratory symptoms with patients. The aim of the study is to evaluate the incidence of local and systemic reactions during the HDM immunotherapy. We investigated a group of 1200 patients allergic to dermatophagoides pt. The age and sex analyses included patients aged 15-30 showing no significant difference. We divided two groups depending on degree of the skin reactivity:

First group: skin reactivity of 2+ and 3+(no-1100) and second group: skin reactivity of 4+ (no=100). We have commenced with the subcutaneous specific immunotherapy in accord with the standard protocol. We analyzed the distribution if local and systemic reactions during the immunotherapy in both groups. Out of 27000 injections (97%) had not reaction.

810 injections caused local reaction (190 in the first group and 620 in the second group). We treated them with oral antihistaminic and local with ice. We can conclude that specific immunotherapy with house dust mites is safety. The majority of reactions were local and occurred within less than 30 minutes after inections. Premedication with antihistaminics is important especially in the beginning of the immunotherapy.

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Specific immunotherapy for house-dust mite in a patient with anisakis allergy: adverse reaction

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The nematode Anisakis simplex belongs to the Anisakidae family. The third-stage larvae are commonly found in the entrails and in the muscles of many fishes and if accidentally ingested can cause clinical disorders and sensitization in humans. Previous studies have shown cross reactivity between Anisakis simplex and different dust mite species. We observed gastrointestinal symptoms during specific immunotherapy for house dust mite in a patient with Anisakis sensitization. In a patient suffering from allergic asthma with sensitization to house dust mite developed gastrointestinal symptoms during Sublingual Immunotherapy (SLIT). Sensitization was evaluated by skin prick tests with common inhalant allergens, standard food extracts, commercial extracts of Anisakis simplex and Radioallergosorbent Test (RAST) to specific IgE antibodies. We obtained that Skin Prick Tests (STP) with common inhalant allergens were positive to graminacee (++−), cypress (++−) and house dust mite (+++−). SPT with standard food extract and commercial extract of Anisakis simplex demonstrated sensitization to Anisakis simplex. RAST showed specific IgE antibodies against allergens of Anisakis simplex. Moreover, it is important to underline that allergenic cross-reactivity between Anisakis simplex and other nematodes, such as arthropods, is well known. In a recent work, Johansson and his group underlined an association between sensitization to Anisakis simplex and four different dust mite species (Acarus siro, Lepidoglyphus destructor, Tyrophagus putrescentiae and Dermatophagoides pteronyssinus). In their study, serum samples collected from 69 subjects allergic to dust mites were analyzed for IgE to Anisakis simplex by CAP FEIA and immunoblotting inhibition. They found that 14/69 patients had detectable levels of IgE antibodies to Anisakis simplex. In the CAP FEIA inhibition, all four mite extracts inhibited, to various degrees, the IgE response to Anisakis simplex on the solid phase. In the light of these findings, it is possible to hypothesize that immunotherapy for dust-mite could pose an unintentional challenge in patients with gastroallergic anisakiasis, due to cross-reaction with anisakis simplex antigens. Even if further observations of adverse reactions to sublingual immunotherapy with dust mite in patients sensitized to Anisakis are needed, our data suggest that oral treatment should be discouraged in these patients.

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Double-blind placebo-controlled study of treatment with a depigmented and glutaraldehyde-polymerised extract of dermatophagoides pteronyssinus and D. farinae: improvement in skin sensitivity

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Background: There is a high prevalence of asthma in the Canary Islands, Spain. Most asthmatic patients are sensitised to house dust mites. Exposure to mite allergens is constant and levels regularly exceed 10 mcg of Der p 1 per gram of dust. Immunotherapy using depigmented and glutaraldehyde modified allergen vaccines produces clinical benefit, as evaluated by improvement in the shock organ, symptom and medication scores, quality of life and skin sensitivity.

Objective: To evaluate the evolution of skin sensitivity in a group of asthmatic patients sensitised to both mites in a double-blind and placebo-controlled study of 1 year of duration.

Materials and Methods: Sixty-four patients were included, and 52 finished the study (26 in the active group and 26 in placebo). The native extract of D. pteronyssinus contained 20.35 mcg of Der p 1 and 12.30 mcg of Der p 2, and the D. farinae extract contained 19 mcg of Der f 1 per mg of freeze-dried extract. Due to the polymerisation process, Der p 1, Der p 2 and Der f 1 were not detectable in the modified extract. Patients were skin tested at baseline and at the end in duplicate, on the volar surface of the forearm using the same batch of native D. pteronyssinus extract at 0.002, 0.02, 0.2 and 2 mg/ml. The vial of maximum concentration had a potency of 100 HEP/ml and contained 40.70 mg of Der p 1/ml and 17.22 of Der p 2. Glycerinated saline solution and histamine HCl 10 mg/ml were used as negative and positive controls, respectively. Skin reactions were recorded after 15 minutes. The results were expressed as the 10 HEP value. The Wilcoxon test was used to compare the results.

Results: Patients treated with the modified allergen extract received a total accumulated dose of 593 mcg of freeze-dried modified material of D. pteronyssinus and 453 mcg of D. farinae. The active group showed a significant difference from baseline (P=0.029, Wilcoxon), whereas the placebo group did not (P=0.900). The active group needed at the end of the study a median of 21.9 mcg more of allergen extract to achieve the value of 10 HEP, whereas in the placebo group the figure was −0.31.

Conclusion: This double-blind placebo-controlled trial demonstrates that the treatment of mite allergic asthmatic patients with a mixture of depigmented and glutaraldehyde-polymerised extracts of D pteronyssinus and D. farinae induces a significant decrease in the skin sensitivity.

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Specific Immunotherapy with depigmented polymerized allergen extracts of domestic mites in patients with persistent rhinoconjunctivitis

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Background: There is limited knowledge about the clinical relevance of a sensitization with domestic mites s.a. Euroglyphus maynei (Em), Lepidoglyphus destructor (Ld), Acarus siro(As) and Tyrophagus putrescentiae (Tp). The efficacy and safety of a SIT with these mites has not been described until now.

Patients and Methods: We investigated 107 patients suffering from persistent rhinoconjunctivitis by i.c test, specific IgEs and placebo controlled nasal challenge test with domestic mite allergens. If the patient had a positive i.c test and/or specific IgEs and a positive nasal challenge test SIT was initiated with the respective depigmented polymerized allergen extract. 20 patients have been treated with the following allergen extracts either alone or in combination for 6 months: 9x As, 6x Ld, 6x Tp, 4x Em. Patients symptoms and medication consumption was recorded using a standardized questionnaires.

Results: 17/20 patients (85%) improved during the course of SIT. The symptom and medication score was reduced in parallel. 1 patient did not change during SIT and 2 patients were lost to follow-up and therefore considered as treatment failure also. There were no systemic reactions and only few and mild local reactions reported.

Conclusion: This is the first report of a successful SIT with Euroglyphus maynei, Lepidoglyphus destructor, Acarus siro and Tyrophagus putrescentiae. The treatment was efficacious and safe.

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Improvement in symptom and medication score after successful treatment with a depigmented and glutaraldehyde-polymerised extract of D. pteronyssinus and D. farinae: a double-blind placebo-controlled study

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Background: There is a high prevalence of asthma in the Canary Islands, Spain. Most asthmatic patients are sensitised to house dust mites. Exposure to mite allergens is constant and levels regularly exceed 10 mcg of Der p 1 per gram of dust. Immunotherapy using glutaraldehyde modified allergen vaccines demonstrated clinical benefit in several studies. The introduction of a depigmentation step before polymerisation inactivates the enzymatic activity, removes pigments and enhances the solubility of the polymer.

Objective: To evaluate the clinical efficacy of a depigmented and polymerised vaccine containing 50% D. pteronyssinus and 50% D. farinae in patients sensitised to these mites in a double-blind and placebo-controlled trial.

Materials and Methods: Fifty-one patients were randomly allocated to either receive active treatment (n=26), or placebo (n=25). The native extract of D. pteronyssinus contained 20.35 mcg of Der p 1 and 12.30 mcg of Der p 2, and D. farinae contained 19 mcg of Der f 1 per mg of freeze-dried extract. Due to the polymerisation process, Der p 1, Der p 2 and Der f 1 were not detectable in the modified extract. Patients recorded the symptom and medication scores on a diary card. The area under the curve (AUC) and the number of days free of symptoms and/or medication were calculated for each patient. The values of AUC were compared groupwise using the Mann-Whitney test, and the Hodges-Lehmann estimator was used to measure the effect size. Fisher's exact p value was used for the number of days free of symptoms or medication.

Results: Patients treated with the modified extract received a total accumulated dose of 593.3 mcg of freeze-dried modified material of D. pteronyssinus and 452.7 mcg of D. farinae. The difference of AUC of symptom and medication scores between both groups was significant ($p<0.05$). The values of the Hodges-Lehmann were −1.6 (95%CL −18.9, −4.4) and −8.6 (95%CL −14.9, −2.2) for symptom and medication scores, respectively. Patients treated with the modified allergen had more days free of symptoms and medication (Fisher's exact p value <0.0001).

Conclusion: This double-blind placebo-controlled trial demonstrates that the treatment of mite allergic, asthmatic patients with a mixture of depigmented and polymerised extracts of D pteronyssinus and D. farinae produces a clinical benefit in symptom and medication scores, with an important effect size (zero value was not included in the 95% confidence limits).

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Clinico-immunologic study on immunotherapy with *Periplaneta americana* extract: a double blind placebo controlled trial

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Background: Cockroaches produce several potent allergens associated with sensitization and development of asthma. In highly susceptible or symptomatic patients, allergen-specific immunotherapy may be beneficial, but the data are limited. The present study is aimed to evaluate the role of immunotherapy in cockroach allergic patients using standardized whole body extract.

Methods: Twenty patients belonging to allergic asthma, rhinitis or both were recruited in active group and 20 in placebo group. The patients in active group were administered with P. americana extract subcutaneously whereas the other group received placebo injections for one year. To assess the benefits of immunotherapy, the data was analyzed for symptom/drug score, airway reactivity and immunoglobulins (IgE, IgG1 and IgG4) at the baseline and after completion of one year of immunotherapy. Immunoblots of specific IgE and IgG4 were made before and after therapy.

Results: Good improvement was observed in clinical status of patients with respect to in vivo and in vitro parameters. Changes in symptom score, airway reactivity and IgG4 values were highly significant in comparison to baseline values and placebo group. Skin test and IgE values showed non-significant reduction. Immunoblotting of specific IgG4 has shown development of more bands of increasing density after 1 year of immunotherapy. While IgE blot showed similar pattern of bands before and after immunotherapy.

Conclusion: Cockroach immunotherapy is well tolerated in patients of allergic rhinitis and asthma. IgG4 immunoblotting has shown a correlation of clinical efficacy with the immunologic response.

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At the question of the cytokines therapy of the nosocomial pneumonia

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Purpose: There were 74 patients with the hospital pneumonia of serious current under our observation. Patients were distributed to the way of introduction of the immunotroping drug. The first group (22 patients) received recombination IL-2 on 500 000 ÌÅ dose intravenously. The second group (24 patients) received hypodermic introduction of the same dose of the drug, and patients of the third group (28 person) received the combined introduction of recombination IL-2: 250 000 ÌÅ dose of inhalation with the help the ultrasonic inhaler simultaneously with hypodermic introduction of 250 000 – 500 000 ÌA dose. The course of treatment consisted of 2–3 procedures. The dynamic research of the basic clinic-laboratory indexes were given to all our patients.

Results and Conclusion: It was fixed, that all three ways of recombination IL-2 introduction provided positive dynamics of indexes of immunity on the background of basic anti-inflammatory therapy of the pneumonia as the result of our research. However, the condition of the local immunoreactivity of lungs

was recovered only at combined inhalation and hypodermic introduction (patients, contents of metabolites of oxide of nitrogen and α1-acidic glycoprotein in the condensate of the exhaled moisture was normalized, reaching similar indexes of healthy people). It is important to note, that complete regress of radiological indexes was only at combined application of recombination IL-2 inhalationly and subcutaneously. The analysis of frequency rate of recombination IL-2 introduction showed, that it was not enough single-pass introduction of the drug for the reduction of indexes of the local immunity, and rates of their reduction in the course of the repeated combined way of introduction were identical and depended on gravity of the condition and the expressiveness of the inflammation in bronchopulmonary system. The increase of frequency rate of recombination IL-2 introduction more than 2–3 times did not accelerate rates of clinical convalescence, reduction rate of immune indexes and biochemical criteria of the degree of the expressiveness of the inflammation in respiratory paths. Thus, we believe that it was efficient combined recombination IL-2 application of the 250 000 ÌÅ inhalation dose and 500 000 ÌÅ dose subcutaneously 2 – 3 times with the interval of 72 hours for treatment of patients with the serious hospital pneumonia.

DIAGNOSIS

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Discrepancies between reported symptoms, lung function measurements and PC20 in a Clinical Asthma Research Centre

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Background: Diagnosis of asthma or non-asthma is critical for clinical research. Using methacholine challenge testing (MCT), a well established bronchial challenge test for assessing airway responsiveness, we have identified groups of patients who have been incorrectly diagnosed as asthmatic or non-asthmatic.

Methods: We screened 68 volunteers participating in clinical research in asthma after they provided informed consent.

All volunteers were questioned about their general health, asthma symptoms medication and family history of asthma. Atopic status was determined with skin prick tests to a standard panel of aeroallergens. A baseline spirometry was performed prior to MCT.

MCT was conducted following a recognised dosing protocol (Crapo, R.O. et al Am J Respir Crit Care Med 2000 Vol 161 pp 309–329) using a Mefar MB3 breath activated dosimeter. Volunteers considered to be asthmatic commenced at the lowest methacholine dose of 0.03125 mg/ml and continued up the dosing protocol until a PC20 was reached. After each dose their FEV1 was obtained at 1 1/2 mins and at 2 1/2 mins post-inhalation with a subsequent 2 mins rest prior to the next dose.

Those considered to be non-asthmatic started their MCT challenge from 0.5 mg/ml dose of the methacholine and continued up-dosing until the highest dose.

The definition of asthma for the purpose of our studies was a PC20 below 8mg/ml at MCT. Non-asthma was defined as completion of the MCT at 16mg/ml without attaining a PC20.

Results: Of the 68 subjects screened for clinical research studies, 29 claimed to have a physician diagnosis of asthma, but in 8 this was not confirmed by our MCT criteria. Of these 8 subjects, 7 had normal spirometry (FEV1 >80% predicted) while 1 had FEV1 of <80% predicted.Conversely, 39 volunteers reported non-asthma. Of these 2 had symptoms at methacholine challenge testing and attained a PC20 <8mg/ml, thus fulfilling the definition of asthma according to our criteria.

Conclusion: Diagnosis of asthma and non-asthma based on existing symptoms, lung function testing and physician based diagnosis is surprisingly unreliable.

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Asthma in pregnancy: diagnostic methods

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Introduction: Asthma is the most common respiratory disorder to complicate pregnancy and represents a significant public health issue.

In many occasions, especially in the last trimester due to the pressure of unborn child, these patients are unable to perform standardized spirometry. The Impulse Oscillometry Technique (IOS) may be suitable in this application since it allows the assessment of airway obstruction by requiring minimum patient cooperation. Aim: Our aim was to identify the relationship between R5, X5, and Resonant Frequency to the 1s forced expiratory volume (FEV1) and forced vital capacity (FVC) in pregnant women.

Methods: In 42 asthmatic pregnant patients aged 18–41 yr (mean 28.5 yr) spirometry and impulse oscillometry (Erich Jaeger, Germany) was performed according to standard guidelines and manufacturers instructions. Relationships between spirometric indices and total Resistance (R5), central resistance (R20), peripheral reactance (X5) and Resonance Frequency (FRes) were obtained using correlation and regression analysis. Volumes are expressed in litres and resistance in kpa.1.s.

Results: Mean ± SD of FEV1 was 2.32±0.46, FVC was 2.93±0.64 , FEV1/FVC was 80.90±15.81 , R5 was 0.49±0.14 , X5 was −0.11±0.14 and FRes was 19.71±5.00. There was significant ($p<0.001$) correltion between the R5 and FRes, each with FEV1.R5: FEV1 = 3.14 – 1.34 R5, R^2= 0.21FRes: FEV1 = 3.90 – 3.24 fres, R^2=0.22.

Conclusion: These results suggest that Impulse Oscillometry is a valuable tool to assess airway obstruction as it is simple and requires minimal subject cooperation. This new field of oscillometry application may be particularly useful in patients who are unable to perform spirometry. Further studies are required to determine the sensitivity and specificity of this technique.

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The association between FeNO and acute exacerbation of asthma in children

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Rationale: Asthma is a Th2 cell mediated chronic inflammatory airway disease. Bronchoscopic biopsy is the best method to evaluate airway inflammation but it is too invasive for children. Measurement of exhaled nitric oxide (FeNO) is a relatively simple, noninvasive, and very reproducible test for detection of endogenous inflammatory signals in childhood. Recently measurement of FeNO level is emerging as a non-invasive tool for airway inflammation. The aim of our study was to determine the relationship between FeNO and acute asthmatic exacerbation, and to decide whether measurement of FeNO could predict acute asthma exacerbation.

Methods: Thirty eight children with mild to moderate persistent asthma aged from 3 to 15 years who presented to Kyunghee Medical Center were studied. FeNO levels were measured by chemiluminescence during exhalation into the NO analyzer. Patient's data were based on out-patient records. We compared FeNO levels between exacerbation group and non-exacerbation group.

Results: The mean FeNO level in the asthmatic children was 16.7±13.9 ppb. In the range of abnormal FeNO level (≥10 ppb), there was significant difference between asthma exacerbation group and non-exacerbation group ($p=0.004$). There was also significant correlation between FeNO level and acute asthma exacerbation ($p=0.003$).

Conclusion: Exhaled nitric oxide levels were related with acute asthmatic exacerbation in childhood asthma. Thus measurement of FeNO can be a promising clinical tool for predicting asthma exacerbation in mild to moderate persistent asthmatic children.

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The analysis of the level of bronchial hyperresponsiveness in astmatic children(2 - 18 years) by dosimetric, oscilometric and tidal breathing methods to histamine and methacholine and analysis of polymorphisms genes ADAM33 and STAT6

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Background of the study: The main pathogenic mechanism of bronchial asthma is chronic eosinophil-based inflammation of the bronchial mucosa. Airway hyperresponsiveness (AHR), is known to be a major risk factor for the development of asthma and to be equivalent of asthma severity.

Methods: Main aim of this project, supported by IGA MZ CR No. NR8383-3/2005, is to test if the level of AHR is associated with certain alleles or haplotypes of genes (ADAM 33 and STAT 6) involved in immune system activity in course of allergic illness development and airway hyperresponsiveness. 80 children with bronchial asthma had histamine and methacholine brochnoprovocation tests in a sequence with increasing levels using dosimetric method and MedicAid and DeVilbiss nebulizers. Oscilometric assessment I was followed by baseline spirometry, and with oscillometric assessment II after a decrease in FEV1> 20% versus baseline. Examinations of responsible polymorphisms of ADAM 33 and STAT 6 genes were accomplished by sequence analysis in 80 children with asthma bronchiale, their sibs and parents and 50 control healthy children.

Results: Examinations of responsible polymorphisms of ADAM 33 and STAT 6 genes were accomplished by sequence analysis in 80 children with asthma bronchiale, their sibs and parents and 50 control healthy children. In the evaluation of the result it could be demonstrated that changes in resistance R5 and R20 between oscillometric assessments I and II more than 40% are consistent with a decrease in FEV1 more than 20% in both dosimetric method. The statistical evaluation of the association of genes polymorphisms with asthma bronchiale occurrence and airway hyperresponsiveness level in children will be presented by poster.

Conclusion: These results show that resistance R5 over R20, as measured with IOS (impulse oscillometry), significantly correlates with the-gold standard-FEV1. Description of influence of particular sequence changes could contribute to additional insight to the airway hyperresponsiveness genetical background.

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Differential flow analysis of exhaled nitric oxide in asthmatic patients

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Background: The majority of asthmatic patients achieve control of their illness, but not all. It is therefore crucial to develop strategies that help the clinician monitor the disease, improving the response to treatment.

Methods: We have quantified the inflammation in central and peripheral airways by measuring exhaled nitric oxide (NO) at multiple exhalation flows in 42 asthmatic patients at different levels of severity (mild = 11; moderate stable = 10; moderate during exacerbation = 9; severe = 12, 6 of whom were receiving oral corticosteroids) and 15 healthy control subjects.

Results: Bronchial NO (bNO) in patients with mild asthma (2,283+/−346 pL/s) [mean+/−SD] was higher than in patients with moderate stable asthma (1,317+/−61 pL/s, $p< 0.001$), in patients with severe asthma receiving inhaled corticosteroids (ICS) [1,023+/−72 pL/s, $p< 0.001$], and healthy control subjects (697+/−31 pL/s, $p< 0.001$). There were no differences between bNO in patients with mild asthma compared to patients with severe asthma receiving ICS and oral corticosteroids (2,335+/−269 pL/s). Patients with exacerbations showed a higher bNO (3,521+/−373.2 pL/s, $p< 0.01$) compared to the other groups. Alveolar NO was higher in patients with severe asthma receiving oral corticosteroids (3.1+/−0.2 parts per billion [ppb], $p< 0.0001$) than in the other groups but was not significantly higher than in patients with moderate asthma during exacerbation (2.7+/−0.4 ppb). No differences were seen in NO diffusion levels between the different asthma groups. All the measurements were highly reproducible and free of day-to-day and diurnal variations.

Conclusion: Differential flow analysis of exhaled NO provides additional information about the site of inflammation in asthma and may be useful in assessing the response of peripheral inflammation to using therapy.

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Monitoring and treatment of childhood asthma in singapore: a questionnaire study in physicians

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Introduction: Asthma is the most common chronic disease in children and in Singapore over 25% of children have ever been diagnosed with asthma by age 15. Most children are seen in the primary care setting, thus it is of value to study the management practices of general practitioners and pediatricians in comparison with gold standards. The aims of the study were to investigate: a) methods of monitoring of childhood asthma, b) practices in managing acute asthma exacerbations, and c) choice of therapy in maintenance treatment.

Methods: 2100 questionnaires of 35 questions were sent by post to general practitioners and pediatricians throughout Singapore. 173 valid responses were received and results were compared to 2006 Global Initiative for Asthma (GINA) guidelines.

Results: Of respondents 76.3% were general practitioners. For monitoring childhood asthma, most physicians (89.1%) did not use symptom score cards/diaries, while 37.6% did not use peak-flow meters/spirometry. For acute treatment, 83.8% used a short-acting â-agonist, but only 41.0% used oral corticosteroids in outpatients. In the maintenance treatment of asthma, across all age-groups (infants, preschoolers, and older children) the most commonly used first line drug was uniformly an inhaled corticosteroid. However, combinations of long acting beta-agonists (LABA) and inhaled corticosteroids were uniformly the second most popular maintenance treatment. Maintenance treatment was frequently used by most doctors in treating preschoolers (62.4%) and older children (64.7%). In contrast, most doctors seldom (45.1%) or never (21.4%) applied maintenance treatment to infants. The vast majority (158; 91.3%) of doctors never used immunotherapy in asthma management, and 2.9% of respondents believed immunotherapy was not available locally.

Conclusion: Greater usage of diaries/score cards can be encouraged along with objective peak flow/spirometry measurements. Management of acute exacerbations is appropriate, but corticosteroids are under-prescribed by most doctors. LABA continues to be prescribed for maintenance despite a lack of established safety profile for infants, along with recommendations that they only be used selectively in patients that are poorly controlled by inhaled corticosteroids. Immunotherapy is not widely used and more awareness/instruction should be generated among doctors.

ASTHMA-ASSOCIATED CONDITION

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Risk of HBV reactivation in asthma or COPD patients treated with corticosteroids

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Background: Viral reactivation in hepatitis B virus (HBV) carriers has been reported to occur in association with immunosppresive agents in cancer or transplantation patients. However, effects of inhaled corticosteroids (ICS) or systemic corticosteroids (SC) in immunocompetent patients with chronic respiratory diseases are not known.

Objective: To evaluate the course of HBV infection in stable HBV carriers using ICS or SC for the treatment of asthma or chronic obstructive pulmonary disease (COPD).

Methods: HBsAg seropositive patients using ICS were reviewed retrospectively from Jan 1997 to Mar 2007. We reviewed data of 183 patients who met our inclusion and exclusion criteria including dose and duration of ICS and SC treatment, HBV DNA titer, HBeAg/Ab and AST/ALT serum level, and usage of antiviral drugs such as lamivudine.

Results: Among the 183 patients who were using ICS, 66 patients were treated with SC at least on one event. HBV reactivation occured in 5 out of 66 (7.6%) subjects who have ever used SC and 2 out of 117 (1.7%) subjects under ICS treatment alone. Among the 5 subjects with HBV reactivation who have ever used SC, 4 (80%) subjects were treated with SC chronically due to difficult-to-control asthma with more than 9000mg of total cumulative dose of prednisolone. 2 subjects with HBV reaction who were using only ICS were using moderate dose ICS.

Conclusion: Chronic use of SC and high total cumulative dose of SC are risk factors of HBV reactivation in patients with asthma and COPD. ICS treatment alone is not a risk factor of HBV reactivation. Prophylactic antiviral therapy should be considered in patients who needs chronic or high dose SC treatment.

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Gastro-esophageal reflux in allergic asthmatic patients

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Background: The prevalence of asthma is increasing in the last years. Also gastro-esophageal reflux disease (GERD) is increasing in the last years. It is known that gastro-esophageal reflux (GER) may be present and also may aggravate asthma. There are no information regarding GERD and asthma in Romania.

Aim: Our study was to evaluate the presence of GER in allergic asthma in Transylvania.

Methods: We evaluate the patients with 56 allergic asthma for the presence of GER. The pts had moderate persistent asthma treated with glucocorticoid inhalers (and/or LABA). The pts had a 24-hours oesophageal pH-monitoring. Reflux episodes identified reflux events as the percentage of time where the pH was less than four. The pts had also an upper endoscopy. The pts were also ask about there asthmatic symptoms during nights and regarding the heart burns.

Results: The mean age was 36 years (18-54 years); 32 of the pts were women. There were only 3 pts (5.7%) having heart burns. On 24-hours oesophageal pH-monitoring we found 18 pts (32%) having acid reflux during day and night. On endoscopy there were only 7 pts (12.5%) with esophagitis. There was no relationship between spirometry results on one hand, and parameters of gastroesophageal reflux on the other. The pts receive also for 4 weeks term a proton pump inhibitor (PPI) (pantoprazol 80 mg/day). The measures of symptoms (mean daily daytime and nighttime asthma symptom scores) did not change significantly during the 4 weeks treatment with PPI . There was no influence on rescue medication in the majority of the pts. Only 2 pts reported an improvement of the asthma, with lower need for rescue medication. In our Romanian patients we found no correlations between GER.
Conclusions: Our results indicate that acid in oesophagus or its short term inhibition by proton pump inhibitors, has no influence on allergic asthma.

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The clinical implication of different pathophysiologic manifestations between asthma and pure COPD

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Background: Although physiologic and anatomic differences of chronic obstructive pulmonary disease (COPD) and asthma are separated into distinct categories of disease, in clinical practice a significant proportion of patients have characteristics that are classically associated with both conditions.
Methods: To determine whether patients with fixed airflow obstruction have distinct cellular and biochemical characteristics depending on their asthmatic features, we characterized 30 consecutive outpatients presenting with fixed airflow obstruction by bronchodilator response, peak expiratory flow variability, serum total immunoglobulin E, allergen skin prick test, methacholine bronchial challenge test (MCT), and bronchoalveolar lavage (BAL). In addition, to determine significance of MCT on differentiating asthma from COPD, we exam MCT on 111 outpatients divided to asthma, pure COPD, and control group.
Results: We found that subtypes of T lymphocytes did not show significant difference in number and ratio on both groups, but neutrophils in BAL fluid was significantly lower in group with asthmatic features. Additionally, their cytokine analysis showed significantly higher level of IL-4 in asthmatic patient compared with COPD patients, while lower level of IL-5 and INF-gamma in asthma group compared with COPD group. In MCT results, area under the receiver operating characteristic (ROC) curve of sensitivity and specificity has shown that best cut-off value of PC_{20} is 15.4 mg/ml in classifying the asthma and pure COPD.
Conclusion: We conclude that despite of similar fixed airflow obstruction, subjects with asthmatic features have distinct cellular and biochemical characteristics compared with pure COPD. In addition, we suggest that the MCT using the new cut-off value could be a more useful diagnostic tool for distinguishing asthma from pure COPD. Based on these findings, we expect that this distinction of asthma from pure COPD using the different pathophysiologic manifestations could provide more proper and specific therapeutic management for the patients.

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Airway remodeling in patients with allergic bronchopulmonary aspergillosis

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Background: Allergic bronchopulmonary aspergillosis (ABPA) is a hypersensitivity reaction to Aspergillus fumigatus (Asp.f) induced by IgE and IgG antibodies which progress to central bronchiectasis (CBE) even at an early stage. We hypothesized that the airway remodeling (=irreversible change of bronchial wall) in patients with ABPA is accompanied by not only CBE but also persistent bronchial narrowing at an early stage of ABPA.
Methods: We evaluated bronchial reversibility (BR) using salbutamol and airway hyperresposiveness (AHR) using acetylcholine (Ach) in ABPA patients with CBE (=classical ABPA) and without CBE (=seropositive ABPA), and compared the BR and AHR of these patients with those of patients with allergic fungal asthma without the IgG antibody to Asp.f. Nineteen classical ABPA, 11 seropositive ABPA, and 21 allergic fungal asthmatics were participated in this study.
Results: All the subjects were positive for the IgE antibody to Asp.f. All the ABPA patients with or without CBE were positive for the IgG antibody to Asp.f. In all classical ABPA patients, but not in seropositive ABPA patients, typical CBE findings were detected in HRCT. There were no differences in age, total IgE level (RIST), and %FEV1 among the three groups. However, not only classical ABPA patients, but also seropositive ABPA patients showed significantly attenuated BR after salbutamol inhalation (median increase in FEV1: 170, 210, 340 ml, $p<0.05$). Moreover, about one-half of ABPA patients showed normal or very mild AHR, on the other hand, none of the allergic fungal asthmatics showed normal AHR.
Conclusion: These results suggest that ABPA patients, even at an early stage, may show irreversible bronchial narrowing resistant to bronchodilators or bronchostimulator inhalation.

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Follicular bronchiolitis: A rare mimicker of "severe asthma"

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Background: Atypical presentations of asthma and abnormal imaging are factors that prompt the pursuit of a lung tissue diagnosis. We present a case of follicular bronchiolitis (FB) in a patient thought to have severe asthma.
Methods: A 35 year old male non-smoker developed productive cough, dyspnea and wheezing for 2 years. He was treated with inhaled steroids, long acting beta-agonists, montelukast, antibiotics and prednisone for late onset asthma. Symptoms worsened with oral steroid taper.
Results: Pulmonary function tests revealed severe obstruction (FEV1 36%, FEV1/FVC of 86%) with significant bronchodilator response (30%) and mildly decreased diffusion capacity (68% predicted). Skin tests were positive to dust mite, mold, cats and dogs. Immunological evaluation (Table 1) revealed mild eosinophilia (557-1320 absolute eosinophils), normal T and B cells (absolute lymphocyte count 1467, CD4 1040 cells/mcL, CD8 452 cells/mcL, CD19 218 cells/mm3, CD4/CD8 2.06, CD56 143 cells/mm3), normal total complement (76 U), normal immunoglobulin levels (IgE 50, IgA 206, IgM 9) except for mild decrease in IgG1 and IgG3 (IgG 810, IgG1 387, IgG2 302, IgG3 16.4, IgG4 49.1 mg/dL), negative cystic fibrosis screen, protective titers to tetanus (0.41), and pneumococcal vaccine (protective titers of 8 out of 14 serotypes: 1,3,8,9,14,26,51,56). Further workup for infection (HIV, HTLV-1/2, AFB, yeast, bacteria, CMV), sarcoid (ACE levels, calcium and gallium scan), autoimmune disease (ANA, ANCA, MPO, Pr-3 Ab), Alpha-1 antitrypsin deficiency and hypersensitivity pneumonitis was negative. CT scan of the chest showed bilateral peribronchial thickening in central bronchi with few small non-specific lymph nodes in the mediastinum. Right upper lobe wedge resection showed acute and chronic bronchiolitis with lymphoid hyperplasia consistent with FB. He improved with chronic steroids and azithromycin (FEV1 72%, FEV1/FVC 92%).

Conclusion: FB is a rare entity consisting of abundant lymphoid follicles limited to the peribronchiolar area. It may be idiopathic or associated with hypersensitivity phenomenon or collagen vascular disease. A few cases are reported associated with immunodeficiency such as HIV or common variable immunodeficiency (CVID). Initially, he was thought to have an immune deficiency but repeat analysis revealed normal immunologic function. His disease may, therefore, be idiopathic or related to a hypersensitivity reaction not well understood. He is controlled with steroids and azithromycin.

TEST	RESULT
FEV1	32 – 51% (1.37 – 2.18 L)
FVC	47 – 62% (2.45 – 3.21 L)
FEV1/FVC	60 – 81%
Absolute eosinophil count	557 1320 cells/mcL (15–550)
Absolute lymphocyte count	2113 cells/mcL (850–3900)
CD3+/CD4+ absolute helper count	1040 cells/mcL (490 – 1740)
CD3+/CD8+ absolute suppressor count	452 cells/mcL (180 – 1170)
CD4+/CD8+ ratio	2.06 (0.865)
CD19 absolute count	218 cells/mm3 (106–220)
CD56 absolute count	143 cells/mm3 (14–402)
IgM	810 mg/dL (48–271)
IgG	359 mg/dL (694–1618)
IgG1	359 mg/dL (455.9–892.6)
IgG2	393 mg/dL (199–526.6)
IgG3	19.8 mg/dL (16.8–99.8)
IgG4	38.9 mg/dL (13.5–73.9)
IgA	206 mg/dL (81–463)
IgE	50 mg/dL (</=114)
Pneumococcal IgG serotype 1	pre -6.5, post-10.3 mcg/mL
Pneumococcal IgG serotype 3	pre -6.5, post-15.7 mcg/mL
Pneumococcal IgG serotype 4	pre -6.5, post-<0.2 mcg/mL
Pneumococcal IgG serotype 5	pre -5.6 post-0.5 mcg/mL
Pneumococcal IgG serotype 8	pre -4.7, post-8.9 mcg/mL
Pneumococcal IgG serotype 9	pre ->12.5, post-7.4 mcg/mL
Pneumococcal IgG serotype 12	pre -3.3, post-0.3 mcg/mL
Pneumococcal IgG serotype 14	pre -1.6, post->18.8 mcg/mL
Pneumococcal IgG serotype 19	pre -1.7, post-<0.2 mcg/mL
Pneumococcal IgG serotype 23	pre -6.5, post-0.5 mcg/mL
Pneumococcal IgG serotype 26	pre -6.5, post-1.4 mcg/mL
Pneumococcal IgG serotype 51	pre -6.5, post-13.8 mcg/mL
Pneumococcal IgG serotype 56	pre -6.5, post-6.4 mcg/mL
Pneumococcal IgG serotype 68	pre -6.5, post-0.7 mcg/mL
Tetanus antitoxoid Ab	0.41 IU/mL (>0.15)
Aspergillus precipitins (A. niger, fumigatus, flavus)	Negative
HIV	Negative
HTLV-1/2	Negative
RPR	Nonreactive
CMV	Negative
Bronchial wash/Cultures of blood and sputum	Negative for bacteria, yeast/fungus, AFB, pneumocystic carinii, CMV
ANA	Negative
c-ANCA, p-ANCA, anti-MPO, anti-Pr3	Negative
ACE	31 U/L (9-67)
Whole body gallium scan	Negative
ESR	35 mm/hr (0-15)
CRP	0.2 mg/dL (<0.8)
Total complement	76U
Cystic fibrosis	Negative for 23 mutations
Alpha-1 Antitrypsin	No mutations

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The prevalence of respiratory viral infection in exacerbation of asthma in hospitalized children

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Background: Respiratory viral infections are common exacerbating factor of asthma in both younger and older children. Of these, Rhinovirus are recognized as the most common respiratory virus associated with acute asthma in school-aged children, whereas RSV in young children. The aim of this study is to evaluate the seasonal trends and etiology of viral agents among children hospitalized for exacerbated asthma.

Methods: Nasopharyngeal aspirates were obtained from hospitalized asthmatic children at Dankook Univ. Hospital. from November 2005 through Febraury 2007. Polymerase chain reaction (PCR) assays were used for the detection of respiratory viruses(influenza virus type A, B, parainfluenza virus type 1,2,3, rhinovirus, resipiratoy syncytial virus type A, B, adenovirus, coronavirus 229E and OC43, human metapneumovirus). The demographic and clinical characteristics were reviewed retrospectively from the patients medical records.

Results: During study period, 1106 nasopharyngeal aspirates were examined from hospitalized children with respiratory symptom, of these, viruses were detected in 785(70.9%). From this population, 58 had hospitalization due to acute asthma exacerbations. Of these, infectious agents were detected in 38(65.5%) of the patients. RSV was detected in 18(31%), IFV in 10(17.2%), RV in 7(12.1%), CoV in 5(8.6%), AdV in 4(6.9%), PIV in 5(8.6%), HMPV in 2(3.4%). Coinfection was identified in 12(20.7%) of the samples. Exacerbation of asthma symptoms was greatest in winter(38%). The highest number of samples positive for respiratory syncytial virus was recorded in January(59%).

Conclusion: In conclusion, we confirmed that acute exacerbations of asthma necessitating hospitalization was often associated with respiratory viral infection, especially RSV in young children. Also, there were no significant differences between young children and school-aged children in the prevalence of respiratory virus.

We found that the causative virus in hospitalized children with asthma exacerbations follow the epidemics of respiratory virus in hospitalized children with respiratory infection.

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Chlamydia pneumoniae infection in asthmatic patients

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Introduction: It has been suggested that chronic chlamydia pneumoniae infection could be a cause for adult onset of asthma. There are data to suggest that infectious organisms, particulary the atypical bacteria, Clamydia pneumoniae, may be involved in asthma pathogenesis. It is not known whether this organism was allowed to persist after an infection, or was present prior to the development of asthma. The significance of atypical bacteria in the exacerbation of asthma is stil unclear.

Aim of the study: The purpose of this study was to determent weather anti-chlamydial treatment with azitrhomycin will improve asthma symptoms and lung function in asthmatic patients.
Materials and methods: 30 patients (mean age 35,5 years) with moderate to moderately severe asthma were treated a median of 6 weeks with azithromycin 1000 mg once weekly. All patients had chlamydia pneumoniae infection detected by specific IgA=1:40 and specific IgG= 1:256 against Chlamydia pneumoniae. Post treatment lung function and symptom score (caugh, wheezing, dyspnea) were compared with baseline values.
Results: After 6 weeks of treatment with azithromycin there was significant reduction in symptom score (p<0,01) and significant improvement in lung function FEV1 (p<0,01), Wilcoxon matched Pairs test.
Conclusion: Treatment with azithromycin significantly improved asthma symptoms and lung function indicating that Chlamydia pneumoniae may play an important role in enhancing the inflammatory processes in lower airways. Awareness of inflammation as the main pathogenetic mechanism in asthma, renewed the interest for the role of infaction in the ethiology.

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Influence of 8week therapy with Omeprazole on asthma symptoms, pulmonary function and quality of life in asthmatics with acid reflux symptoms

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Acid reflux symptoms has been found to occuere in 32 to 80% of asthmatic patients. Although the association is evident, it has been difficult to establish a casual relationship between them. Acid-suppressive treatment has been inconsistent in improving asthma control.

The aim of the study was to determine whether a proton-pump inhibitor improves asthma control in asthmatics patients with acid reflux symptoms.

Twenty-two patients receiving usual asthma cove including an inhaled corticosteroid. Patients had acid reflux symptoms and moderate to severe persistent asthma. They received omeprazole, 20mg bid for 8 weeks. The measurements were daily asthma symptoms by diary, daily morning and evening peak expiratory flow, FEV1, FVC, asthma quality of life with standardized activities(AQLOS) questionnaire score, rescue salbutamol use.

In this study we have shown that daily asthma symptoms, salbuterol use, peak expiratory flow, FEV1, FVC at 8 week did not improve significantly with omeprazole treatment. But there has been significant improvement in nighttime asthma symptoms in patients with nocturnal asthma. The AQLOS emotion function domain improved at 8 weeks(p=0,025) with omeprazole therapy.

The conclusion is that daytime asthma outcome did not improve with 8-week omeprazole therapy,but there seams to be a subgroup of asthmatic patients who benefit from excessive antireflux therapy.

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A 9 years old male with cervical lymphangioma and asthma: a case report

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Lymphangioleiomyomatosis (LAM) is rare idiopathic interstitial lung disease predominantely affecting women. Although Van Stossel first described pulmonary LAM in 1937 the natural history of LAM remains poorly understood. The true incidence and prevalence of LAM are unknown. Although current worldwide registries report LAM as a disease with no geographic preference. Most of the delay in diagnosis results from misdiagnosis and the lack of early specific clinical findings. Common misdiagnosis include asthma, emphysema, and chronic bronchitis.

Tazelaar and coworkers describe nine cases of a distinct lymphatic vascular lesions that they termed diffuse pulmonary lymphangiomatosis. Six patients included wheezing or asthma (44%) and dyspnea (22%) presented for 3 months to 20 years.

We presented a 9 years old boy, who began sufferind at 8 months of life with 38 grades fever, hiporexia, wheezing, dyspnea, cianosis. He received Cefuroxime and Ceftriaxone treatment without diminished symptoms. He enter to National Pediatrics Institute with sinusitis or laringothraqueitis probably diagnosis, the patient continued with dyspnea and inspiratory estridor, the cervical ultrasonography described hemangioma, biopsy reported right lateral lymphangioma without malignancy, it was partially extracted. Tha patient suffered thraqueostomy because compression in hypopharynx posterior wall with 50 percent diminution, supraglotic inflammation and edema. Moreover, our patient presented severe bronchoespasm with salbutamol and dexamethasone nebulizers treatment.

Because asmatic mother background, at 1 year old, he received allergy valoration, He developed nasal constipation, pruritus, and rhinorrea in the last 3 months. He received beta 2 agonist and budesonide, transfer factor, bacterial vaccine. Diagnosis of severe moderate asthma, later he developed again, volume increase in lymphangioma zone, he received OK 432. Laboratory results: dermatophagoides farinae specific IgE 1.30 UI/ml, dermatophagoides pterinissinus 2 UI/ml, total IgE 1922 Ui/ml, 7.6% eosinophilia, Prick test positive: Chenopodium, Lolium, Quercus, Timothy, Fraxinus, ligustrum, and DPT, he started sublingual immunotherapy.

This is an interesting case because lymphangioma is a rare disease, mainly in children; the association with atopia, causes a difficult control asthma. There are not references with lymphangioma and asthma in the literature, our treatment is controverted and novel.

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A severe acute asthma exacerbation complicated by pneumomediastinum and atelectasis of the lungs: a case report

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A severe exacerbation of asthma is a subacute or acute episode of asthmatic symptoms aggravation accompanied by an increased airway obstruction, respiratory insufficiency and a greater risk of complications. Pneumomediastinum and subcutaneous emphysema are rare but well-known complications of acute asthma. The generally accepted explanation for the development of pneumomedistinumis that free air tracks from ruptured alveoli along peribronchial vascular sheaths toward the hilum of the lung. From there, it extends proximally within the mediastinum. Atelectasis of the lungs is due to the formation of mucous plugs and usually responds well to the treatment of the underlying disease. This is a csse report of a 47-year old female patient L.D., who developed pneumomedistinum and atelectasis in the course of a severe asthma exacerbation. The patient has a 27-year long history of asthma and in 2004 she was vitally endangered and mechanically ventilated. In the course of this exacerbation, she developed progressive dyspnea, cough and fever. During acute coughing, an edema and crackles developed in the face, neck and chest regions, accompanied by a sudden deterioration of dyspnea, tachypnea and cyanosis. The arterial blood gas analysis at rest revealed a severe manifested partial respiratory insufficiency, and the chest X-ray established subacute emphysema. As no satisfactory regression of the disease was achieved, the chest CT scanning was performed on the eighth day, revealing atelectasis of the posterobasal left lung segment. **Conclusion:** pneumomediastinum and atelectasis are rare but possible complications which may

accompany a severe asthma exacerbation. Dramatic in appearance, they usually have a benign course and good response to conservative treatment.

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Bronchial asthma in surgical patients

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Background: The respiratory complications in patients undergoing the abdominal surgery frequently develop and aggravate course on the presence in these patients the bronchial asthma. Mechanisms of its pathogenesis and possible ways of prophylactic remain the debatable. By that, the important role plays cytokines, chemokines, adhesion molecules.

Methods: The blood levels of interleukins 4, 5, 8, 13, 16, 18, eotaxin, myeloperoxidase, VCAM-1, IgE were measured in patients with bronchial asthma, which underwent upper abdominal surgical intervention. Concentrations of some cytokines were measured by bronchoalveolar lavage. Results of investigation in patients without asthma who underwent surgery were regards as control.

Results: The increasing of IL-5, 8, 16, 18, VCAM-1, IgE, eotaxin, myeloperoxidase serum levels was noted in asthmatic patients. The level of IL-13 decreased in these patients simultaneously. The high levels of IL-8 and myeloperoxidase were noted in bronchoalveolar fluid. These changes was more significant be respiratory complications (pneumonia, edema). Asthmatic patients with bacterial infection had significantly higher serum MPO and IL-8 levels in compared with asthmatic patients without infection.

Conclusion: Inhibition synthesis of proinflammatory cytokines, chemokines is an important link in the prevention of respiratory complications during surgery in patients with asthma. The applying of IL-13 and interferon gamma may be a perspective in this view.

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Etiology of chronic cough in a population of children reffered to a pulmonologist

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Chronic cough, has been estimated to affect 7 to 10p.100 of children. When the diagnosis proves elusive, children with chronic cough are often referred to a paediatric pulmonologist for consultation to determine the reason for chronic cough. During the period from July 1994 to June 2005, 160 children were referred for chronic cough (longer than 3 week's duration). The mean age at presentation with chronic cough was 6 years. there were 83 girls and 77 boys. The most cause of chronic cough in this series was asthma in 48p.100. Other causes are gastro oesophageal reflux in 28p.100, tuberculosis in 8p.100, bronchiectasis in 8p.100, sinusitis in 5p.100 and psychogenic cough in 2p.100. cough resolved after evaluation and treatment in all cases

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A family with irritant cough from March 2007

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Background: Cough is a reflex response of the lower respiratory tract to stimulation of irritant or cough receptors in the airways mucosa. The most common cause in children is reactive airways(asthma). Specific lower respiratory stimuli include excessive secretions, aspirated foreign material, inhaled dust particles or noxious gases,and a inflammatory response to infectios agents or allergic processes. Useful information that may aid in distinguishing the cough origin may include a history or family history of atopic conditions (asthma, eczema, urticaria, allergic rhinitis) a seasonal or environmental variation in frequency or intensity of cough.

Case Report: We presented 3-members of a family with chronic cough (more than 6-weeks duration) which started after an upper respiratory tract infection and coninued in spite of antibiotic and bronchodilator therapy, In their past history there was not any atopic condition, The young son had difficult breathing especially in their house and got better outside, In allergy clinic we couldn t find wheezy breathing in rest and also after activity in their son, and PFC tests were in normal limits for parents, (the 4-year-old boy was incooperative) In the next step (and second visit) when they admitted in emergency ward we could find a problem in their house, they had been using a wall heater and its airway has been closed by a bird nest. They were exposed to noxious gases for a long time.

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Montoux test in BCG-vaccined children with asthma

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Introduction: Recent studies have proposed a decline in tuberculosis infection is a factor underlying the rising severity and prevalence of atopic disorders in developed countries. There are also conflicting reports on the effect of Bacillus Calmette-Guerin (BCG) vaccination on the subsequent development of atopy and asthma. BCG as the most potent inducer of TH1 is believed to suppress TH2 dependent atopic reaction.

Methods: Trying to prove this hypothesis, in a case-control simple sampled 100 patients younger than 5 years old who were known case of asthma with variable severity received 5 units intradermal tuberculin test. The patients were also BCG- vaccined at birth. As the control group 100 healthy children previously vaccined at birth, who were age-adjusted with case group, were also test with tuberculin test (PPD). After 48 hours, induction area was measured in two vertical and longitudinal diameters. At the same time, the severity of asthma in the case group was determined (according to classification of National Institutes of Health, National Institutes of Heart, lung and blood), by asking mothers about the frequencies of night and day symptoms or intervals of attacks. The patients were divided to three groups: Mild, Moderate and severe groups (mild intermittent and mild persistent groups were classified as one group).

Results: In our case group, mean size of PPD skin test response was 1.7mm±2.99mm but in the control group mean size was 4.42mm±3.69mm($p<0.0001$). Also in another test we divided our case group to two groups ; PPD skin test less than 5mm and more than 5 mm; again with a significant P value.

Conclusion: As a conclusion, patients with definite asthma had a significant weaker response to PPD test. This is indicator of weaker TH1 response in allergic patients, can be proposed that stimulation of TH1-immune system by BCG vaccination at birth can influence subsequent development of allergic diseases dominantly mediated with TH2-Immune system. This may promise a revolution in the future of asthma. Regarding the severity of asthma, no significant evidence supported the relation between severity of asthma and PPD skin test response, it may be explained by dominancy of moderate asthma group.

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Relationship between coronary spasticity and airway responsiveness in patients with coronary spastic angina

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Background: Several reports have suggested a possible link between bronchial asthma and coronary spasm, but the possibility of a relationship in coronary spastic angina (CSA) has not been clarified.

Methods: Airway responsiveness to methacholine and coronary spasticity to acetylcholine were examined in 39 patients with CSA and 28 patients with

chest pain syndrome (CPS). Sixteen healthy subjects as the control group were enrolled in the study and their airway responsiveness compared with that of the CSA and CPS patients.
Results: The incidence of airway hyperresponsiveness was significantly higher in the CSA group (71%) than in the CPS (21%) and control (16%) groups ($p<0.001$). The geometric mean of the log minimum dose (Dmin), defined as the cumulative dose at the point at which respiratory conductance began to decrease, was significantly lower in the CSA group (0.73 log units) than in the CP (1.18 log units) and control (1.41 log units) groups ($p<0.01$).
Conclusion: Results of this study demonstrate that acetylcholine-induced coronary spasticity is significantly related to methacholine-induced airway responsiveness in patients with coronary spastic angina. A generalized hyperresponsiveness of the vascular and nonvascular smooth muscles, including that through cholinergic mechanisms, may exist in patients with coronary spastic angina.

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GERD and anxiety in patients with severe COPD

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Objectives: To detect the impact of anxiety on shaping the prevalence of gastro-intestinal reflux disease (GERD) in patients with sever chronic obstructive pulmonary disease (COPD) based on FEV1 % less than 50%.
Methods: We examind the prevalence of symptomatic GERD ,using the Vigneri score, cough and dyspnoea with the modified Medical Research Council questionnaire in 29 male patients with COPD. Esophageal 24 h pH monitoring was used to document the diagnosis of GERD in symptomatic group. Beck Anxiety inventory was used to figuer out the impact of anxiety on expressing GERD Symptoms.
Results: Reflux disease symptoms was recorded in eighteen patients of the studied group (62%). GERD was diagnosed, based on esophogial 24 h PH monitoring, in only 11 patients of those who expressing symptoms of GERD. Mean of Anxiety score was higher among symptomatic group than non symptomatic reflecting the attribution of differnt symptoms of anxiety to the GERD syndrom.
Conclusion: Patients with severe chronic obstructive pulmonary disease have a high prevalence of symptomatic gastro-oesophageal reflux. However True GERD was documented in a fewer number of them. Psychological factors, such as anxiety and somatisation may play a role, particularly in those patients without esophageal inflammation.

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An experimental study on effect of antioxidants on pulmonary functions of smokers

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Backround: Cigarette smokes contain large number of free radicals, which cause oxidative damage of airways and lower lung functions. A prospective interventional study was carried out to observe the effects of supplementation with antioxidant vitamins on lung functions of smokers.
Methods: 200 healthy male smokers were divided into 4 equal groups of whom 3 groups were supplemented daily with either vitamin A 10,000IU or vitamin C 500mg or vitamin E 200mg and 4th group with combination of all three in same dose for 2 months. FVC, FEV1, FEV1/FVC and PEF were measured at beginning, after 2 months of supplementation and at the end of next 6 months without supplementation.
Results: After 2 months all groups showed significant improvement of all measured parameters lung functions. The difference of improvement among various supplementation groups was not significant. However, combined vitamins yielded higher trend of improvement. All groups showed decline of lung functions at the end of six months without supplementation indicating insignificant retention effect.
Conclusion: Clinicians may prescribe anti-oxidant supplementations for smokers to improve their lung functions. However, these supplementations should be given for a long period as no significant retention effect was detected.

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Symptom-limited stair climbing as a valuable predictor of postoperative cardiopulmonary complications after thoracic surgery

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Study objective: Thoracotomy is associated with high rate of postoperative cardio-pulmonary complications (POCs). We hypothesized that symptom-limited stair climbing predicts POCs after thoracic surgery.
Methods: A prospective evaluation of 40 patients undergoing thoracotomy. The 28 men and 12 women completed symptom-limited stair climbing. A separate investigator, blinded to the number of flights of stairs climbed, assessed 30-day actual outcomes for POCs, including pneumonia, atelectasis, mechanical ventilation for > 48 h, reintubation, myocardial infarction, congestive heart failure, arrhythmia, pulmonary embolus, and death within 30 days of surgery. The operations performed included 30 lobectomies, 1 wedge resection, 1 pneumonectomy, and 8 decortications.
Results: POCs occurred in 12 of 40 patients (30%). Of those unable to climb one flight of stairs, 85.5% developed a POC. No patient able to climb the maximum of five flights of stairs had a POC. The inability to climb two flights of stairs was associated with a specificity of 92%, negative predictive value of 81% and positive predictive value of 75% for the development of a POC, while the inability to climb the maximum of five flights of stairs was associated with a sensitivity of 100% and negative predictive value of 100% for the development of a POC. The number of days in the hospital postoperatively decreased with a patient's increased ability to climb stairs.
Conclusion: Symptom-limited stair climbing offers a simple, inexpensive, valuable means to predict POCs after thoracic surgery.

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Hepatopulmonary syndrome among patients of cirrhosis of liver and portal hypertension

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Background: The hepatopulmonary syndrome(HPS) is characterized by a clinical triad of liver disease, pulmonary gas exchange abnormalities leading to arterial deoxygenation, and widespread pulmonary vascular dilatation (IPVD).
Objectives: The study was undertaken to understand the frequency and clinical and pulmonary functional characteristics of (HPS)among patients with chronic liver disease.Methods:We studied 90 patients for the presence of HPS using two-dimensional transthoracic air contrast echocardiography(CE)for detection of pulmonary vasodilation, pulmonary function tests and Arterial blood gas analysis.Those patients in whom(CE)showed intrapulmonary

vascular dilatations were classified as the positive group while others were labeled as the negative group.

Results: 90 patients were included in the study, of whom 24 (26.66%) had a positive(CE); 12(13.33%) of them were found to have PaO2<70 mmHg and were qualified for the diagnosis of "clinically significant" HPS;and other 12(13.33%) with PaO2>70 mmHg were diagnosed as "subclinical HPS". Using an increased alveolar-arterial difference for the partial pressure of oxygen (AaDO2) as an indication of hypoxaemia,the prevalence of HPS was considerably higher (>15 mm Hg,(26.66 %); and>20 mm Hg,(22.22%); than using reduced partial pressure of arterial oxygen (PaO2) as a threshold (<80 mm Hg, 20.0%; and<70 mm Hg, 13.33%). For AaDO2 as the cut off, the positive predictive value for a diagnosis of HPS was low (35.29%, and 38.46% respectively). In contrast, PaO2 as a cut off had considerably higher positive predictive values (52.94%, and 75% respectively). Introducing PaO2<60 mm Hg as the cut off, the positive predictive value increased to 100%. The Child-Pugh score correlated significantly with the severity of HPS. Cyanosis, clubbing and orthodeoxia were significantly commoner in the 12 patients of clinically significant" HPS.

Conclusion: The study results showed presence of HPS and (IPVD)among patients of chronic liver disease.

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Expression of c-met in malignant pleural mesothelioma: an immunohistochemical study

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Background: Most patients with malignant pleural mesothelioma present in advanced stages of disease. Response rates and survival with currently available therapies are poor. Therefore, it is critical to identify the molecular markers of mesothelioma which would provide a way of understanding this neoplasm and targeting these markers in therapy.

Objectives: To assess the immunoreactivity of c-Met in malignant pleural mesothelioma and to analyze the potential link of the c-MET expression to some clincopathological parameters such as tumor subtype, TNM stage and patients' survival. Methods: A total of 20 patients (7 females and 13 males) with pathologically confirmed MPM; age range, (35 to 63 years) were included in the study. The patient records for the clinical, radiological and laboratory investigations and the results of closed pleura biopsies were analyzed. Pleura biopsies were stained for c-Met using immunohistochemical technique applied to paraffin sections.

Results: Of the studied tumors 18 (90%) were immunoreactive for c-Met. There were no significant relations between c-Met and patient age (p=0.569) or gender (0.755). Also there was no relation between c-Met expression and clinical symptoms. All tumors that showed distant metastasis were c-Met positive. While all c-Met negative tumors showed no metastasis. However, the difference was statistically insignificant. There was also no relation between c-Met and tumor subtype (p=0.40) or tumor stage (p=0.257). However, all T3 and T4 tumors were c-Met positive and the two c-Met negative tumors were of T2. The 2 c-Met negative patients showed one-year survival. Whereas (7/18) of patients with positive c-Met died. However, again the difference was statistically insignificant (p=0.755).

Conclusion: c-Met receptor was expressed in a high proportion of MPM. It may have a significant role in the development of MPM and could be a beneficial target for therapy. Though there was no statistically significant relation between c-Met expression and one-year survival or with different prognostic factors in MPM, we observed more c-met expression in more extensive cases and more deaths in c-Met positive cases. Additional larger-scale studies of MPM are needed to confirm the prognostic role of c-Met expression.

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Elevated c-reactive protein in patients with obstructive sleep apnea hypopnea syndrome (OSAH)

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Background: Sleep disordered breathing (SDB) is a prevalent condition in adults associated with increased cardiovascular morbidity. Circulating levels of C-reactive protein (CRP), a pro-inflammatory protein, are associated with increased risk for atherosclerosis.Therefore, CRP may be one of the links between OSAH and cardiovascular disease (CVD).We hypothesized that Patients with OSA have higher CRP values than do control subjects.

Methods and Results: We studied 28 patients (20 males and 8 females) with newly diagnosed (OSA), who were free of other diseases, had never been treated for OSA, and were taking no medications. After polysomnography, venous blood was collected at 5 AM and serum levels of CRP were investigated. We compared CRP measurements in these patients to measurements obtained in 20 control subjects (14 males and 6 females) who were matched for age and body mass index, and in whom occult OSA was excluded. Levels of CRP were significantly higher in patients with OSAS than in obese control subjects (CRP P<0.000). We evaluated relationship between the Levels of CRP and sleep study parameters such as Apnea Hypopnea Index (AHI), Desaturation Index(DI), Average of Mean Saturation ,Average of Lowest Saturation (LSAT) and Percentage of Sleep Time with Saturation less than 90 %. The results showed significant statistical positive correlations between CRP values and AHI (p< 0.00), DI (p< 0.00) and percentage of sleep time with saturation less than 90 % (p< 0.00) in the OSAS patients.

Conclusion: Levels of CRP are elevated in patients with OSAS. Therefore, OSAS is associated with increased risks for cardiovascular morbidity and mortality. The severity of OSA is proportional to the CRP level.

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Validity of multidimensional body mass index (B), airflow obstruction (O), dyspnea (D) and exercise capacity (E), bode index as predictor of hospitalization for COPD

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Background: We hypothesized that the BODE (body mass index, airflow obstruction, dyspnea, and exercise capacity) index would better predict hospitalization for COPD than FEV1 alone. Study objectives: the purpose of this study was to test in a cohort of patients with COPD, how well a multidimensional grading system that assessed the respiratory and systemic expressions of COPD would better categorize and predict outcome in these patients. Patients: A total of 150 patients with COPD(ages 45-83 yr; 89% male) recruited from the outpatient clinic of Suez Canal University Hospital were enrolled in 30 months, prospective study and followed up for a mean period of 12 months from January 2004 –June 2006.

Measurements: The BODE index was calculated for each patient using variables obtained within 4 weeks of enrollment. The main outcome measure was the number of hospital admissions for COPD during follow-up. The following variables were assessed for each patient: age, sex, pack years of smoking, FVC%, FEV1%, the best of two 6 minute walk tests done 30 minutes apart, degree of dyspnea, body mass index (BMI). We evaluated the relationship between FEV1%, the level of dyspnea, BMI, the best of two 6 minute walk tests done 30 minutes apart and BODE scores with the number of hospital admissions.

Results: After 30 months, 126 patients were available for the follow-up examination (follow-up rate, 84 %). During the follow-up period, 85 (67%) of patients required at least one hospital admission and 6(4.8%) died. In multivariate analysis a significant effect of BODE score on the number of hospital admissions was found (95% confidence interval [CI], 0.36 to 0.61; p< 0.000). In comparison, there was a significant but smaller effect of the pack years of smoking, BMI and BMI score on the number of hospital admissions {(95% confidence interval [CI], 0. 03 to 0.05; p< 0.000), (95% CI,-0.32 to-0.09; p<0.01) and (95% CI,-1.6 to-0.12; p<0.05) respectively}. FEV1%, the level of dyspnea, and 6 minute walk test were significant predictors of hospitalization in univariate analysis (p<0.000) but were excluded in multivariate analysis.

Conclusion: The BODE staging system, which includes in addition to FEV1 other physiologic and clinical variables, is a better predictor of hospital admissions than FEV1 in COPD.

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Acute and chronic eosinophilic pneumonia-our own experience with diagnosis and treatment

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Eosinophilic pneumonia (EP) is a rare disease. Some EP may result from other diseases, while in others the cause is unclear. EP is divided into acute (AEP) and chronic (CEP) forms.

The aim of the study: The authors share their own experience with the diagnosis, differential diagnosis and treatment of AEP and CEP.

Methods: Since 1995, AEP or CEP have been diagnosed in 21 patients (pts). Clinical examination, radiograph, CT or HRCT scan, laboratory examinations with peripheral blood count of eosinophils (Eo), microbiology, mycobacteriology, fiberbronchoscopy or histology were used. The pts were divided into two groups: AEP and CEP. The ANOVA and Kruskal-Wallis tests were used for statistical analysis.

Results: 13 female, median age 66 yrs (36–87) and 8 male, median 67 yrs (55–85) had one of the two forms: AEP in 7 pts and CEP in 14 pts. The main clinical symptoms were dyspnoe in 20 pts (95.2%), cough in 19 pts (90.5%), fever in 17 pts (80.9%), pain in 5 pts (23.8%). AEP or CEP was the result of concomitant disease in 3 pts with toxoplasmosis and in 1 pt. with sarcoidosis, rhinitis and allergic bronchopulmonary aspergillosis. In 15 pts no concomitant disease was identified. A statistical significance was found between the age of pts with AEP (56±12.2) and CEP (67.8±12.9) (p<0.05). There were no statistically significant differences between the absolute Eo count in male (614.8±279.3) and female (709.3±811.3) and in the groups of AEP (860.7±976.7) and CEP (579.6±429.3). 9 pts have been continually treated with corticosteroids, 8 pts temporarily and 4 pts have not received any treatment. The longest treated patient has been receiving corticosteroid for more than 9 yrs.

Conclusion: EP is a very interesting disease. The clinical symptoms are similar to typical pneumonia, but treatment with antibiotics is unsuccessful. The treatment with steroids is determined on an individual basis. Dividing AEP and CEP is not so clear cut. We could consider using the results of corticosteroids treatment as a way dividing AEP and CEP since the treatment in AEP is only temporary and in CEP on-going.

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Bone mineral density changes in male patients with chronic obstructive pulmonary disease: clinical and biochemical variables in correlation with glucocorticoids use

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Recent studies have shown that osteoporosis and vertebral fractures are quite common in postmenopausal women with chronic obstructive pulmonary disease (COPD). Few data are available in correlation between bone mass density (BMD) and men with COPD.This study was designed to investigate the prevalence of osteoporosis in men with COPD. with special regards to the role of glucocorticoids (GCs) use in these patients. We aimed to determine factors that influencing bone metabolism and the clinical variables of this group of patients. We also tried to answer the arising question: should COPD patients be routinely evaluated for BMD?

Methods: The study included 56 male patients with documented COPD for at least two years, their age ranged 24-66 years. Subjects were divided into 3 groups: group 1. consisted of 18 patients, who were oral GCs users, group 2. consisted of 18 patients who were inhaled GCs users and group 3. consisted of 20 patients, never GCs users (this group was considered as the control group). All subjects underwent measurement of BMD. pulmonary function tests (PFTs) and a number of biochemical markers of bone metabolism. The associations between BMD. PFTs. GCs use. biochemical markers and clinical variables were analyzed.

Results: Of all 56 patients with COPD. the prevalence of osteopenia and osteoporosis. as defined by WHO criteria was 26.8% and 21.4% at the lumber spine. 30.4% and 23.2% at total hip. 35.7% and 28.5% at femoral neck and 32.1% and 28.8% for total body respectively. Patients included group 1 had the lowest BMD at any site (p<0.0001). group 2 patients had over all bone mass loss, that was indistinguishable from those who were received oral GCs. group 3 patients had less bone mass reduction than the other two groups. Of the clinical and biochemical markers measured. N-telopeptide was significantly correlated with bone mass (P<0.01), but there was no correlation with other markers. The lowest mean of FEV1 (Forced Expiratory Volume in one second) was observed in group 1 patients. BMI (Body Mass Index) was weakly correlated with bone mass in the 3 studied groups.

Conclusion: Bone mass loss is a common problem in male patients with COPD. while the use of oral GCs increase the frequency of osteoporosis, inhaled GCs therapy offered no protection from bone loss. COPD patients who had never treated with GCs had also a substantial risk for osteoporosis. We advocate early screening and preventive intervention.

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Psychotherapy improve exercise tolerance in patients with Pink Puffer syndrome

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Anxiety is common in the "pink puffer" syndrome associated with chronic obstructive pulmonary disease (COPD). The degree of anxiety correlates well with perceived dyspnoea. This study examines the effect of psychotherapy on anxiety, exercise tolerance, and dyspnoea. 10 patients with stable COPD (mean forced expiratory volume in one second (FEV1)=1.25 L) had six 90 min sessions of cognitive and behavioural psychotherapy at weekly intervals. Patients completed the Beck Anxiety Inventory (BAI), 1 day before and 1 weeks after therapy. FEV1, forced vital capacity (FVC), blood gas tensions and 6 min walking distance (6MWD) were measured. 10 control patients attended weekly for lung function and 6MWD for 6 weeks, but had no psychotherapy. There were no differences in mean baseline (BAI) score, lung function, blood gas tensions or 6MWD between groups. After treatment, the BAI score had decreased from 43.10 to 20.6 (p<0.001), in association, the mean 6MWD had also improved in the psychotherapy group only, from 333 to 559 m (p<0.001), an increase of 75%. In conclusion, six sessions of cognitive and behavioural psychotherapy produced a good improvement in exercise tolerance in anxious patients with chronic obstructive pulmonary disease (Pink puffer).

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Exhaled breath analysis with electronic nose in patients with lung cancer, chronic obstructive pulmonary disease, asthma and pneumonia

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Background: Since developement of electronic nose (EN), it has been well recognized that there is a great potential in applying this technology to the field of medicine. Analysis of various biomarkers in exhaled breath allows completely safe, noninvasive, sensitive, and specific tool for the primary screening for different lung diseases. The aim of this study was to test whether exhaled breath analyzed with artificial nose could identify and discriminate between asthma, COPD, lung cancer and pneumonia.

Methods: Exhaled air was collected in plastic bags and immediately analyzed by electronic nose instrument (9185, Nordic Sensors AB) containing 14 different odour sensors. Multifactorial logistic regression analysis and support vector machine was used to find relationships between the sensor response, on derivative, on integral and the clinical diagnoses of patients.

Results: 62 individuals – 25 asthma, 13 lung cancer, 13 pneumonia, 12 other lung disease patients and 10 healthy volunteers were tested. Given table represents p values of significant relationship between electronic nose sensors and clinical diagnosis. Some sensors (1, 6, and 13) gave specific responses to particular disease; some other sensors (3, 5, and 7) shared the response with two diseases. The highest sensitivity and specificity was found for lung cancer diagnosis compared to healthy control group It was 100% and 92,9% respectively.

Sensor Nb.	Lung cancer	COPD	Asthma	Pneumonia
1	0.957027	0.350632	0.039670	0.072061
3	0.073306	0.082622	0.003552	0.005401
5	0.030928	0.822417	0.236019	0.021888
6	0.038484	0.070998	0.457104	0.241149
7	0.046645	0.136587	0.092022	0.017031
13	0.088195	0.740809	0.079040	0.023023

Conclusion: Artificial nose is able to discriminate among different lung diseases. Further development of this approach is necessary to create new screening and monitoring methods for different lung diseases.

AEROBIOLOGY

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Proteolytic activity of Per a 10 from Periplaneta americana enhances allergenicity in a mice model

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Background: Cockroach allergen sensitization has been implicated as a major cause of allergy/asthma. Recently we have demonstrated a serine protease (Per a 10) as an immunodominant allergen from *Periplaneta americana* (American cockroach). So, we aimed to investigate the immune-inflammatory responses to active serine protease in mice model of airway inflammation.

Methods: Balb/c mice were sensitized with proteolytically active allergen or OVA through intraperitoneal and subcutaneous routes and challenged intranasally with Per a 10 allergen (active / inactive), PBS or OVA. The cellular infiltration (total cell count and EPO activity in BALF), Th1/Th2 cytokines in BALF and spleen culture supernatant, serologic (specific IgE, IgG1) and histopathologic events in the various animal groups were compared.

Results: Mice challenged with active serine protease allergen or OVA showed tissue damage in the lungs, elevated serum IgE, IgG1, eosinophils and expression of inflammatory cytokines (IL-4, IL-5) in BALF and spleen culture supernatant, indicating the induction of a Th2 response. Administration of proteolytically inactive allergen (blocked irreversibly with AEBSF) induced significantly lower levels of cellular infiltration in lungs, systemic IgE production and Th2 cytokines. Quantitative evaluation of lung tissues showed decrease in total inflammation score in mice challenged with inactive protease.

Conclusion: Serine protease allergen from *P. americana* induces allergic response in a mice model and its proteolytic activity enhances the airway inflammation. The inactivated allergen may have potential for therapeutic applications.

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Cassia siamea pollen: an important aeroallergen of tropical countries

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Background: It is well known that allergy in India and other countries appear to be one of the commonest major health problems. Cassia siamea Lam. of the family Fabaceae is an important avenue tree in tropics. Its pollen grain has been known as one of the important pollinosis causing airborne bioparticles. The present study was demonstrated to identify its important allergenic components and also to study the cross reactivity between the members of the family Fabaceae.

Methods: The seasonal periodicities of airborne Cassia pollen were recorded in a 2-year aerobiological survey using a seven-day Burkard volumetric sampler (Burkard manufacturing Co., Rickmansworth, Herts, England). The allergising potential of Cassia pollen antigen was investigated by in vivo skin prick tests and in vitro IgE-enzyme linked immuno sorbant assay (ELISA). The IgE specific immunoblotting confirm the IgE sensitive reaction between the pollen allergen and patient's sera. In the present study, the whole pollen extract of C. siamea pollen was characterized by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). The presence of glycoprotein in the crude allergenic extracts of Cassia siamea pollen was detected by PAS (Periodic Acid Schif) technique.

Results: An intensely positive result (+++) was observed in response to SPT with the antigenic extract of C. siamea pollen. Within 175 patients 30.85 % shows positive allergic reactions. Immunochemical studies revealed the presence of ten allergenic components in the molecular weight ranging between 8–83 KDA. Three major allergenic protein bands (22, 17, 9 KDA) were isolated by IgE-specific immunoblotting with individual sensitive patients sera. Crude pollen extract of C siamea showed four bands of molecular weight 89, 66, 55 and 19 KDA contain glycoprotein. In the present study, the result

of dot blotting analysis indicated the presence of cross-reacting allergens in the relevant pollen extracts between the members of the Fabaceae family.
Conclusion: The aerobiological, clinical and immunological studies demonstrated that C. siamea pollen is an important aeroallergen to cause respiratory disorders in the area of their occurrence. Immunochemical studies with cassia siamea pollen revealed three major allergenic components which will be helpful for the clinicians for treatment of allergy.

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Human sensitization to different species of brassica (mustard) pollen in India

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Backed by the legacy of India's 5000-year-old civilization, Rapeseed- mustard is the second most important source of edible oil in India. On the Indian subcontinent, B. campestris and B. juncea are extensively cultivated but can share the same growing areas with B. nigra and B. napus as well. A systematic evaluation of allergenic properties of the various species of Brassica has been lacking. Therefore, the present work was aimed at assessing the sensitization as well as heterogeneity in the allergenicity of pollen of the four widely cultivated species of Brassica in India. Allergen extracts from pollen of Brassica were prepared from four different species of Brassica. Skin prick test was performed with the aqueous extracts of mustard pollen and other common inhalant allergens in 159 atopic cases suffering from bronchial asthma and allergic rhinitis and referred to allergy units for treatment. Among four species of Brassica pollen, B. campestris is the highest sensitizer (2+ and above) showing skin prick response in 13.20% cases, followed by B. juncea (11.94%) . B. nigra and B. napus pollen showed positivity in 5. 0 % and 4.4% cases respectively. Raised specific IgE in cases positive to pollen are 0.2795 + 0.08423 to B. campestris, 0.3025 + 0.1137 to B. juncea, 0.3380 + 0.1128 to B. nigra and 0.3430 + 0.1648 to B. napus respectively based on O.D values. Immunoblotting of B. campestris, showed 20, 25, 32, 37, 56, 76, 87 and 90 proteins as major allergens (more than 70% of cases). In B. juncea, however individual heterogeneity was observed and proteins of mol wt 14, 20, 25, 32, 37, 56 and 90 kD were major (more than 70% of cases). IgE binding proteins of B. nigra and B. napus were also identified. Thus, we conclude that the pollen from four different species of Brassica grown in India lead to sensitization in atopic patients.

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Aerobiological and immunochemical studies on Fungal aerosol of North Calcutta, India

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There is an increasing concern about the exposure to fungal aerosol in our environment that results in different allergy related respiratory disorders of the sensitive subjects. Besides allergenicity and mycotoxicity, microbial volatile organic compounds also get released from airborne fungi, which may cause lethargy, headache, and irritation of the eyes, nose and throat. The objectives of the present study are to prepare a fungal spore calendar in the air of North Calcutta, India, including their diurnal and circadian periodicity pattern throughout the year; to study the effect of the meteorological parameters on their frequency; and to determine the role of these spores to cause respiratory allergy.

Burkard 7-day volumetric sampler and Andersen 2-stage viable sampler were used to monitor the airborne fungal spore and culturable mold concentration over the studied area continuously for four years (Nov.'02-Nov.'06). Diagnosis of their respiratory allergic disorders was based on detailed medical history followed by their lung function test using Spirometer, skin prick test and measurement of specific IgE level in patients-sera by ELISA. Healthy asymptomatic patients were taken as control. Major allergens of the selected allergenic molds have been identified by 12% SDS-PAGE and Immunoblotting. A total of 45 fungal spore types and 24 culturable mold types were identified and recorded throughout the total sampling period. Air sampling showed 14 dominant (4.5–27% contribution) spores, among which concentrations of Cladosporium sp, Ascospores, Basidiospores and Aspergilli/Penicilli group are notably high. Different Aspergillus species, Cladosporium sp, Penicillium citrinum, Trichoderma harzianum and Alternaria alternata were among the prevalent culturable molds (>500 CFU/m3 air) as recorded. Common occurrence of respiratory symptoms and airway obstruction and mild type of lung function impairment was noted. Molds like, Aspergillus niger, A. fumigatus, Trichoderma harzeanum, Rhizopus sp etc. showed high reactivity to the patients. From Trichoderma harzianum two major allergens (35.48 kDa and 15.49 kDa) have been detected by Immunoblotting. The present survey and both qualitative and quantitative information, obtained from the study could be useful for Aerobiologists and Clinicians to forecast fungal spore load to the atmosphere and for therapeutic studies including allergy diagnosis.

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Regional importance of Alnus pollen as an aeroallergen: a comparative clinical and aeropalynological study of Worcester (UK) and Poznan (Poland) regions

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Genus Alnus is common and widespread in Central and Northern Europe. Major Alnus allergens (Aln gI) are known to cross react with other members of the Fagales order.
Aim: Assesment of contrasting regional importance of Alnus pollen in 1996–2005 within Worcester and Poznan regions, which have differing biogeographical and climatic regimes.
Methods: Daily average Alnus pollen counts were collected using Burkard volumetric spore trap. Furthermore we analyzed skin prick test (SPT) results (1996–2005) and allergen-specific IgE (asIgE) measurement results (2002–2005) against the above aeroallergen in patients of Allergic Diseases Diagnostic Center at the Department of Dermatology.
Results: The average number of Alnus pollen grains recorded annually in Poznan was more than 2.5 times that of Worcester. Furthermore, daily average Alnus pollen counts exceeded the thresholds of 100 grains/m3, 500 grains/m3 and 1000 grains/m3 more frequently in the area of Poznan in comparision to Worcester. Approximately 11% of the population (n = 5032) tested for pollen allergens (SPT) presented positive results for tree pollen allergens and 430 of these were simultaneously positive for Alnus pollens. Annual number of positive SPT results for Alnus pollen allergens was significantly related ($p<0.05$) to seasonal variations in the magnitude of the Alnus pollen catch in Poznan region. Patients with positive SPT results for Alnus pollen allergens were diagnosed as: pollinosis – 51%, atopic dermatitis – 43%, asthma – 4%, chronic urticaria 1% and eczema – 1%.
Conclusion: There is an undoubtful importance of Alnus pollen allergens, being present in high amount over Poznan region, particularly considering the

possibility of so called priming effect leading to patient sensitization in early spring and resulting in marked exacerbation of clinical symptoms later on, during birch (Fagales) pollen season.

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A novel group 2 mite allergen from dermatophagoides farinae

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Background: Mite allergy is a disease induced by inhalation of mite bodies and feces present in the house dust, leading to asthma, nasal inflammation, and dermatitis. The identification of allergens is needed for the specific immunotherapy. Our allergenomic analysis of *Dermatophagoides farinae* body extract by two-dimensional immunoblotting followed by mass spectrometry has revealed a number of new allergens. The purpose of this study is to characterize a 15-kD allergen, DFA22, with high frequency and intensity of patients' IgE binding.

Results: Based on the partial amino acid sequences obtained by mass spectrometry, a full length gene encoding DFA22 was isolated by PCR. Analysis of its deduced amino acid sequence and predicted molecular conformation suggested that DFA22 is a new member of group 2 mite allergens (ML protein). It was noted that DFA22 has a higher homology with a sheep scab mite allergen, Pso o 2 (62% identity in amino acid sequences), and a storage mite allergen, Lep d 2 (46%), than a house dust mite allergen, Der f 2 (40%), indicating that there are at least two subgroups in the group 2 mite allergens. In order to characterize the immunochemical profile of the new allergen, a recombinant protein was produced using a vector pCold-TF, which can express trigger factor (TF) fusion protein at low-temperature, and *Escherichia coli* Rosetta-gami, which has an ability to form precise disulfide-bond. The rDFA22 was purified by His-tag affinity chromatography, removal of TF, and anion exchange chromatography. Mass spectrometric analysis demonstrated that three disulfide-bonds characteristic to the group 2 mite allergens are formed in the rDFA22. ELISA indicated that its IgE binding frequency was 51% (n = 35; RAST scores to mite > 3). The binding of patients' IgE to rDFA22 was partially inhibited by rDer f 2 (~32% inhibition) and Pso o 2 (~56%), implying that these allergens shear common IgE epitope(s).

Conclusion: DFA22 is an important novel allergen that cannot be disregarded to achieve successful specific immunotherapy against mite allergy. It is also necessary to reconsider the molecular differentiation of the group 2 mite allergens.

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Experiences with the setting of a new polllen trap as a member of the Hungarian airborne pollen network in southwest Hungary

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The Hungarian network of aeropalynology includes traps thruout the country. In order to optimise this network, the Hungarian Institute of Aerobiology has set up a new trap in the Hospital for Chest Diseases of the Hungarian Calvinist Church In Hungary in Mosdós, Somogy county. The area has high impotance in the view of the researchers, because it is located close to the most poulated recreactional region, the Lake Balaton and this part of the country is probably the most pollen "infected" territory in Hungary. The equipment is a standard spore trap (Burkard), which is used all over the counry. Our aeropalinlogy team includes: three assistants, two clinicians and a technician. The team has already been reporting the pollen counts from the begining of the polens season to the National Network on a weekly basis.

Objectives: To assess the situation of pollen counts and epidemiological data on allergies in the region. Second objective was study the influence of temperature, relative humidity, rainfall and wind direction on the occurrence of pollen allergens in the atmosphere. Our results showed that, the occurrence of pollen grains in the atmosphere markedly relates to meteorological factors. We evaluated a correlation between the concentration of pollen grains in the atmosphere of the region and temperature, relative humidity, rainfall and wind direction during the vegetation period. For our analysis we selected one representative of each phytoallergen group (trees, grasses, weeds). We have chosen the Betula genus of trees, the whole Poaceae family of grasses and ragweed (Ambrosia artemisiifolia) to represent weeds. The taxons mentioned represent the most significant allergens in Hungary. Our data provides predictive information on the forthcoming patient flow and the severity of their allergic symptoms.

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Comparison of allergen spectrum in asthma and allergic rhinitis children with 3~14 years of age

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Background: To compare the allergen spectrum in asthma and allergic rhinitis children with 3~14 years of age.

Methods: Inhalant allergen skin prick test (SPT) were done in 527 asthma and 620 allergic rhinitis children respectively. The positive rates of different allergen and allergen spectrum in asthma and allergic rhinitis were analyzed.

Results: There was no significant difference on the positive rates of SPT in asthma and allergic rhinitis (77.8% vs78.9%, $P > 0.05$). The main inhalant allergens related to asthma and allergic rhinitis were Dermatophagoids pteronyssinus, Dermatophagoids farinae ,alternaria, cat dander and mugwort. The positive rates of Der.p, Der.f and molds mixture in asthma were higher than those in allergic rhinitis (64.6% vs 49.5%, 59.8% vs 47.9%, 8.8% vs 3.9%, $P < 0.05$); The positive rates of summer-autumn pollen and mugwort in allergic rhinitis were higher than those in asthma (25.6% vs 19.3%, 26.0% vs 19.3%, $P < 0.05$). 40.2% asthmatics and 46.2% allergic rhinitis children were single allergen sensitization. The multiple sensitization to mites, molds and pets were common in asthma and allergic rhinitis.

Conclusion: Mites, molds, pets and summer-autumn pollen were the main inhalant allergens related to asthma and allergic rhinitis children with 3~14 years of age. The allergen spectrums were similar in asthma and allergic rhinitis. Mites and molds allergy were more popular in asthma while summer-autumn pollen in allergic rhinitis.

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Epidermal skin prick test of airborne environmental and food allergens

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Background: Sensitization to food and airborne environmental allergens data is still rare reported. This study evaluated the epidermal skinprick test

of patients who applied to Dermato Venereology Department outpatient, retrospectively.

Methods: Retrospective study has been done for patient who came to Dermatology Venereology Department of Dr. Wahidin Sudirohusodo Hospital, Jaury Akademis hospital and private practice for 1 year from April, 2006 until April 2007. Epidermal skin prick test (ESPT) of 27 patient consisted of 12 airborne environmental allergens and 21 food allergens. Interpretation of this ESPT was divided into 3 groups: negative, positive-negative, and positive group (positive 1, positive 2, positive 3).

Results: ESPT of 27 patients were evaluated. Of 27 patient 17 (62%) female and 10 (58 %) male patient; 22 (81%) adult and 5 (19 %) pediatric patients. Most positive result were airborne environmental allergens respectively: mixed fungi 40,7 %, house dust 29,6 % and cockroach 25,9 %, while food allergens were (peanut, cockle shells, crab, shrimp) 25,9 % (nut large fish) 22,2 % and (soybean, milk fish, tuna fish) 18,5 %; whereas rice pollen and carrot allergen showed negative results.

Conclusion: The result of this study showed common allergens that wa found in patients related to tropic environmental and food habit of the patient.

Key words: retrospective, airborne environmental allergen, food allergen

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Allergic sensitization to Cheyletus eruditus

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Background: Cheyletidae mites have been described as an etiological factor for human dermatitis. In our subtropical area this family accounts as one of the most frequently found, after Dermatophagoides spp and Blomia spp, in house dust samples. The potential allergenicity of this mite family is still unknown.

Methods: We selected 15 non consecutive patients sensitized to Dermatophagoides spp and/or Blomia spp with perennial respiratory symptoms of rhinoconjuctivitis and asthma without dermatological symptoms, and no pets (dogs, cats and/or birds) at home. Ten subjects with non allergic rhinitis/asthma were selected as control group. Skin prick test with Cheyletus eruditus extract (1/50 w/v) were performed in the forearm followed by immediate reading at 15 minutes. A skin prick test was regarded as positive with a wheal of at least 3 mm.

Results: Thirteen out of 15 subjects showed a positive skin prick test. All patients of the control group had negative readings of the cutaneous test with the same extract.

Conclusion: We describe, to our knowledge, the first study to show IgE mediated sensitization to Cheyletidae mite family in patients to respiratory symptoms. More studies are needed to evaluate the actual prevalence in the general population and the clinical relevancy of this allergic sensitization.

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Allergenicity of pollen grains and pollen cytoplasmic granules

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Background: Since decades, epidemiological studies show an increased frequency of respiratory allergies. These diseases coincide with the pollination season but in most cases, no evident correlation with the pollen count is observed. In parallel, several studies have shown that in contact with water and atmospheric pollutants, pollen grains can release pollen cytoplasmic granules PCG (0.5–3 μm). Due to their small size, they may penetrate deeper into the lungs and might induce higher allergic response. These granules are suspected of playing a role in "thunderstorm asthma". The aim of our study was to compare the allergenic potential of PCG with those of the pollen, in the Brown-Norway rat.

Methods: PCG are isolated from *Phleum pratense* pollen by osmotic shock in pure water, following by filtration, centrifugation and two washes. Rats were sensitized (D0) and challenged (D21) intratracheally with pollen (0.5 mg per rat) or purified PCG ($1.5x10^6$ and $1.5x10^7$ PCG per rat). These two quantities correspond to the number of granules released by 0.5 mg of pollen, if 10% or 100% of pollen burst, respectively. Blood samples, bronchial lymph node and bronchoalveolar lavage fluid (BALF) were collected from the rats 4 days after the challenge. The number and type of cells were determined in BALF. The IgE and IgG1 levels in sera were assessed by ELISA. Pollen and PCG-induced proliferation of lymph node cells was monitored by [^{3}H]-thymidine incorporation in a lymph node assay.

Results: The number of cells in the BALF was 2 times higher in the rats exposed to pollen and to the strongest concentration of PCG ($13.8x10^6$and $14.1x10^6$ respectively) compared to rats instilled with NaCl ($6.7x10^6$). Only the rats sensitized to pollen show IgE and IgG1 levels higher than those of the controls. The PCG do not induce an increase of the IgE and IgG1 levels. Cultured lymph node cells of pollen or granules-sensitized rats significantly proliferated in the presence of pollen or PCG. However, pollen appeared to be more potent than PCG to induce the lymphocyte proliferation either in pollen and granule-sensitized rats.

Conclusion: *Phleum pratense* pollen induced humoral and cellular allergic responses in our rat model whereas PCG induced only a cellular response. This difference may be due to an extensive wash of the granules and thus a loss of soluble allergens. In perspective, we want to evaluate the allergenicity of non-washed granules.

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Aerobiology and respiratory allergies in Santa Cruz de Tenerife

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Background: In the Canary Islands the prevalence of respiratory allergy is 10–12% of the general population. The climate is very mild (mean annual: 21.2° C) and poor rainfall (mean annual:214 mm), but a high ambiental humidity (60–80%). This climatic characteristics determine that the most frequent cause of allergy in our area are house dust mites. Although

prevalence and types of pollen causing allergic symptoms have not been still well defined.

Methods: We selected consecutive patients with respiratory symptoms sugestives of allergic respiratory pathologies. They were studied by means of standard prick tests with a battery of pollen extract, and seric specific IgE analysis. A Hirst spore trap runs on a continuous-basis on the roof of the "Observatorio Atmosférico de Izaña" headquarters, since October 2004. The results corresponding to the period 2004–2006, obtained following the counting method of the Spanish Aerobiology Network (Red Española de Aerobiología, REA), are presented.

Results: The most abundant pollen types have been (by decreasing order of the annual indexes): Urticaceae, Asteraceae (99% Artemisia), Arecaceae, Myrica, Poaceae, Ericaceae, Chenopodiaceae-Amaranthaceae, Olea, Cupressaceae, Polygonaceae, Pinus, Moraceae, Plantago Mercurialis, Myrtaceae, and 43 other pollen types (<1% of the pollen spectrum). Herbs predominate in the pollen spectrum (60%) during the first half of the year. Trees (34%) predominate during the summer time. Finally shrubs (5%) pollinate mostly in spring. Most of the cited pollen types are able to cause allergy. Prevalence of pollen allergy during the period 2004–2006 was 16% of patients with respiratory allergy studied in our medical center. The most frecuent types of pollen sensitization were: Artemisia vulgaris (64,5%), Poaceae family (Gramineae) (45,2%), Parietaria judaica (24,6%), Chenopodium album (9,4%), Salsola kali (6,2%) y Plantago lanceolata (4%). The prevalence of sensitized patients to molds were less than 5%.

Conclusion: Pollen is a relevant cause of allergic respiratory diseases in our area. More studies are needed to improve the accuracy of diagnosis of pollen allergic patients.

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Distribution of allergens among allergic rhinitis patients living shiraz (Iran) region

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Objective: Allergic rhinitis is the single most common chronic allergic disease worldwide. House dust mites, grass pollens and fungal spores play has been identified to play a major role in the pathogenesis of allergic rhinitis. To determine the distribution of allergens among allergic rhinitis patients living in the shiraz region.

Methods: We investigated 56 allergic rhinitis patients (29 females, 27 males; mean age 22.3+/−14.1 years) who had symptoms.

Results: The history and clinical examination revealed that 14 patients (25%) had seasonal symptoms and 42 patients (75%) had perennial symptoms. Common pattern of multiple allergens were tree mixture allergens (58.9%), grass mixture (42.8%), mite (19.6%), candida (10.7%), tomato (5.3%), fruite (3.5%), and feather mixture (3.5%) and cladosporium, oak, ash (3.5%), alternaria, maple, hen egg, aspergillus (1.7%). Skin prick test were negative in 12 cases (21.4%). According to the prick test results, sensitivity to one allergen was found in 5 patients (8.9%) and sensitivity to multiple allergens in 51 patients. Family history were positive in 29 patients.

Conclusion: The most common allergens were tree mixture and grass in the shiraz region. It was concluded that the distribution of allergens was associated with the climatic, environmental and socioeconomic features of the region. Larger representative sample involving multi-centers in Shiraz should be encouraged in the near future.

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A research on triticum aestivum allergy in east of turkey

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The patients who visited the Center of Allergy and Immunology in Malatya/Turkey and diagnosed with seasonal allergic rhinitis (SAR), bronchial asthma (BA) and SAR+BA. The result of the skin prick tests applied to these patients with aeroallergens showed that 120 patients had allergic reaction to only triticum aestivum (TA) and 37 patients tested positive for ta with other aeroallergens in a total of positive 293 tests. There were 134 male test participants and 159 female participants in this research. 48 male and 72 female tested positive for TA only. 18 male and 19 females tested positive for TA and other aeroallergens. All these patients are currently in our allergen immunotherapy program.These results were worth giving attention to and I was informed by the Malatya Agriculture Bureau that, Malatya and surrounding areas are the capital of growing wheat wh to make bread, which is TA. On the other hand, Malatya is exporting the 20% of the world's apricot consumption. I am working on a detailed cross-sensitisation research between TA and apricot tree pollen. Since, there is no apricot tree pollen extract produced by any company, I gathered this pollen in a powder form and made connection with companies to produce the antigen to use in aeroallergen skin prick tests.

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Aerobiology of Islamabad

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Islamabad is one of the most beautiful capitals of the world with mountains and greenery all arround. But it has also highest pollen counts in asia with huge number of with Pollen allergy patients. In spring and fall numerous patients visit emergency department of different hospitals. In 2004 WAO gave Burkards Sampler on loan basis to study the aerobiology of the capital. There are two main flowering seasons in Islamabad i.e. spring season starting from the end of February to the end of April and the second flowering period during Mon-soon (fall) season starts in the middle of July to the end of September Although there are about 350 species of plants in Islamabad but the pollen trapped on the sampler were fewer.

With the Burkards Sampler we collected data for two years i.e 2005 and 2006. In 2005 total of 638017 pollen grains & 548647 mold spores per cubic meter of air were trapped. While in 2006 total pollen grains were 510954 pollen grains & 528383 mold spores per cubic meter of air were collected. The pattern of pollens and molds is nearly the sameIn spring season the highest pollen counts were those of B. papyrifera i.e. 1149/cubic meter of air per hour on 22nd March in 2005 while 1430/cubic meter of air on March 10th in 2006. During fall season pollens of Cannabis sativa were prevalent. On 18th August 2005 highest pollen counts of Cannabis was 85 per cubic meter of air per hour, while in 2006 highest pollen count of the same pollen was 25/cubic meter of air per hour on 18th August.

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% SPT + to Dust	% SPT + to POLLEN of	% SPT + to POLLEN of	% SPT + to POLLEN of	% SPT + to POLLEN of	% SPT + to POLLEN of
House Dust	B. papyrifera	Grass	Cannabis sativa	Morus alba	Dandelion
50%	48%	29%	20%	7%	18%

Mould spores found through out the year with periodic variation Pithomyces spp, Alternaria spp, Cladosporium spp, Drechslera spp, Aspergillus spp, Curvularia spp have allergenic importance while Stachybotrys spp produce saratoxins causing toxic symptoms in humans. In our study 10% patients coming to allergy centre had skin prick test positive to molds like alternaria, cladosporium and Aspergillus spp.

Skin Prick Tests (SPT) were done on the patients coming to the Allergy Centre with the extracts of these pollens and house dust. Following table illustrate the results with other pollens. The SPT results of dust and B. papyrifera are nearly the same but the intensity of reaction and size of wheal with extract of B. papyrifera is much greater. Total No. of Individuals = 702, Persons with SPT Positive 412, Persons with SPT Negative 290.

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Evaluation of allergenicity to Brassica juncea in Indian atopic population

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Rapeseed mustard is the second most important edible oil in India. Allergy to *Brassica* seeds have been reported earlier from Austria, Finland etc, but only scant information is available from Indian subcontinent.

We have investigated the allergenicity to *B. juncea* seed flour by in vivo and in vitro methods in Indian atopic cases. Antigen from *B. juncea* was extracted in 1:20 w/v by standard protocol. To assess sensitization, skin prick test was carried out with antigen extract (1:10 w/v) of *B. juncea* on the above cases. Total IgE and *B. juncea* specific IgE was estimated by Enzyme Linked Immuno Sorbent Assay. To determine the allergenically important protein, immunoblot was carried out.

A total of 159 atopic cases suffering from allergic rhinitis, bronchial asthma or both were selected from general atopic population, referred to clinical units for treatment. Of these, 37 (23.27%) were suffering with allergic rhinitis, 44 (27.67%) with bronchial asthma and 77 (48.42%) with bronchial asthma as well as allergic rhinitis. The mean age was 28.5 yr. (10–50 yr.) and sex distribution (female vs. male) was 80 vs. 79. The mean age of 50 healthy volunteers included was 25.63 yr. Of these, 15 cases (9.43 %) showed 2+ & above skin test response against seeds of *B. juncea*. The total IgE of 159 cases ranged from 12 IU/ml to 2208 IU/ml. Raised allergen specific IgE was also observed in the sera of skin test positive cases ranging from 0.221 to 0.848 Optical Density value. The antigen extract separated into 23 protein bands on Sodium Dodecyl Sulphate- Poly Acrylamide Gel Electrophoresis, in the mol wt range of 14–97 kDa. The Immunoblot revealed 12 IgE binding protein fractions in the sera of patients showing raised IgE *Brassica* specific antibodies. Some heterogeneity in the IgE binding protein fractions amongst individual patients is observed. Thus we conclude, that sensitization against seeds of *B. juncea* does exists in Indian population.

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Identification of aeroallergens in Lebanon

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Background: Aerosolized pollen are an important cause of respiratory allergy worldwide. The availability of pollen counts in westernized countries is essential for patient education and medical treatment. Aerobiological pollen data is scarce in the Middle East and had never been previously assessed in Lebanon. In order to identify and quantify pollen prevalence in Lebanon, a national aerobiology program was launched starting in the capital city of Beirut.

Methods: A *Lanzoni* pollen trap was placed on a roof top in Beirut 20 meters above ground level. A seven-day cycle drum with a 168-hour film coated with silicon adhesive was used to capture pollen grains. Slides were stained with gelatin fuschin & examined with a light microscope using a 40x objective. Pollen identification and counting was recorded on a weekly basis.

Results: A total of 9491 pollen grains/m3 were collected for the 12 month period of 2006 (except for a brief interruption during the 2006 summer military conflict). Twelve different types of pollen were identified which were Alnus, Cupressaceae, Oleaceae, Pinaceae, Juglandaceae, Mercurialis, Chenopodiaceae, Artemesia, Parietaria, Plantago, Apiaceae, and Poaceae (table 1). The 4 most common pollen found over the study period was Oleaceae (27% of total) followed by Cupressaceae (24%), Pinaceae (18% of total), and Mercurialis (17%) (table 2). The peak pollen level recorded was 327 pollen grains/m3 on April 17th with an average daily count of 93 pollen grains/m3 throughout the month of April. The lowest total pollen count was in the month of September with an average daily level of <2 pollen/m3/day.

Conclusion: This was the first aerobiologic pollen data ever recorded in the country of Lebanon. Typical elevation of pollen counts in the spring was

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	January	February	March	April	May	June	September	October	November	December	Total 2006 specific pollen count/m3
Alnus	51	24	10.2	20.4	5.4	1.8	0	41.4	3.6	34.2	192
Cupressaceae	26.4	369	1277.4	336	85.8	10.2	5.4	74.4	58.2	26.4	2269.2
Merculiaris Annua	76.8	129.6	143.52	365.4	0	0	0	838.8	19.2	76.8	1650.12
Pinaceae	0	7.8	416.52	701.4	347.4	42.6	2.4	206	0	0	1724.12
Oleaceae	0	0	86.28	1372.2	1029	87.6	6.6	0	0	0	2581.68
Parietaria	0	0	9	211.2	160.8	31.8	0	160.8	0	0	573.6
Juglandaceae	0	0	0	13.8	0	0	0	0	0	0	13.8
Plantago	0	0	0	27.6	0	0	0	0	0	0	27.6
Chenopodiaceae	0	0	0	0	0	0	13.2	3.6	0	0	16.8
Apiaceae	0	0	0	0	0	0	3.3	133.8	2.4	0	139.5
Poaceae	0	0	0	0	41.4	33.6	1.8	26.4	0	2.4	105.6
Artemesia	0.6	2.4	0	0	0	0	0	0	0	0	3
Indeterminate	27	30	53.52	42.6	30	7.8	3.6	0	0	0	194.52
Total 2006 monthly count (pollen/m3)	181.8	562.8	1996.44	3090.6	1699.8	215.4	36.3	1485.2	83.4	139.8	9491.54

Pollen	Percentage of Total 2006 Pollen Count
Alnus	2.02
Cupressaceae	23.91
Mercurialis Annua	17.39
Pinaceae	18.16
Oleaceae	27.20
Parietaria	6.04
Juglandaceae	0.15
Plantago	0.29
Chenopodiaceae	0.18
Apiaceae	1.47
Poaceae	1.11
Artemesia	0.03
Indeterminate	2.05

observed however the level was surprisingly high for an urban area. The finding of tree pollen (cupressaceae and Alnus) in winter months along with identification of a particular pollen (Mercurialis) not previously suspected were important findings for this allergen information-poor country. Since pollen collection took place in only one location (Central Beirut) and for only one year, extrapolation to a nationwide impression can not be made. Nonetheless the exciting pollen data generated in 2006, a first for Lebanon, will serve a pioneering role for the study of aerobiology and allergic disease in this country and region.

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A ten year survey (1996–2006) of the major pollen aeroallergens in Sydney, Australia

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Background: Airborne pollen is a major cause of allergic symptoms during the spring months in Sydney, Australia. Establishing the predominant pollen aeroallergens and the timing of highest pollen levels greatly assists medical personnel in developing effective treatment plans for those individuals suffering from allergic symptoms related to pollen sensitivity. This survey identifies which pollen aeroallergens are most prevalent during the spring months in Sydney and suggests possible reasons for the differences observed between seasons in an attempt to determine effective predictors of severe pollen seasons.

Methods: Aeroallergen monitoring was performed during the spring months 1996–2006 in Sydney, Australia. A Burkard 7 day volumetric spore trap was used for pollen collection and slides were analysed using standard protocols. A daily pollen count was expressed for each species in grains per cubic meter (CuM). Data was collected from August to November of each year (spring in the southern hemisphere).Daily rainfall data was obtained from the Bureau of Meteorology and expressed as millimetres of rain on a daily basis for the entire ten year period of the survey. Pollen data was then collated into 3 groups – tree, grass and weed pollen. Each group was analysed for seasonal trends and correlated with rainfall data. For each year of the survey, the season duration was identified, as well as peak day and peak count, average daily pollen count and total season pollen count for each group.

Results: Tree pollens make up the majority of the total pollen count in Sydney, followed by grasses and weeds. Major tree pollen species identified in Sydney include Cypress,Pine, Plane and Casuarina. Tree pollens tend to reach peak levels in mid to late September, and grass and weeds pollens in mid to late October. The highest average daily pollen counts for all groups occurred in years with the highest autumn/spring rainfall. Average daily pollen counts ranged from 23–147 grains/CuM (trees), 1–25 grains/CuM (grasses) and 0–15 grains/CuM (weeds).The period of pollen season duration also varied considerably each year.

Conclusion: The results of this survey indicate the major pollen aeroallergens present during the spring months in Sydney, Australia. The survey also highlights considerable variability between pollen seasons and the need for ongoing aerobiological pollen monitoring in the future because of highly variable climatic conditions that clearly impact upon the pollen counts.

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Influence of meteorological variables on pollen counts in Sydney, Australia: longitudinal data examining effects within and between pollen seasons

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Background: Airborne pollen is a significant cause of allergic symptoms in sensitised individuals. Various meteorological factors have an effect on pollen levels, on a day to day basis throughout the pollen season, and on a seasonal basis over time. By understanding the effects of these meteorological factors, it may be possible to develop effective indicators which may assist in predicting high pollen days within a season to assist in symptom management.

Method: Airborne pollen was collected between 1996–2006 in Sydney, Australia. A Burkard 7 day volumetric spore trap was used to collect data and slides were made up and analysed using standard protocols. Pollen counts were expressed as grains per cubic Meter (CuM) for various species and then grouped into trees, grasses and weeds. Meteorological data was obtained and included minimum temperature, maximum temperature, daily rainfall and humidity. Spearman rank correlations were used to assess the effect of the meteorological variables upon the day to day pollen counts and the seasonal pollen counts as a whole. We then examined cross correlation plots between first differenced 4 day moving averages in order to detect short term associations between fluctuations in meteorological variables and fluctuations in pollen counts.

Results: Examining daily counts within a given pollen season, there was a statistically significant positive correlation between daily maximum temperature and the daily pollen count for all pollen species ($p < 0.05$). Daily rainfall and high humidity were negatively correlated with the daily pollen count for all species ($p < 0.05$). Day to day changes in maximum temperature were moderately positively correlated with a simultaneous change in the total pollen count ($r = 0.35$), but weakly negatively correlated with a change in the total pollen count 4 days later ($r = -0.15$). Examining season to season associations, mean seasonal pollen counts were negatively correlated with mean seasonal daily maximum temperature ($r = -0.782$ total pollen and $r = -0.600$ for grasses).

Conclusion: On a day to day basis, increases in daily maximum temperature lead to an increase in pollen levels and daily rainfall leads to a simultaneous decrease in the daily pollen level. Overall, cooler temperatures across a season often lead to a higher total pollen count. Ongoing pollen monitoring is needed to further examine the effects of meteorological factors on daily and seasonal pollen counts.

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The characterization of native thaumatin-like allergen named Cry j 3 from Japanese cedar *(Cryptomeria japonica)* pollen

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Research Center for Allergy and Rheumatol, Kanagawa, Japan; [5]Azabu University, School of Veterinary Medicine, Kanagawa, Japan.

Background: Japanese cedar *(Cryptomeria japonica)* pollinosis is one of the most prevalent allergies in Japan. Four allergens named Cry j 1, Cry j 2, CJP-4, and CJP6 have been identified as the causative allergens from Japanese cedar pollen. Recently Cry j 3 has cloned as a homologue of Jun a 3. Jun a 3 is a major allergen from mountain cedar *(Juniperus ashei)* pollen. However native Cry j 3 has not been isolated and there were no reports on its allergenic activity.
Objective: The aims of this study were to isolate native Cry j 3 and assess its IgE-binding capacity in patients with Japanese cedar pollinosis.
Methods: Cry j 3 was purified from Japanese cedar pollen by multi-dimensional chromatography. We assessed the IgE-binding capacity by immunoblot analysis and enzyme-linked immunosorbent assay (ELISA). Moreover, we assayed the capacity of Cry j 3 to induce histamine release from the patients' leukocytes. We cloned cDNA corresponding to purified Cry j 3 from a cDNA library of Japanese cedar pollen.
Results: We purified native Cry j 3 from Japanese cedar pollen as a 27 kDa protein. The IgE-binding frequency of Cry j 3 from the sera of patients allergic to Japanese cedar pollen was estimated as 27% (27/100) by ELISA. Cry j 3 induced the release of histamine from leukocytes. We cloned the cDNA and named it Cry j 3.8. Cry j 3.8 cDNA encoded 225 amino acids and had significant homology with thaumatin-like proteins.
Discussion: Cry j 3 is an additional causative allergen in Japanese cedar pollinosis and may play crucial rules in the cross-reactivity with oral allergy syndrome.

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Structural characterization of norcoclaurine synthase suggests the enzyme to be a true member of the Bet v 1 protein family

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The enzyme Norcoclaurine synthase (NCS) found in the common meadow rue, *Thalictrum flavum*, and other plants shows sequence homology to members of the class 10 of pathogenesis related (PR 10) proteins that contains allergens such as the major birch pollen allergen Bet v 1, the major cherry allergen Pru av 1, and the major apple allergen Mal d 1. The enzyme catalyzes the condensation of dopamine and 4-hydroxyphenylacetaldehyde to (S)-norcoclaurine, an important step during synthesis of benzylisoquinoline alkaloids. This group of plant secondary metabolites includes pharmacologically active substances like the analgesic morphine.

Whereas the physiological function of PR10 class allergens is still unknown, NCS activity was studied in detail. Investigation of the structural properties of NCS by NMR spectroscopy can thus not only provide new information concerning the reaction mechanism of the enzyme, but is also expected to help clarify the long standing and heavily debated question on the physiological function as well as the reasons for the allergenic potential of members of the Bet v 1 allergen family.

As the first important step towards the three-dimensional solution structure, we optimized expression of recombinant NCS in *E. coli* and established an efficient purification protocol yielding high amounts of pure isotopically labeled protein. Spectroscopic analysis by circular dichroism and NMR spectroscopy showed that the protein is properly folded with well defined secondary and tertiary structure, probably similar to that of its allergen homologue Bet v 1. In consequence, comparison of NCS and Bet v 1 concerning structural and allergenic properties might help to elucidate which structural features are important for the allergenicity of Bet v 1 allergens. Enzyme kinetics were investigated by monitoring substrate turnover using 1D-^{1}H-NMR. Additionally, NMR titration experiments with substrate analoga were carried out to characterize substrate binding.

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Measurement of Alternaria spores and Alt a1 levels in the atmosphere of Ciudad Real (Spain)

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Background: Alternaria alternata is one of the mayor fungal antigen producers. The mayor allergen Alt a1 is expressed by Alternaria and other taxonomical species related.
Aim: Analyze the correlation between Alternaria spores levels and Alt a1 in the atmosphere of Ciudad Real.
Method: Daily spore concentrations were sampled daily during 2003 an 2004 using a Burkard volumetric spore trap. The total number of spores was expressed as n° spores/m3/day. An Air Sentinel and monoclonal binding were used to measure the mayor allergen Alt a1 (pgr/ml). Meteorological data of the years 2003 and 2004 were obtained.
Results: A wide variety of fungal spores were compiled. Cladosporium was the most prevalent fungi (77%), followed by Alternaria (6,6%). The highest airbone Alternaria spp. spore concentration were obtained during spring and summer (may-june and august-september), with a decrease in july due to the high temperature (less than 100 spores/m3/day, as the rest of the months). Correlation may be established between the total spore counts and the atmospherical conditions (temperature 20–30° and accumulate rainfall but not daily rainfall). Alt a1 levels are more homogeneous along the year. These are no correlated with spore counts.
Conclusion: Fungal spore release is influenced by meteorological conditions. No relation between Alt a1 levels and meteorological factors was observed.

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Changes in the allergenic protein content of the Cupressus arizonica pollen exposed to air pollution

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Cupressus spp. pollens are an increasing cause of allergies in various regions. It is a common opinion that recent increases in this pollinosis are due to extensive plantation of cypress trees, and the potential adjuvant effect of the chemical pollution generated by the urbanism. In Tehran, the perfect adaptation of cypress trees to climatic conditions, have led to large-scale plantation causing a significant augmentation of allergic diseases from the middle of winter to the beginning of spring.

In this study, we tried to evaluate effects of air pollutants on pollen protein content of the *Cupressus arizonica* planted in the highest polluted areas of Tehran. Polluted areas of the city were selected according to data provided by Air Quality Control company. In the first sampling, pollens were collected just after microsporangia bursting, and the second sampling was conducted 2 weeks later, in order to permit a natural pollen exposure to the urban air pollution. After two weeks exposure to urban air pollution, the colour of purified pollens turned from a brillant yellow to a dark green. Bradford protein assay showed an important diminution of the total protein content in polluted pollens from 220 μg/ml to 150 μg/ml. Comparison of protein components of polluted and non-polluted pollen extracts via SDS-PAGE and densitometric analysis revealed appreciabe differences between their electrophoresis profiles. In non-polluted pollen extracts, a protein band with an approximate molecular weight at about 45 kDa, called Cup a 1, (known as the major allergen of C.arizonica pollens) represented 75% of all protein content

and another protein at 35kDa constituted only 7% of the total protein content. In pollutted pollens the concentration of the protein at about 45 kDa considerably diminished to 14.6 % of the total protein content but the amount of the protein fraction at 35kDa increased to 50.2%. Immunoblots, conducted to evaluate sera IgE reactivity of cypress allergic subjects to the new major protein of polluted pollen extracts, showed that the protein of 35 kDa was also the major allergen of *C.arizonica* pollen extracts in allergic subjects living in Tehran.

Important differences observed between protein profiles of non-polluted and polluted pollen extracts, reflected the fact that environmental pollution induce the augmentation of an allergenic protein which may have a negative repercussion on the health of urban popullations.

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The allergen-specific IgE reactivity pattern of Chinese house dust mite allergic patients

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Background: Two species of house dust mites (HDM), Dermatophagoide pteronyssinus (Der p) and Dermatophagoide farinae (Der f), have been recognized as major allergen sources in Chinese allergic patients according to epidemiological surveys using in-vivo or in-vitro assays. The specific IgE reactivity pattern of European and American HDM allergic populations are well characterized. However, few studies have addressed the allergen-specific IgE pattern of Chinese HDM patients.

Methods: Serum samples from 62 patients who had allergic asthma and/or allergic rhinitis and who showed skin prick test and UniCAP positive to Der p and Der f were obtained from South and Southwest of China. Sera were analyzed by crossed radioimmunoelectrophoresis (CRIE) and immunoblotting to visualize the allergen-specific IgE reactivity patterns.

Results: 23 allergens of Der p and 16 allergens of Der f were detected by CRIE. The most frequent allergens were identified to belong to HDM group 1 and 2. All sera had specific IgE against both Der p 1 and Der f 1, while 92 % of sera recognized Der p 2 and 95 % reacted against Der f 2. Antigen 16 of Der p showed IgE binding by 69%. After immunoblotting, patients' IgE mainly showed binding to proteins of around 25 and 14 kDa which were identified as HDM group 1 and 2 allergens, respectively. The reactions of the sera by immunoblotting were 53 % for Der p 1, 63 % for Der p 2, 31 % for Der f 1 and 59 % for Der f 2.

Conclusion: The HDM specific IgE reactivity patterns of Chinese mite allergic patients are similar to that of other patient populations. In addition to the commonly recognized Der p 1, Der p 2, Der f 1 and Der f 2, a novel major allergen, denoted antigen 16, was identified in the Der p extract. The analysis indicates that the Der p and Der f extracts used in this study can be used worldwide both for diagnosis and treatment.

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Immunoglobulin E and Immunoglobulin G epitope-mapping of Blo t 5, the major allergen of Blomia tropicalis

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Background: *Blomia tropicalis* is one of the most predominant domestic mite species in tropical and subtropical countries. Blo t 5 is a major allergen with reported IgE sensitization rates of 43–92% in *Blomia tropicalis* allergic subjects.

Aims: The objective of this study was to determine the IgE, IgG1 and IgG4 epitopes of Blo t 5 in mite allergic children.

Methods: Five overlapping peptides, F1 (aa 1–45), F2 (aa 41–80), F3 (aa 70–117), F4 (aa 1–80), and F5 (aa 41–117) were generated by PCR from Blo t 5 full-length cDNA. The IgE, IgG1, and IgG4 reactivities to these peptides and full length Blo t 5 were determined by ELISA assay with 20 Blomia tropicalis skin test positive children. The IgE ELISA unit was determined by a semi-purified human IgE as standard curve, whereas IgG ELISA unit was determined by OD405nm X dilution factor.

Results: The IgE epitope mapping experiments revealed very low levels of IgE binding to small peptides at N-terminal, F1 and F2 ($p < 0.005$) compared to F3, and larger peptides, F4 and F5, indicating that Blo t 5 major IgE epitopes were located at C-terminal and conformational dependent. In contrast, higher levels of IgG1 to F1 and F3 were found compared to F2 ($p < 0.005$) indicating that the major IgG1 epitopes are located at both the N- and C- terminal of Blo t 5. The peptides F1, F2, F3 and F4, bound lower levels of IgG4 ($p < 0.005$) than F5 suggesting that the IgG1 and IgG4 epitopes are unique and the major IgG4 epitopes seem to be located at C-terminal and conformational dependent.

Conclusion: The data revealed both IgE and IgG4 epitopes are found mainly in the C-terminal of Blot 5 and are highly conformational dependent, whereas IgG1 epitopes are found throughout the whole Blot 5. The results suggest that recombinant hypoallergenic Blo t 5 for immunotherapy can be achieved by modifying the IgE epitopes while retaining the IgG epitopes.

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Aeroallergen sensitivity of Iranian patients with allergic rhinitis

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Background: Allergic rhinitis is a prevalent allergic disease throughout the world. A recent study showed that allergic rhinitis is a common problem in Mashad, Iran, However, Little is known about the relation between skin test reactivity to aeroallergens and allergic rhinitis in Iran.

Objective: The aim of this study was to characterize the most frequent indoor and outdoor (grass and tree) inhalant allergens involved in allergic rhinitis in Mashad, Iran.

Methods: A total of 99 cases (females: 49, males: 50, aged between 3 to 68, mean age 25 years), that were diagnosed with current active allergic rhinitis based on the ISAAC rhinitis Core questionnaire and clinical presentation, underwent a standardized skin prick test with 28 (7 indoor, 13 grasses and 8 trees) aeroallergens. Results: 75% of patients were reactive to at least one of the aeroallergens and 69% of patients had positive reactions to more than 3 allergens but just 3% of patients were mono sensitized. Sensitization was more prevalent for outdoor allergens than for indoor allergens in patients (74% vs. 31%).The individual pollen allergens with the highest positive rates were pollens of Careless weed (Amaranthus palmeri) 67.4%, Russian thistle (Salsola kali) 64%, Redroot (Amaranthus retroflexus) 61%, Burning bush (Kochia scoparia) 58%, Lambsquarters (chenopodium album) 56%, Eastern cotton wood (Populus deltoides) 54%, White ash (Fraxinus americana) 53%, American sycamore (Platanus occidentalis) 43%. The most common Indoor allergens were Dermatophagoides pteronyssinus %20, cockroach mix (American and German) 20%, house dust 20%, Dermatophagoid farinae 17%.

There was a significant but weak correlation between sum of all wheal sizes and serum total IgE level (P<0.001) and also between the number of positive skin test responses and the serum total IgE.The sum of mean wheal sizes was larger in men than in women (371 vs. 219 mm2) for outdoor allergens but it was not statistically significant. Redroot, Russian thistle and Careless weed had the greatest mean wheal size (60, 40 and 37 mm2, respectively).

Conclusion: The present study revealed that the prevalence of the Skin Prick reactivity to outdoor allergens particularly grasses (weeds), is high among

Iranian allergic rhinitis patients, although Indoor allergens do not seem to be very common among patients.

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Solution structure of Phl p 3, a major allergen from Timothy grass pollen

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More than 20% of population in industrial countries suffer from seasonal allergic rhinitis, commonly known as "hay fever". Pollen from grasses, trees and weeds are some of the most frequent allergen sources. Structural similarities found among the allergens from diverse allergen sources represent the molecular and structural basis for immunologic cross reactivity and thus explain why allergic patients can exhibit clinical symptoms and IgE reactivity to various unrelated allergen sources. Binding of an allergen to the corresponding IgE is an important step in the immunoreaction. Knowledge of the structure and binding regions is therefore important to understand this process in order to improve diagnosis of disease and to prepare better allergic-specific immunotheraphy.

Timothy grass (*Phleum Pratense*) pollen is an important allergen source in Northern and Central Europe and its set of allergens is the best characterized within different grasses. Phl p 3 was recently isolated from Timothy grass pollen extract. From the amino acid sequence and homology comparison belongs to the group 3 allergens. In this work we determined solution structure of Phl p 3 by NMR. This solution structure and comparison with the structures from other member of the group 3 (like Phl p 2 and Phl p 1) should help us to find specific IgE-reactive eppitopes and to find better cure for the disease.

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Investigating pollen asthma deaths in islamabad, Pakistan

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Introduction: Severe asthma and deaths have been reported in Islamabad during the spring allergy season, which affects thousands of residents. We investigated the deaths of 3 patients due to asthma, during the last ten days of March. In order to find a possible cause of these symptoms, we investigated the siblings of the deceased. Case 1: Male, 19 years, with a history of rhinitis during spring season, developed sudden difficulty in breathing while walking in the market, just after sunset. He was shifted to the nearest hospital. He died just on reaching the hospital, approximately 15 minutes after the onset of the attack. Case 2: A female medical doctor, 27 years, mother of 2 children aged 1 and 2 years, developed asthma suddenly about 1 hour after sunset. She was rushed to the hospital but, like the previous case, expired before reaching the hospital, approximately 20 minutes after onset of symptoms. Case 3: A 30 year old female, mother of 2 children aged 2 and 4 years, expired at home just before midnight, approximately 25 minutes after the onset of the first symptoms of asthma. Commonalities in all cases: All the deceased patients had a previous history of allergic rhinitis and occasional asthma during the spring pollen season in Islamabad. None had ever suffered from severe asthma, or been hospitalised. Very prominent is that the counts of Paper mulberry pollens were in excess of 30,000 grains / cm3 that day, and humidity was >80%, due to a forthcoming thunderstorm and rain. None of the patients was taking regular preventative treatment for allergy or asthma, at the time of the episode. None had been investigated for allergy during their lifetime.

Methods: We contacted the siblings of the expired patients and found a positive history of seasonal spring allergy in 2 brothers of case 1, 4 sisters and one son of case 2, and one sister of case 3. One sibling each of cases 1 and 2 never reported any symptoms of asthma ever, while case 3 did not have any other sibling.

Results: Skin prick allergy testing of all the symptomatic patients revealed strong results (more than 10 mm wheal) to pollens of paper mulberry (Broussenetia papyrifera) pollens.

Conclusion: It is probable that the deceased patients suffered from allergy to pollens of paper mulberry, and expired due to the thunderstorm effect, during the peak of paper mulberry pollen allergy.

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Incrimination of Blomia tropicalis as a potent allergen in Indian house dust

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Allergy to house dust mites particularly to the genus Dermatophagoides is a common feature in tropical climate like Kolkata, India leading to various nasobronchial allergic manifestations. Analysis of house dust samples collected from the houses of the asthmatic patients of Kolkata revealed a variety of acarine fauna of which the mites belonging to the genus Dermatophagoides predominated, comprising 60.4% of the total acarine population followed by Blomia tropicalis (16.6%) and Austroglycyphagus geniculatus (6.8%). Perusal of literature revealed that works on the role of house dust mites in allergic etiology mainly concentrated with the genus Dermatophagoides alone. No work incriminating the role of Blomia tropicalis in house dust allergy has yet been reported from India, although it contributes significantly towards the domestic acarine population. High prevalence of this mite species in the patients' environment tempted us to search for an allergic etiology towards this species of mite. Allergy skin test with house dust and house dust mites revealed an encouraging result against Blomia tropicalis. 72% allergic asthmatics of Kolkata responded positively towards this species of mites. The total serum IgE level in patients varies between 54 to 4040 KU/ml (mean 369+487 KU/ml). Detection of allergen specific IgE antibodies against this species of mites also confirms its role in house dust allergy. 30% patients showed very high, 20% high and 40% showed moderate IgE level against this mite antigen. The present study incriminates Blomia tropicalis as an important source of allergen in house dust in Indian condition.

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Aeroallergen sensitivity in patients with allergic rhinitis and or asthma

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Background: Allergic rhinitis and asthma are prevalent through out the world. For the efficient diagnosis and its effective therapy it is very important to know about the prevalance and pattern of sensitisation in a located geographical area. Little is known about this in south coastal city of Chennai in India, Southern Asia.

Aim: Aim of this study was to characterise the most frequent inhalant allergen involved in Allergic rhinitis and Asthma.

Subjects and Methods: A total number of 104 cases [males 54 and females 50, aged from 6 to 60] who were diagnosed to have Allergic rhinitis and or asthma were subjected to undergo Standradised SKIN PRICK TEST with 22 common Allergens. Reactions was considered to be positive if the mean wheal diameter was atleast 3 mm more than that of the negative control. Each patient was evaluated for sensitisation to allergen, number of positive reaction and mean wheal size.

Results: 78 % of the patients were reactive to atlest one of the allergen and 61 % of the patients had positive reactions to more than 3 allergens. The most common allergens were, D.PTERONYSSINIUS 65%, D.FARINAE 60% HOUSE DUST 52% MOSQUITO MIX 44%. Among Pollens PARTHINIUM 40% AND CHENOPODIUM 28% and Among food allergens CITRUS MIX 45%.

Conclusion: The present study revealed that the prevalance of SKIN PRICK REACTIVITY to allergens particularly to HOUSE DUST MITE is high among with peoples in Chennai.

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Sensitization to locally prepared allergenic extracts of house dust mites in patients with respiratory allergy at the eastern of kingdome of Saudi Arabia

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Introduction: The prevalence of sensitization to allergens from house dust mites (HDM) in patients with respiratory allergens appears to be directly correlated with exposure.

Objective: To explore sensitization to locally prepared allergenic extracts of HDM in patients with respiratory allergy at the Eastern of Kingdome of Saudi Arabia (KSA).

Methods: House dust collected from different houses at KSA was put on culture media plates containing a mixture of charcoal, gypsum and water, which was then dried and wood excelsior with crushed maize for HDM nutrition was added. These were incubated in chamber with an adjustable relative humidity at 73% and temperature at 23 °C for one month with regular observation. Three types of HDM were isolated and cultured: {(Dermatophagoid Pteronyssinus (DP), Dermatophgoid Farinae (DF) and Blomia Tropicalis (BT)}. Allergenic extracts were prepared locally using the weight/volume method concentration 1/10 for skin prick test (SPT). Sensitization was based on a positive reactivity to standard SPT performed on patients with respiratory allergy.

Results: 100 patients with the primary diagnoses of allergic rhinitis in 64 patients (64%) and bronchial asthma in 34 patients (34%) were studied. They aged from 8 to 62 years old (mean=32 ±13SD). SPT was positive to: DP in 53 patients (53%) (44% polysensitization with other HDM and 9% monosensitization), DF in 46 patients (46%) (42 % polysensitization with other HDM and 4% monosensitization) and BT in 2 patients (2%) (2% polysensitization with other HDM).

Conclusion: Prevalence of sensitization to locally prepared allergenic extracts of HDM, especially DP followed by DF, was predominant in patients with respiratory allergy seen in the Eastern of KSA. The sensitization to BT in few patients is described for the fist time and needs further evaluation in relationship to its significance in this geographical area. Properly manufactured extracts to other local allergens may help to determine new regional environmental factors for allergy.

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Sensitisation to house dust and house dust mites in nasobronchial allergic patients of Kolkata Metropolis, India

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Background: Recent studies revealed the presence of huge quantity of reported allergenic mites viz. Dermatophagoides pteronyssinus, D. farinae and Blomia tropicalis in the indoor environment of Kolkata. Immediate hypersensitivity to these dust mites along with house dust may play a role in pathogenesis of nasobronchial allergic diseases. So, The present study evaluated the sensitisation rates towards house dust and house dust mites among patients suffering from various nasobronchial allergic manifestations residing in Kolkata metropolis area, India.

Methods: The Skin Prick Test was performed on a total of 1079 patients(585 male and 494 female and 50 healthy controls) using extracts of four allergens viz. Dermatophagoides pteronyssinus, D. farinae and Blomia tropicalis and house dust allergens. Total serum IgE was measured by using EIA technique and specific IgE to these allergens measured by following Pharmacia Immuno CAP 100 System. The influence of age and gender, if any, on allergen sensitivity was also investigated.

Results: The sensitisation rates among nasobronchial allergic patients to house dust and house dust mite allergen tested were as follow: house dust (96.22%), Dermatophagoides pteronyssinus (75.06%), Blomia tropicalis (72%) and D. farinae(63.72%). The frequency of positive skin response was found to be independent of age and gender. The total serum IgE level in patients varies between 54–4040 IU/ml (mean 369 ± 26.51). Specific IgE antibody test proved that 83% patients showed sensitivity towards at least one of the allergen tested.

Discussion: Nasobronchial allergic patients are highly sensitive to the house dust and other three allergenic mites. Thus, these dust mites and house dust should be considered important allergenic sources of this area.

DRUG ALLERGY

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Adalimumab causing vasculitis

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A 19 year old white male with past history of Crohn's disease presented to ED with one-week history of pain, rash and edema involving lower extremities. Pain was localized to his lower extremities bilaterally extending from mid shin to distal toes, particularly worse on the medial aspect of left foot. He also noticed pruritis and palpable rash in the same area. His palpable pruritic rash had improved to just a petechial rash by the time he presented. He did bring pictures of the rash when it was at its worst. There were no symptoms of exacerbation of Crohn's disease. He was compliant with his medication. The occurrence of rash was temporally related to injection of Adalimumab, his last dose was 2 weeks prior to admission. Physical exam revealed edema of both lower extremities with scattered nonblanching petechial rash. There was also tenderness and decreased range of motion of ankle. There was no sensory deficit.

Laboratory results including, CBC, CMP, PT, PTT, Rheumatoid factor, Antihistone antibody, Anti-DS DNA antibody, C3 and C4 levels were within normal range. ESR was 19, CRP was 9.080. ANA, ANCA, Serum Cryoglobulin and Hepatitis screen were negative. Skin biopsy results revealed mixed cellularity vasculitis, suggestive of resolving leukocytoclastic vasculitis.

After discharge, he received next dose of Adalimumab resulting in redevelopment of the same rash only more severe in presentation.

Vasculitis appears to be a rare but potentially serious complication with anti-TNF therapy that likely represents a type III hypersensitivity reaction. In patients who develop this reaction, safe practice is to stop the TNF antagonist and treat with a regimen of steroids and antihistamines with or without immunosuppressive agents. This case represents a very rare hypersensitivity reaction to humanized antibody like Adalimumab.

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Allergy to etanercept in a patient with rheumatoid arthritis

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Drugs that antagonize TNF-á have demonstrated their effect controlling inflammatory activity and inhibiting the progression of structural damage on patients with chronic arthropathies. Otherwise the use of these drugs can produce quite frequently some side effects, being skin reactions one of the most frequent after infectious diseases. A wide amount of skin reactions have being described with these drugs, such as systemic erythematous lupus, necrotizing vasculitis, discoid lupus, multiform exudative erytema, Stevens-Johnson syndrome, toxic epidermal necrolysis, eczema, urticaria, etc. We introduce the case of a 57 year old patient with rheumatoid arthritis, which developed a skin reaction with a high suspicion of being a hypersensitivity drug reaction. After two years being treated with etanercept (25 mg, twice a week) and leflunomide (20 mg a day), he developed 15 minutes after administrating the drug a vesicular itchy rash on the injection sites with itchy isolated wheals all over his body. We performed some skin tests including prick-test, intradermoreaction and patch-test with etanercept, infliximab, adalimumab and leflunomide. We used a blood sample of this patient to do a basophiles activation test (BAT) with the same drugs. We performed subcutaneous provocation with etanercept and adalimumab as well. In order to rule a possible irritative reaction out, we performed prick-test and intradermoreaction with etanercept, in five healthy controls.

Prick-test and intradermoreaction with etanercept were positive and negative for the other drugs. BAT was positive with etanercept and negative with leflunomide, infliximab and adalimumab. Due to these results, we carried out subcutaneous provocation with etanercept and adalimumab, being positive to etanercept and negative to adalimumab. The skin test in healthy controls was in all of the cases negative.

We presented the first case of a patient with a type I hypersensitivity reaction with etanercept, demonstrated with test in vivo and in vitro, and confirmed with subcutaneous provocation. Despite the skin reactions with this type of drugs are quite frequent, there is not enough evidence published trying to clarify the immunologic mechanism implicated on it. This article opens a new expectative on the study of the side effects and skin reactions of these drugs, taking into account that they are very effective drugs in chronic arthropathies and that their use will probably be widely spread.

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Oral hydroxychloroquine desensitization in a 50 year old male with cutaneous lupus

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Background: Hydroxychloroquine is an antimalarial drug with immunomodulatory and cytotoxic properties used in the treatment of lupus erythematosus. Adverse cutaneous reactions in the form of rashes is its most common side-effect occurring in about 3–10%. Though, a mild reaction, it is the often the reason for cessation of treatment. This is the first reported case of rapid oral desensitization to hydroxychloroquine.

Methods: We present a rapid oral hydroxychloroquine desensitization protocol in a patient who developed pruritus to hydroxychloroquine.

Results: This 50 year old patient diagnosed with cutaneous lupus presented with a 1-year history of malar rash, arthralgia and diagnostic tests consistent with cutaneous lupus. He was started on prednisone, calcium carbonate and aspirin. After one year, he has was started on hydroxychloroquine for better control at 200 mg per day. On the 2nd week of treatment, he developed pruritus within two hours of intake. He was then referred for hydroxychloroquine desensitization. We used a rapid desensitization protocol as opposed to a published slow desensitization protocol by Mates, et al covering 36 days. Our patient was started at 1/30th of the total dose doubling q 30 minutes reaching the cumulative dose for the day of 200 mg. There was no pruritus and other adverse reactions observed. The patient is still being maintained on hydroxychloroquine at the present time.

Conclusion: This is the first reported rapid hydroxychloroquine desensitization protocol. This demonstrates that rapid desensitization may be used when a drug is essential in an allergic patient.

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Oral mitotane desensitization in an 18 year old female with adrenocortical carcinoma

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Background: Mitotane is a chemotherapeutic drug used in the treatment of unresectable adrenocortical carcinomas. It is associated with various side-effects that may affect intake. This is the first reported case of oral desensitization to Mitotane.

Methods: We present an oral mitotane desensitization protocol in a patient who developed pruritic maculopapular rash and angioedema to mitotane.

Results: This 18 year old patient diagnosed with adrenocortical carcinoma presented with a 5-year history of amenorrhea, hirshutism, striae and obesity. She underwent work-ups and excision biopsy on exploratory laparotomy revealed adrenocortical carcinoma. Prednisone was started and her symptoms subsequently resolved. On the 6th month post-surgery and medical treatment, there was recurrence of the same symptoms. She was restarted on prednisone and underwent another mass excision. Prednisone was discontinued. Mitotane was initiated at 500 mg tablet three times a day. After 1 week, she developed pruritic maculopapular rashes associated with angioedema and periauricular swelling within two hours of intake. She was then referred for mitotane desensitization. We used a protocol starting at an initial dose of 1/300th of the total dose doubling q 15 minutes reaching the total cumulative dose after 3 days of 1500 mg. She tolerated the procedure well with no untoward side-effects. Presently, she is still on mitotane.

Conclusion: This is the first reported successful mitotane desensitization protocol. This demonstrates that desensitization may be an option when a good alternative drug is not available in an allergic patient.

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A case of unusual insulin allergy and successful desensitisation

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Background: Immunologic responses to insulin in the form of IgG and IgE antibodies are common. Clinical problems related to these immunologic reactions are uncommon. Gell and Coombs type I, II, III and IV reactions have been reported. The antigenicity may be the result of any of several factors: insulin as a heterologous protein, altered tertiary structure of insulin, presence of non-insulin protein contaminants, or pharmaceutical formulation additives. IgE mediated local reactions are the most common and almost always subside spontaneously. With the use of human insulin these hypersensitivity reactions have become rare.

Case report: We report a Type 2 diabetic who developed an interesting combination of significant immediate cutaneous reactions and delayed Type III and Type IV reaction to all the insulins including Human and recombinant preparations. Skin tests performed with all forms of commercially available insulin and insulin analogues were strongly positive, but skin tests were negative for solvents, excipients and additives. High titre specific IgE and IgG antibodies against insulin were demonstrated against bovine, porcine and human insulin. Attempted desensitization with subcutaneous insulin- human mixtard was initially unsuccessful. An intensive desensitisation regimen with subcutaneous administration of Human actrapid followed by human mixtard over 72 hours was done with the following Modified Insulin Desensitisation Schedule

Day	Time (hours)	Insulin (Human)	Dosage (Units)
Day 1	0800	Actrapid	0.00001
	0815	Actrapid	0.0001
	0830	Actrapid	0.01
	0845	Actrapid	0.1
	0900	Actrapid	1
	0915	Actrapid	2
	0930	Actrapid	4
	1000	Mixtard	8
	2000	Mixtard	8
Day 2	0800	Mixtard	16
	2000	Mixtard	8
Day 3	0800	Mixtard	20
	2000	Mixtard	10

Results: On day 1, 0930 dose resulted in systemic urticaria after 45 minutes, the next dose was reduced to 1/3rd of the previous dose which caused the reaction and increments done by 1 units per injection. Hypoglycemia was prevented by infusing 5 % dextrose throughout the desensitisation procedure. After 96 hours patient was tolerating a Human mixtard dose 20 units and 10 units.

Conclusion: This is an unusal case of insulin allergy resluting from multiple immunologic mechanisms which responded to a intensive desensitisation regimen. Within 6 months of successful induction of tolerance there was reduction in the IgG and IgE anti – insulin titres.

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Cutaneous hypersensitivity reactions to inhibitors of cyclooxygenase-2

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Background: Most patients with cutaneous hypersensitivity to nonsteroidal anti-inflammatory drugs (NSAIDs) exhibit clinical tolerance to specific inhibitors (coxibs) of cyclooxyge-nase-2 (COX-2). However, a subset of such patients will develop urticaria or angioedema when exposed to coxibs. The mechanisms of urticaria and angioedema due to coxibs are presently unknown. In this paper we discuss the prevalence of skin reactions due to coxibs and possible mechanisms mediating such adverse manifestations.

Methods: A total of 206 patients (female 144, male 62, mean age 31.1±13.7 years) with urticaria and/or an-gioedema due to nonselective NSAIDs were studied between September 1999 and September 2005. Drug pattern showed 167 crossreactors (81%) and 39 single-reactors (19%), with a cutaneous clinical pattern in 122 (59.2%), mixed reactions (respiratory/cutaneous) in 82 (39.8%), and systemic reactions only in 2 (0.9%). Oral provocation tests were carried out with preferential and specific COX-2 inhibitors. In addition, a PubMed search of published studies on the tolerance to specific COX-2 inhibitors in NSAID-hypersensitive subjects was performed.

Conclusion: Positive oral challenges with nimesulide were observed in 31% of studied patients, with meloxicam in 20.6%, with celecoxib in 18.4%, with etoricoxib in 11.2%, with valdecoxib in 10.3%, and with rofecoxib in 9.4%. While most NSAID-intolerant patients tolerated coxibs during oral challenges, a small subgroup of patients developed urticaria. Although there are few studies for valdecoxib and etoricoxib, these results are in agreement with the majority of published studies dealing with tolerability to rofecoxib (reaction rates 0–34.7%) and celecoxib (reaction rates 0–33.3%). Cutaneous reaction rates correlated with in vitro inhibition of COX-1 by different COX-2 inhibitors. Two possible mechanisms are proposed to explain urticaria induced by coxibs, an IgE-mediated allergic response, and inhibition of COX-1.

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The importance of drug challenge in the work-up of ADR to chemotherapeutic agents

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Background: Allergic reaction to chemotherapeutic agents among pediatric patients suffering from malignancy is not common and poses an important challenge since alternative agents are not commonly available. We report pediatric cases with adverse drug reaction (ADR) to chemotherapeutic drug reactions who required further management at Siriraj Hospital, Thailand.
Methods: The medical records of children with previous ADR to chemotherapeutic agents from January 2004 through December 2006, were reviewed. They underwent allergic investigations with skin prick tests (SPT), intradermal tests (ID) and drug challenges.
Results: Eight patients (3 boys, median age 9.5 years) were documented to have ADR to chemotherapeutic agents. Their presentations were urticaria (3/8 patients) and anaphylaxis (5/8 patients). Drugs causing reactions were vincristine (3/8 patients), L-asparaginase (1/8 patients), Mesna (1/8 patients), carboplatin (1/8 patients) and cyclophosphamide (1/8 patients). Among five patients with anaphylaxis, two had alternative agents. Three other anaphylaxis patients had no alternative drug and underwent successful graded challenges (all with vincristine). Two of the urticaria patients also underwent successful graded challenges (carboplatin – 1 case and cyclophosphamide – 1 case) whereas one was switched to an alternative agent.
Conclusion: Although urticaria and anaphylaxis could develop among patients required chemotherapeutic agents, a number of these patients could tolerate a careful and successful graded challenge.

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Provocation test in patients with history of aspirin hypersensitivity

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Aim: The aim of the study was to evaluate the usefulness of aspirin provocation test in patients with a history of aspirin hypersensitivity.
Methods: Patients with asthma and/or chronic urticaria/angioedema with a history of aspirin hypersensitivity underwent provocation test with aspirin. Single-blind oral provocation test was performed with increasing doses of aspirin (placebo, 50mg, 100mg, 150mg, 300mg) administered orally. Anterior rhinomanometry, spirometry and clinical monitoring were performed before the test, during and after drug administration. Test was considered positive in case of clinical symptoms of hypersensitivity, decline of FEV1 or positive nasal response measured by anterior rhinomanometry.
Results: 62 patients (15 males, 47 females) aged from 19 to 71 (mean age: 48 years) including 26 patients with diagnosed asthma and/or nasal polyposis (first group) and 36 patients with urticaria and/or Quinke oedema (second group) were examined. All subjects had a history indicating hypersensitivity to aspirin. Provocation test was positive in 28 patients (45,1%). There was not a significant difference between the groups (first group - positive result in 12 patients (46%), in the second one- 14 patients (39%). The mean dose provoking reaction was significantly lower in asthmatics (238,3 mg) than in urticaria/angioedema group (392,8 mg); $p<0,05$.
Conclusion: Oral aspirin provocation test is a valuable tool in final diagnosis in patients with a history of aspirin hypersensitivity. Doses provoking reaction are lower in asthmatics than in urticaria/angioedema patients.

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Skin tests in Nsaids hypersensitivity

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Background: The diagnosis of NSAIDS (non-steroidal anti-inflammatory drugs) hypersensitivity relies on clinical history and skin or provocation tests. In daily clinical practice double blind placebo controlled provocation tests are not always possible and skin tests must be used first.
Methods: 245 patients with a history of rhinitis, bronchospasms, conjunctivitis, urticaria and angioedema, anaphylaxis or anaphylactoid reactions related to ingestion of NSAIDS, mainly to Aspirin (ASA) have been studied. Skin prick tests and intradermal tests when pricks were negative or dubious have been done in 215 cases. Lysine salycilate for ASA and solutions of more commonly used NSAIDS and acetaminophen have been used after discontinuing H1-antihistamines, psychotropic drugs and topical steroids. Informed consent was obtained from all the patients. Aspirin was tested in 215 patients, diclofenac in 202, ibuprofen in 156, piroxicam-meloxicam in 112, indometacin in 122, nimesulide in 186, celocoxibe in 105, etoricoxibe in 102, metamizol in 37, acetominophen in 172, naproxene in 18, niflumic acid in 2, with a total of 1430 skin tests (prick and ID when necessary).
Results: When identification of the drug was possible concordance with positive skin tests was 98%. Positive NSAIDS skin tests: ASA 197 (91.6%), Prick 65 (30.2%), Diclofenac 135 (91.6%), Prick 35 (17.3%), Metamizol 25 (67.6%), Prick 5 (3,5%), Celecoxibe 71 (67.6%), Prick 24 (22.9%), Piroxicam 57 (50.8%), Prick 21 (18.8%), Etoricoxibe 50 (49.0%), Prick 19 (18.6%), Ibuprofen 68 (43.5%), Prick 40 (25.6%), Indometacin 52 (42.6%), Prick 31 (25.4%), Naproxen 18 (33,3%), Prick 3 (16,7%), Nimesulide 50 (26.9%), Prick 24 (12.9%), Acetominophen 25 (14,5%), Prick 5 (2,9%), Niflumic acid 2-2 (both ID). Only local reactions have been observed without any severe reaction.
Conclusion: Prick tests are more specific but less sensitive than ID which can give false positive results. 3 NSAIDS groups can be considered: 1-Aspirin, diclofenac and metamizol with a higher percentage of positivity. 2-For others NSAID including the newer coxibes positivity around 50%. 3-Nimesulide and acetaminophen with a lower of positivity. Skin tests can be used in NSAIDS hypersensitivity to confirm clinical history but also to choose, case by case, a drug less prone to give hypersensitivity reactions.

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IgE mediated anaphylactic reaction to omeprazole. Cross reactivity among Proton-Pump Inhibitors

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Background: Omeprazole is a proton pump inhibitor (PPI) with an excellent therapeutic index for Helicobacter pylori infection. Anaphylactic reactions to omeprazole have rarely been reported.
Case report: A 38 year-old woman was referred to our department with pruritus in the palms, facial angioedema, oedema of eyelids and nose, generalised urticaria, bronchospasm, nausea, dizziness and collapse 15 minutes after the administration of an omeprazole capsule. No other drugs had been administered during the previous 12 hours. She was effectively treated at the emergency department. She had a past medical history of peptic ulcer for which she had been taking Omeprazole for many years. She had no past medical history of atopy.
Methods and Results: Skin prick tests with common inhalant, food allergens and other Imidazole derivatives (ketoconazole, metronidazole) were negative. The skin prick test with PPIs was positive to Omeprazole (cap 20mg/ml) and negative to Lansoprazole (cap 30mg/ml) Pantoprazole (tab 40mg/ml) Rabeprazole (tab 10mg/ml) Esomeprazole (tab 20 mg/ml). Intradermal tests were positive for Omeprazole (vial 4mg/ml, 0.004mg, 0.04mg/ml) but they have not been done for other PPIs. Five normal controls were tested and were negative for skin tests to all substances. The serum was tested for IgE Ab (ImmunoCap/Phadia) against omeprazole with experimental prototype reagents and it was negative. The challenge test with omeprazole was not carried out for ethical reasons. Also no adverse reaction occurred when the patient took the capsule shell without the pharmaceutical substance. Subsequently oral challenge in a single blind with placebo procedure was curried out with lansoprazole and rabeprazole and no reaction was reported.

Conclusion: Proton pump inhibitors are a rare cause of anaphylaxis but health care professionals need to be aware of this possibility. Skin prick tests proved to be an effective diagnostic tool for the IgE mediated hypersensitivity to PPI. An anaphylactic reaction to one of the PPIs does not necessarily imply cross reactivity with all PPIs. Oral challenge test with negative prick tests to PPIs turned out to be safe and offered the patient the chance to be treated with a different PPI with out any complication.

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Effective desensitization for allopurinol for oral way, in allergic patient. Procedure of dilution

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The desensitization with allopurinol is an effective treatment in selected patients, with cutaneous symptoms that do not respond to therapies alternatives. Clinical case. Old patient, with gout arthropathy (uric acid:11 milligram/decilitre), allergic to allopurinol, with pruritus and maculopapular rash. Associate pathologies: renal insufficiency (creatinine: 2,1 milligram/decilitre); Myelodysplastic syndrome. The therapy with altenative drugs (sulfinpyrazone, rasburicase) was not effective in reducing the level of uric acid.

Method preparation: one tablet of 100 mg allopurinol was weighed and crushed in a mortar ;1800 mg of rice starch (F.U.Italian) were added progressively in the mortar, by standard geometric diluition technique to obtain an homogeneous allopurinol dispersion in the mixed powder (total weigth 2000 mg).To obtain unit doses containing 50 mcg, 100 mcg, 200 mcg, 500 mcg, 1mg, 5 mg, 10 mg, respectively were weighed 1 mg 2 mg 4 mg 10 mg 20 mg 100 mg 200 mg from the mixed powder. To obtain unit dose containing allopurinol 25 mg, one tablet of 100 mg allopurinol was crushed in a mortar and 1:4 of one tablet was weighed. The doses have been manifactured in capsules. The successive treatment comes carried out following the outline proposed from Fam AG. In consideration of associated pathologies, in particular of the renal insufficiency, the dosage increment has been carried out every 5 gg. Employed doses: 50, 100, 200, 500 mcg, 1 mg, 5, 10, 25, 50, 100 mg. Treatment is lasted in total 60 days and cutaneous adverse reactions have not been taken place, neither pruritus symptoms. The first dosages have been given in hospital, and the following in outpatients department, with the necessary observation. We think that this procedure is easy and sure for patients with this type of pathology.

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Stevens-Johnson Syndrome from hemodialysis-associated hypersensitivity reaction in a 61-year old male

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Background: Stevens-Johnson syndrome (SJS) is often described as diffuse, severe, mucocutaneous eruption involving two or more mucosal surfaces and majority of cases are attributed to drug exposures. Hemodialysis-associated hypersensitivity reactions are well documented in literature and majority would present as flushing, urticaria, periorbital and facial edema, bronchial hypersecretion and hypotension. This is the first reported case of hemodialysis-associated SJS.

Methods: To present a patient who developed SJS after hemodialysis.

Results: This is a 61-year old male diagnosed with chronic kidney disease, in uremia. Less than 24 hours after the initiation hemodialysis, patient developed generalized, pruritic, erythematous macular rahes. This was thought to be an adverse reaction to present antibiotics thus, all antibiotics were discontinued. After the second hemodialysis, rashes progressed with increase in erythema and exfoliation. This was thought to be a an anaphylactoid reaction to ethylene oxide present in dialyzers. Thus, pre-medication prior to subsequent hemodialysis was done. But on subsequent hemodialysis these lesions became localized flaccid bullae, later developing epidermal detachment involving the eyes and oral mucosa. He received intravenous hydrocortisone up to 200 mg/kg/day (equivalent to 3mg/kg/day methylprednisolone). However, patient's condition worsened requiring endotracheal intubation. He subsequently died due to multi-organ failure. The strong history surrounding each hemodialysis sessions with the progression of his lesions plus the high index of suspicion pointed to ethylene oxide hypersensitivity as the cause of the SJS.

Conclusion: This is the first reported case of hemodialysis-associated SJS. This case report demonstrates the importance of early detection, referral, and management of hemodialysis-associated hypersensitivity reaction. Replacing ethylene oxide with steam or gamma radiation may help solve this problem.

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The impact of polypharmacy intervention on adverse drug reactions and cost of care in new orleans seniors

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Background: In the United States (U.S.), more than 100,000 deaths annually are attributed to serious adverse drug reactions (including but not limited to drug allergy). The estimated total cost of these events in 1996 was 12.2 billion (U.S.) dollars. Polypharmacy, particularly in seniors, contributes to the increased likelihood of adverse drug reactions (ADRs). Polypharmacy is defined as the use of five or more prescription medications on a long-term basis. There are no reports of intervention studies from the state of Louisiana (U.S.) where fifty percent of seniors are on polypharmacy in contrast to twenty percent prevalence in the rest of the United States. The goal of this pilot clinical observational study is to demonstrate whether cessation of inappropriate medications would decrease adverse drug reactions and lower the monthly cost of medications.

Methods: Nine patients, ages 66 to 83 years, that met the definition of polypharmacy were evaluated. Patients with known Human Immunodeficiency Virus (HIV/AIDS), transplant recipients, and malignancy were excluded secondary to known requirement of multiple medications. The Medication Appropriateness Index (MAI) was utilized to stop inappropriate medications. ADRs, Mini Mental Status Exam (MMSE) scores and monthly cost of medications were recorded before and after intervention.

Results: The mean number of medications used daily decreased from 13.5 medications to 7.3 medications before and after intervention respectively. The ADRs decreased from 3.0 occurrences before intervention to 0.3 occurrences after intervention ($p<0.001$), while MMSE mean scores improved from 25.5 prior to intervention to 28.1 post intervention ($p<0.05$). Monthly Cost of Care (COC) values decreased from 352 U.S. dollars per month prior to intervention to 128 U.S. dollars per month post-intervention ($p<0.001$).

Conclusion: Seniors on polypharmacy can be benefited by stopping inappropriate medications with intervention having a favorable outcome on mental status, frequency of adverse drug reactions and the monthly cost of medications. Further prospective studies are in process on a larger number of patients.

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Desensitisation to antituberculosis drugs in cutaneous allergy: about 6 cases

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Tuberculosis continues to be a major cause of morbidity and mortality worldwide. Currently available drugs are effective for treatment of the disease or latent infection, but may cause serious allergic reactions, often difficult to manage. Once the diagnosis allergic drug reactions have been established, re-introduction of the drug, leading to tolerance can be considered, induction of tolerance is usually possible in most patients.

We report the observations of 6 women, their age varies between 29 and 70 years, without immunodeficiency, treated for pulmonary tuberculosis TPM+ (2 patients), pleural tuberculosis confirmed histologically (3 patients) and a TPM-(1 patient) and having received an antituberculosis treatment containing SRHZ for the two first and RHZ for the last four ones with adequate posology. All our patients presented cutaneous reactions of gravity moderated at severe, which have occurred between 3 and 45 days after the beginning of the treatment. The treatment was stopped until disappearance of the lesions in all the cases. The reintroduction was practised successfully at five of our patients and the therapeutic protocol was changed in a patient considering the gravity of the lesions.

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Cutaneous adverse drug reaction induced by a generic substitute of zyloric with a residual sensitization to allopurinol

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Cutaneous adverse drug reaction (CADR) due to a generic drugs seem to be rare or have been rarely reported except with antiepileptic drugs. We report on the first case of a CADR that occurred when a drug that had been well tolerated for 15 years was replaced by a generic substitute. A 59 year old man, who had taken Zyloric* pills for 15 years developed a maculopaplar rash (MPR) 3 days after the replacement of Zyloric* pills by its generic substitute called Allopurinol Ge*. Four days after stopping the intake of Allopurinol Ge* the MPR spontaneously disappeared. Biological findings remained normal without liver involvement. Two months after, the patient took Zyloric* again and developed generalized eruption with a more severe involvement of the flexure areas, the 3 days following the re-intake of the drug. This second CADR was less severe than the previous one. Three months later, the intake of Zyloric* was disrupted and the CADR vanished in 3 days. There was no other modification in the usual treatment. Dermato-allergological investigations were performed three months after the disappearance of the CADR and after stopping Zyloric*. Drug patch and pricks tests were done.

The results were negative for both tests. To our knowledge, we report on the first case of CADR due to allopurinol, appearing after replacing well tolerated Zyloric* by one of its generic substitutes. The generic substitute was responsible for inducing a sensitization to allopurinol, which unfortunately led a relapse of the CADR when Zyloric* was reintroduced, even though the clinical eatures were less severe in the second CADR. There are no common excipients between Zyloric* and its generic. In the few reported cases in literature of CADR occurring after the replacement of a drug by its generic substitute, the adverse effect does not relapse when the original drug is readministrated. Thus, to our knowledge, here is the first case in which the reintroduction of the original drug was associated with a relapse of the CADR. From our observation, new mechanisms must be evoked for this type of therapeutic accident, i.e. the role of excipients other than those listed on the pharmacological notice or a non tolerated commercialized form of the drug that induces a sensitization to the active compound.

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The value of skin tests in the investigation of suspected systemic corticosteroids hypersensitivity

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Background: To establish the value of patch tests (PT), prick tests (pt), intradermal tests (IDT) in suspected immediate or delayed hypersensitivity to corticosteroids (CS) systemically administered.

Materials and Methods: 67 patients (52 women, 15 men, mean age 50,6 y.o) referred for immediate (13,4%) or delayed (86,6%) cutaneous adverse reaction (CAR) possibly due to a CS systemically administered, were enrolled. All had PT with a special series of 12 CS diluted in petrolatum, read at 20 minutes, 48h, 96h and one week. When negative, pt were carried out with tixocortol, budesonide, betamethasone, betamethasone dipropionate, hydrocortisone 17 butyrate, triamcinolone acetonide and the involved CS. If PT and pt were negative, IDT were carried out at least with one CS of each group of the Goossens classification (1): hydrocortisone (group A), triamcinolone acetonide (group B), betamethasone (group C), betamethasone dipropionate (group D1), with progressively increasing dosages. If skin tests were negative, oral provocation tests with the involved CS or another CS (same group or different group) were proposed. Oral administrations were performed under hospital surveillance, versus placebo, with progressively increasing concentrations to the full dosage.

Results: 12 patients (18%) had positive results either on skin testing or oral administration. PT were positive in 5 cases (7.5 %), none had positive pt, 3 patients had positive IDT, one had a positive repeated open application test. In 58 patients all tests were negative. Thirty-eight accepted one or more oral administrations (OA) of CS (50 OA were performed). Among them, 1/21 had a positive OA to the involved CS, 1/9 had a positive OA to a CS belonging to the same group; 2/20 had a positive OA with a CS belonging to another group.

Conclusion: Hypersensitivity to systemic CS is supposed to be rare but do exist, as we confirmed a CS sensitization in 12 patients with an immediate or delayed mechanism. We demonstrated that skin tests are useful in the management of such patients. They have a mean value in investigating suspected CS hypersensitivity, but they are well tolerated and seem to have a good negative predictive value (92.1 % in our series). They allow to confirm a sensitization (positive tests) and to lead the OA in the initially responsible chemical group of CS or in another group with a low risk of reactivity.

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Anaphylaxis to Omeprazole

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Background: Proton pump inhibitors are widely used but only few cases of anaphylaxis have been reported. In some cases cutaneous tests have proved useful and in only one case cytometric cellular allergen test (FAST) was performed and confirmed the existence of specific IgE.

Case report: We present a 26 year old patient whith previous history of urticaria. In one episode he associated nausea and shortness of breath. He did not correlate symptoms to food ingestions. He used ocassionally omeprazole due to acid reflux.

Methods: Skin prick tests and intradermal tests were performed with omeprazole and pantoprazole (1/100 and 1/1000 dilutions) with negative results. Controlled oral challenge was programmed. The patient started with generalized flushing and pruritus, eyelid edema, abdominal pain and shortness of breath 45 minutes after last dose of omeprazole. Physical examination did not reveal wheezing nor hypotension. The patient recovered in two hours following intravenous infusion of 500 ml of saline, an iv push of one ampoule of pheniramine and 80 mg of prednisolone. FAST, also called basophil activation test, was performed with negative result.

Conclusion: We present a case of anaphylaxis to omeprazole which could not be confirmed with cutaneous nor basophil activation tests as previously reported.

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Drug allergy in pollinosis: is the pattern changing?

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Background: We have presented at a previous EAACI meeting a study on the association between pollen and drug allergies. In this study β-lactams were more frequently associated with pollen allergy than non steroidal anti-inflammatory drug (NSAIDS). The aim of the present study is to verify if the association pattern has changed.
Methods: 952 patients allergic to pollens and 141 with hypersensitivity to drugs have been studied in the last five years. 47 patients were simultaneously allergic to pollens and drugs (4,9% of pollinosis) (33,3% of drug allergy). Clinical history, skin prick tests for pollens and prick and ID when necessary for drugs, specific IgE when possible done in all the 47 patients.
Results: 32 females (68,1%) and 15 males (31,9%), previously Females (54,4%) Males (45,6%). 40 patients (85,1%) attended by pollinosis and 7 (14,9%) by drug allergy. In pollen allergy clinics was rhinitis in 45 and asthma alone in 2. Drug allergy: urticaria-angioedema in 32 (68,8%), shock in 2, bronchospasm in 2. Skin prick tests-grass pollens 32 (68%), Parietaria 19 (40,2%), olive tree 12 (25,5%). Drug allergy: β-lactams 19 (40,4%), NSAIDS 24 (51,1%) and other drugs (16,2%). There was no correlation between allergy to one specific pollen and a group of drugs.
Conclusion: The association of pollinosis with drug allergy is not very frequent. Reporting to the results previously obtained in these series there is a greater number of grass pollens allergy instead of Parietaria and a greater incidence of NSAIDS hypersensitivity instead of β-lactams. These results can be explained by a decrease of antibiotics prescription to allergic patients and an increase in the use of NSAIDS in the last few years.

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Adverse drug reactions: a five-year experience in a tertiary care hospital (2002–2006)

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Background: Drug hypersensitivity is a common clinical problem associated with significant morbidity and mortality. According to Volchek (2004), adverse drug reactions are a major health issue encountered frequently in both the inpatient and the outpatient setting. Over the past few years, with the increasing number of medications that have been manufactured, there has been an increasing emphasis on pharmacotherapeutic vigilance in order to reduce the incidence of such reactions.
Objective: The purpose of this investigation was to review records of patients referred to for adverse drug reactions and determine the most commonly implicated drugs, most common clinical manifestations as well as to look into the source of the referral or the consult.
Methods: Chart review and frequency distribution Results: A total of 1,694 patients were referred to the Allergy Section from 2002–2006 with a slowly increasing trend each year. Forty percent of these patients were seen at the out-patient clinic and 60% were in-patient referrals. Among those admitted, the number of adult and pediatric cases were nearly the same (54% and 46% respectively). In the out-patient clinics, adult referrals outnumbered pediatric cases by more than two-thirds. The most common manifestations in the in-patient setting were cutaneous reactions (maculopapular rashes, urticaria, angioedema), followed almost equally by jaundice, difficulty breathing, fever and chills. Rare generalized reactions such as Stevens-Johnson Syndrome (SJS) and toxic epidermal necrolysis (TEN) were also reported, averaging 10 cases per year. The most commonly implicated drugs for both in-and out-patient settings was still beta-lactam antibiotics, followed by anti-tuberculosis medications, anticonvulsants, ASA/NSAIDs and others (L-asparaginase, vitamin K and allopurinol). Over the five year period there was an increasing trend of.
Conclusion: Results have shown that over the past five years, although the implicated medications are the same, there has been an increasing trend in the reporting of adverse drug reactions. This is most probably due to the fact that there is a heightened awareness regarding the issue. An increasing number of brand names and manufacturers may also lead to an increased number of adverse reactions to medications.
Key words: adverse drug reaction

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A 10-year update from the Tan Tock Seng Hospital (TTSH) prospective inpatient drug allergy reporting system (1997 – 2006)

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Background: We set-up the TTSH Prospective Inpatient Drug Allergy Reporting System in 1997 in our 1200-bed hospital.
Aim: To describe the manifestations, risk factors and outcome of drug allergy in hospitalized patients from the 10-year registry.
Methods: All newly developed cases, confirmed or suspected, of drug allergy in inpatients were notified and subsequently verified by an allergist using the network-based electronic system.
Results: From December 1, 1997 to December 31, 2006, 1094 cases were reported from a total of 443,165 admissions. Just over half (51%) of the patients were females. The majority (76.9%) were ethnic Chinese. The patients' mean age was 57 ± 20 (13–96) years with 41% aged = 65 years (elderly). Half of the patients developed drug allergy during the course of hospitalization. In the 4 weeks preceding the allergy, 56.6% had received = 5 drugs. The most commonly implicated drugs were antibiotics (57.8%) and anti-epileptic drugs (14.6%). The majority (72.9%) had delayed reactions. Cutaneous eruptions were the most common presentation (95.6%), with majority having maculopapular rash. Systemic manifestations occurred in 33.8%, of which hepatitis, haematological abnormalities, and drug fever were equally common (16% each). Serious reactions occurred in 28.4% including drug-induced hypersensitivity syndrome, (DIHS, 16.5%), Stevens Johnson syndrome, (SJS, 7.5%), toxic epidermal necrolysis (TEN, 1.5%) and overlap SJS-TEN (0.9%). Anaphylaxis occurred in 3.7%. Death occurred in 19 (1.7%) patients. Systemic involvement (p=0.00), DIHS (p=0.008), anaphylaxis (p=0.008) and SJS (p=0.001) were more common in non-elderly patients. In multivariate analysis, non-elderly age was associated with DIHS (p=0.015) and SJS (p=0.009). There were no statistically significant racial or gender differences, or differences in outcomes between the elderly and non-elderly.
Conclusion: Drug allergy occurred in 2 per 1,000 hospitalizations, with serious reactions in 28.4% and death in 1.7% of affected patients. Serious drug allergies appeared to be more common in the non-elderly with no differences in outcomes. It is possible that immune senescence may be a reason for the milder drug allergy reactions in those aged = 65 years.

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Negative predictive value of drug skin tests in investigating cutaneous adverse drug reactions

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Drug skin tests are useful in etiological analyses of cutaneous adverse drug reactions (CADR), in order to determine if the drug can be re-administered or to avoid a cross reaction with a substitute drug. The aim of this study is to evaluate the negative predictive value (NPV) of drug skin tests.
Patients and Methods: After a retrospective analysis of the files of patients referred for CADR between 1996 and 2006, we have enrolled those having CADR with clinical features, onset's delay after the drug intake and drug imputability strictly determined, having had negative drug skin tests followed by drug administrations. There were oral provocation tests (OPT) or substitution tests (ST) with a drug of the same class of those of the drug suspected during the CADR.

Results: From 1957 files analysed, 200 patients were included. After 403 patch tests, 403 prick tests and 304 IDTs, all negative, 260 OPTs and 143 STs were performed, 307 different drugs were re-administered. The 42 positive drug re-administrations were 27 OPTs and 15 STs (NPV=89.5%). The NPV of our drug skin tests were 89.6% for OPTs and 89.5% for STs. The NPV of betalactams were 95.8% % for OPTs and 86.6% for STs, for corticosteroids 100% for OPTs and 73,7% for STs.
Conclusion: Negative drug skin tests did not eliminate the responsibility of a drug in CADRs even with a low imputability. Except for severe CADRs, drug skin tests must be followed by a drug re-administration under hospital surveillance.

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Drug adverse reactions and asthma

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Background: Studies of epidemiology and clinics of drug reactions in patients attending Allergy Centers are scarce. The prevalence of drug allergy and clinical manifestations in out-patients has been analysed.
Methods: The files of 9 067 patients have been reviewed. Clinical presentation and suspected drugs registered. Skin tests with the group of drug incremated have been done by prick and intradermal techniques in progressive concentrations when prick were negative informed consent was obtained from all the patients. Specific IgE or leucotrienes release has been done in cases these techniques are available.
Results: 448 (4,9%) out of 9 067 patients reported one or more drug adverse reactions: (females 69,7%, males 30,3%). 90% of the patients reported urticaria/angiodema and 10% rhinitis and/or asthma. NSAIDS were implied in 63,6% of the patients, β-lactams in 34,2%, local anesthetics in 4,7%. Associations of reactions to NSAIDS and β-lactams were frequent (22,3%) other drug less implied were sulfa drugs, quinolones, macrolides, ACE inhibitors, angiotensin II blockers, statins, corticosteroids, vitamines, allopurinol, tetracyclin, gentamycin, furadantine, curarisers, tramadol, ranitidin, thyocholchycosid, fibrates, fluconazol. Specific IgE and CAST (leucotrienes release) were positive only in less than 50% of the cases.
Conclusion: NSAIDS and β-lactams are the more frequent cause of adverse reactions in out-patients clinical history, skin tests step by step are the more practical first diagnostic approach of drug allergy in out-patients. Drug hypersensitivity can cause asthma, allergic rhinitis or both (Fermaid Widal triad) in a non-negligible number of patients atopic or non-atopic.

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Current practice of antibiotics allergic skin testing in Korea

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Background: Antibiotics skin testing (AST) is a useful procedure for identifying patients with IgE mediated hypersensitivity to antibiotics. However the procedure is not standardized and is performed in diverse and sometimes inappropriate protocols among hospitals in Korea.
Objective: To examine protocols of AST in major hospitals of Korea and compare it with that in the literature.
Methods: Routine protocols of AST performed in general wards of 12 general hospitals were collected via allergists working in each hospital by using questionnaire. In addition, we reviewed literatures searched from Pubmed/Medline.
Results: All of the 12 allergists responded to the questionnaire. All hospitals carried out AST on volar forearm by intradermal method without any controls. Two hospitals used saline as a negative control only if false positive result was suspected. Penicillins and cephalosporins were tested in 10 hospitals and 11 hospitals. In detail, both were tested in 9 hospitals, only penicillins in one hospital, and only cephalosporins in 2 hospitals, among which penicillin was never prescribed in one. In addition, semisynthetic penicillins was tested in 8 hospitals, carbapenems in 9 hospitals, vancomycins in 4 hospitals, aminoglycosides in 3 hospitals, and monobactams in one hospital. The testing concentration differed upto 10000 folds among the 12 hospitals. Standard of interpretation also varied between hospitals. The generalized method of AST in the literatures was intradermal testing on volar forearm. AST was recommended as a routine practice for penicillins and cephalosporins but not for other antibiotics in most of the literatures.
Conclusion: The protocols of AST varied greatly among 12 general hospitals in Korea and differed from that commonly used in literatures. Thus standardized guidelines for AST are required for the safe usage of antibiotics.

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Dual fixed drug eruption caused by Paracetamol

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Introduction: Paracetamol associated Fixed Drug Eruption is uncommon and occurs in less than 1.5 percent of all fixed drug eruptions.
Case report: A 19-year-old healthy male took amoxicilin for a respiratory infection. After two days of treatment, some hours after the last dose, he suffered pruritus, erythema, pain, and local inflammation of penis, with vessels blisters and wounds of 1–2 cm. He recovered after oral corticoids, antihistamines and topic antibiotic, plus permanganate potassium washing. The wounds lasted 2–3 weeks and left skin pigmentation. On another occasion, he took paracetamol again for a sore throat. After 3–4 days of treatment he suffered similar wounds and needed the same treatment Following this, he has tolerated other NSAIDs.
Methods and Results: With written consent we made the following study: We performed a patch test with paracetamol 10% concentration that was negative at 48 and 96 hours. Prick and ID tests with paracetamol were both negative too. An oral controlled challenge test with paracetamol up to 500 mg was also negative. After this, we recommended that the patient be taken 500 mg. of paracetamol every 8 hours three more days. After two days of treatment the penis wounds appeared again and other wounds between the 1st and 2nd finger of the left hand appeared too. This wound was a 1-cm diameter, erithematous and descamative plaque. These wounds recovered after oral corticoids and antihistamines that were administered during 5–6 days.

After the challenge the patient recalled that in the first reaction he had taken paracetamol plus amoxicilin.

We performed cutaneous and oral challenge tests with amoxicilin with negative results and the patient took amoxicilin for 5 more days with good tolerance. Patch test with TRUE TEST™ and challenge tests with other para-amino drugs (sulfonamides and hydroclorotiazide) were all negative.
Conclusion: We present an infrequent adverse reaction due to paracetamol confirmed by a challenge test. It is a skin reaction suggestive of dual fixed exanthema in both penis and betwen 1st and 2nd finger of the left hand. We have ruled out cross-reactivity with other para-amino drugs. Although paracetamol allergy is infrequent, it is important to investigate and to study all the drugs involved in an adverse reaction.

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Sensitization to cefuroxim

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France; [3]University Hospital of Montpellier, Exploration des Allergies, Services de Maladies re, Montpellier, France.

Introduction: During drug hypersensitivity tests, cefuroxim, one of the most common beta-lactam used in France, is often tested either as the suspected drug or as an alternative. We report those tests and characterized patients with a positive test.

Methods: Using our Drug Allergy and Hypersensitivity Database (DAHD), we conducted an historico-prospective cohort study. All patients who consulted between 1996 and 2007 for a suspected beta-lactam hypersensitivity reaction and who underwent at least skin tests to cefuroxim were included. Diagnosis (skin tests and challenges) followed the European Network on Drug allergy (ENDA) recommendations.

Results: 650 patients [(188, 29.1% of male), 107 (16.5%) asthmatics and 355 (54.6%) atopics] were tested. 38 (5.8%) had a clinical history involving cefuroxim, 17 (44.7%) urticaria / angioedema, 10 (26.3%) exanthema and 6 (15.6%) anaphylaxis / anaphylactic shock. Only 5 (13.2%) were positive (3 skin tests and 2 oral challenges). Among those tested as an alternative (612), 178 (29.2%) were positive to at least one beta-lactam. 10 (1.6%) of them were also positive to cefuroxim (3 skin tests and 7 challenges). The most common clinical presentations were urticaria / angioedema (4, 40%) or anaphylaxis / anaphylactic shock (4, 40%).

Conclusion: 22.2% of patient with a drug allergy to beta-lactam, were positive to cefuroxim. Among those positive to the later one, 20,8% had no history involving this drug.

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An allergic evaluation of adverse drug reactions in the hospital among Thai children as previously categorized according to the World Health Organization-The Uppsala Monitoring Centre System

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Background: Confirmation of adverse drug reaction (ADR) is important for proper further drug management in patients with such reactions. The gold standard for diagnosis adverse drug reaction is drug challenge. Recently, adverse drug reactions as categorized by World Health Organization-The Uppsala Monitoring Centre (WHO-UMC) system has been suggested as a practical tool for assessing and classifying adverse drug reactions (ADR). However, confirmation of such classification by complete allergic evaluation has been not previously studied. The objective in this study is to further evaluate ADR among Thai children reported with adverse drug reactions with the WHO-UMC system by complete allergic evaluation.

Methods: Cases of ADR documented by WHO-UMC criteria between January 2005–June 2006 at Siriraj hospital, Thailand were investigated by skin prick tests (SPT), intradermal test (ID) and challenges to drugs. Serum tryptase was measured in case of anaphylaxis.

Results: Twenty-seven patients with mean age of 6.9 year were documented to have ADR by WHO-UMC criteria between January 2005–June 2006. These patients were classified to have causality terms as certain in 4/27, probable in 6/27, possible in 10/27 and unlikely in 7/27 cases. Antibiotics were the most common drugs causing ADR in this group of patients (44.5%) which sulfonamides and beta-lactam drugs being the common culprits (18.5% and 14.9%, respectively). Skin tests and drug challenges were performed in 26 patients. Serum tryptase were measured in 3 patients with drug-induced anaphylaxis. Drug challenges and serum tryptase were positive in 8 and 1 patient, respectively. Overall, cases of certain and unlikely drug allergy were identified in 9/27 and 18/27, respectively. By performing drug challenges, the sensitivity and specificity were changed from 70% to 100% and 88.2% to 100% , respectively.

Conclusion: Skin tests, drug challenges and serum tryptase are helpful to further confirmed adverse drug reactions among patients categorized by WHO-UMC system.

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Adverse cutaneous reactions due to glycopeptides-induced adverse cutaneous reactions: results of immunoallergical investigations in 9 patients

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Objective: With the emergence of hightly resistant staphylococcus, vancomycin (V) and teicoplanine (T) usage has considerably increased but they can induce cutaneous adverse reactions (CAR) for which the value of immunoallergical investigations has not yet been determined.

Methods: Retrospective study (2006–2006) including 9 patients (6 women, 3 men; mean age = 54.3) with CAR suspected to be due to glycopeptides, V: 4 cases, T: 4 cases and successively V then T: 1 case. Red man syndromes were not included. Imputability was estimated according to Moore's criteria. The CAR were 6 maculopapular rashes, 1 linear bullous dermatosis, 1 DRESS and 1 urticaria. Six weeks later, according to ESCD's guideline for drug skin tests, immunoallergological investigations were carried out with successively drug patch tests, prick tests and intradermal tests with all the drugs taken during the CAR.

Results: Skin tests confirmed the responsibility of glycopeptides in 4/9 cases, with cross-reactivity between V and T in 2/5 cases. Following negative immuno-allergical investigations, 3 patients had well tolerated provocation tests with glycopeptides.

Conclusion: In literature, T seems to be safer than V (ototoxicity, rashes, nephrotoxicity) for a similar efficacy, inducing very few hypersensitivity reactions such as red man syndrome (3 cases published). Allergic cross-reactivity (V and T), already reported in literature, is not so rare (2 of our patients) but unconstant (3 of our patients had indeed well tolerated provocation tests with the other glycopeptide and we precise that all the skin tests were negatives for V and T in these patients. In that way, this study has important clinical implications, demonstrating that skin tests are useful in CAR due to glycopeptides, in determining the responsible drug and if the tests are negative, allowing the readministration of another glycopeptide with hospital surveillance, as we demonstrated that cross-reactivity between glycopeptides is not constant.

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A link between angioedema caused by inhibitors of angiotensin converting enzyme and eotaxin and ECP

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Background: Inhibitors of angiotensin-converting enzyme (iACE) and Blockers of angiotensin II type 1 receptor (AT1 blockers) are commonly used in hypertention and chronic heart failure treatment. These therapies are believed to be safe. However, angioedema is a rare but a well recognized potentially life-threatening adverse effect of iACE therapy. The aim of the study was to estimate angioedema threat in patients treated with short and long acting iACE in comparison to patients receiving AT1 blockers. Eosinophilic cationic protein (ECP) and eotaxin serum concentrations were used as indicators of the angioedema threat, since bradykinin is involved in the activation of this chemokine and subsequently eosinophils.

Methods: Patients were divided into the following groups, according to the treatment they have been receiving: AT1 blockers, short acting iACE, long acting iACE and control. Patients with a history of angioedema of etiology unrelated to the study were excluded. Eotaxin concentration in the serum was determined by standard solid phase sandwich ELISA. ECP level in the serum was assessed by the Pharmacia CAP system ECP fluoro enzyme immunoassay (FEIA) method, according to the manufacturer's instructions. Results are shown as mean ± SD. Student's t test for unpaired samples was used for statistical analysis.

Results: There were no statistically significant difference with regard to serum ECP level between the AT1 blockers and control group. The same was true for eotaxin. The levels of ECP in sera of patients taking short acting iACE (12.9±3.30 μg/mL, $p<0.001$) and long acting iACE (10.8±4.9 μg/mL, $p<0.05$) were significantly higher than in control group (5.22±2.5 μg/mL). The level of eotaxin was higher in serum of patients taking short acting iACE (177.5±57.6 pg/mL, $p<0.05$) and higher in serum of patient taking long acting iACE (156.1±44.8 pg/mL, $p<0.05$) compared to the control group (112.3±21.5 pg/mL).

Conclusion: Serum concentration of eotaxin and ECP play potential role in the pathogenesis of iACE induced angioedema and their concentration may be a predicting factor of this adverse event. Thus, patients treated with AT1 blockers are at the significantly lower risk of angioedema in comparison to patients taking iACE.

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Fixed drug eruption caused by traditional herbal medicine (jamu)

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Background: Fixed drug eruption (FDE) are so named because they recur at same site with each exposure to the medication. *Jamu* is the term commonly used to describe traditional herbal medicine products from Indonesia. The medicinal elements may be made from roots, bark, leaves, fruits, flowers, seeds or other parts of a plant. *Jamu pegal linu* indicated for the treatment of rheumatism, body ache and bone ache, muscle joint, giddiness, toothache and backache usually consumed by hard workers. Commercial products are available in the form of powders, pills, capsules, mixtures and creams. Some illegal retailers adding phenylbutazone, allopurinol and paracetamol in to *jamu*. FDE caused by *jamu* has never been reported yet.

Case: A 40-year-old man presented with multiple black macules on the upper left eyebrow, left and right cheeks, lower lip and left and right hands. The lesions initially appeared small and only a few, enlarge slowly, and became obvious and of concern to the patient in the past 2 weeks. The lesions were slightly itchy but there was no pain. He was otherwise healthy. He denied taking any medication except traditional herbal medicine (*jamu*) for the past 1 year when feeling tired. Family history was negative. Laboratory examinations were within normal limits. Skin biopsy from one of his lesions showed some vaculolar degeneration of the basal cells, pigmentary incontinence, mild superficial perivascular limpho-histiocytic infiltrates with a few eosinophils.

Discussion: The clinical presentation and histopathologic alterations are consistent with the diagnosis of fixed drug eruption. The patient was treated

with oral methyl prednisolone and topical hydrocortisone 2,5% for his face and also topical desoximetasone 0,25% for his body. His condition improved and the lesions disappeared in 6 weeks.
Conclusion: This was the first case of Fixed Drug Eruption caused by traditional herbal medicine (*jamu*). Successfully treated with oral methyl prednisolon and topical hydrocortisone for the face and topical desoximetasone for the body.
Key Words: fixed drug eruption, jamu

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Anaphylactic reactions during general anaesthesia: diagnostic approach and prevalence of anaesthetics involvement

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Background: Although anaphylactic reactions during general anaesthesia show a very low incidence, the mortality reaches 5% or more, and additional patients may experience significant brain damage. Neuromuscular blocking drugs are usually reported as the most common cause of anaesthesia-elicited anaphylaxis, but the possible involvement of induction agents, antimicrobials, latex, plasma expanders and other drugs should be considered. Unfortunately, the diagnostic approach is limited by the lack of validated commercial kits for detection of specific antibodies, as well as by the occurrence of pseudoallergic reactions, that are not mediated by circulating IgE.
Methods: We evaluated 20 consecutive cases (14 F, 6 M; age range 16–68 yrs) of well documented anaphylaxis during general anaesthesia, by laboratory assays (WBC count; Eosinophil cationic protein; Serum Tryptase; Plasma C4 and C1-INH; IgA and total IgE levels; RAST for inhalant and food allergens, latex and beta-lactams) and skin tests (Patch and/or Prick testing; intradermal injection) for inhalant allergens, eggs, soybeans and exotic fruit, latex and drugs administered before or during anaphylaxis onset. Drug skin tests were performed with commercial formulations of beta-lactams, heparin, atropine, benzodiazepins, vecuronium, norcuronium, mivacurium, rocuromium, fentanyl, remifentanyl, propofol, droperidol and thiopental, at different dilutions.
Results: Two patients showed evidence of allergy to norcuronium and mivacurium, respectively. Three subjects were allergic to amoxicillin and six to latex. Heparin skin testing elicited a local weal-and-flare response in one patient, whilst diffuse urticaria and itching were observed in one case. Finally, apparent anaphylaxis followed by cardiac arrest during anaesthesia in an asthmatic subject, seemed to be an adverse effect of a difficult intubation rather than a true allergic or pseudoallergic reaction. Unfortunately, in the remaining six patients we were not able to identify any triggering factors, also because no diagnostic tests are available to assess NSAIDs hypersensitivity.
Conclusion: Latex allergy still represents, at least in our experience, the main cause of anaphylaxis during general anaesthesia. Although the reliability of skin testing with commercial drugs has not been well established till now, this procedure can help to prevent the risk of further episodes in patients with a documented anaphylactic reaction during anaesthesia.

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Four cases of steroid allergy with bronchial asthma

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Background: Corticosteroid preparations have anti-inflammatory and immunosuppressive properties and are widely used in the treatment of asthma and allergic disorders. However, steroids themselves can rarely induce hypersensitivity reactions. The severity of the reaction can vary from a rash to anaphylaxis or death. We report four cases regarded as the steroid allergy that we experienced in our hospital recently.
Case Report: Case 1 was the 9 years old girl and hospitalized with moderate asthma attack. Rash developed in several minutes when she was given aminophilin continuous intravenous infusion and hydrocortisone succinate (HC-S) 6mg/kg intravenous drip. An intradermal test by HC-S was positive and we diagnosed the drug allergy. We changed it to predonine (PSL) intravenous infusion, rash didn't develop but persistence of wheeze and decreased FEV1.0. The symptom and lung function were improved when all steroid were canceled. Case 2 was 10 years old girl with many kinds food allergy and hospitalized with moderate asthma attack. She was given aminophilin continuous intravenous infusion, hydrocortisone succinate (HC-S) 7mg/kg intravenous drip and PSL 1.0mg/kg$B!_#4(B/day intravenous injection for 8 days. She showed persistence of wheeze and decreased$B!!(B-FEV1.0. She was diagnosed steroid allergy and pertussis infection. By an intradermal test, she showed positive for PSL, HC-S, methylprednisolone succinate(MP-S), hydrocortisone sodium phosphate. Wheeze and FEV1.0 were improved by all steroid cancellation. Case 3 was a 23 years old female patient hospitalized with moderate asthma attack. It was similar to case 2, she had intractable wheeze and decreased FEV1.0 persisted. By an intradermal test, she showed positive for PSL, HC-S and she was diagnosed steroid allergy. Case 4 was a 10 years old boy with moderate asthma attack developed urticaria and hypotension after receiving HC-S intravenous drip. In this patient the intradermal tests HC-S and PSL showed positive and diagnosed steroid allergy.
Conclusion: 3 in 4 cases did not show anaphylactic reaction. There were many cases which showed persistence of wheeze and decreased FEV1.0 during treatment of asthma. We thought that the steroid allergy that took the course to protract of asthma attack could be present from these cases. It is important to considerate striod allergy in treatment of acute exacerbation of intractable asthma.

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Angioedema after administration of methylprednisolone to treat drug allergy: a case report

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Background: Despite the large use of corticosteroids (CS), allergic reactions to these drugs remain uncommon; Immediate allergic reactions to CS are uncommon events usually provoked by excipients. Both immunologic and nonimmunologic mechanisms are proposed. Delayed reactions are compatible with a T-cell mediated response. Cross-reactivity has been observed between different topical corticosteroids, but the classification of cross-reactivity between them does not seem to be completely useful in adverse reactions to systemic corticosteroids. Case report. We signal the case of a 39-year-old woman with no atopy but contact sensitization to tixocortol-21-pivalate 1%. The woman, before a recent administration of mepivacaine for dental surgery, had proceeded with CS preventive administration (Prednisone 50mg 13hh, 7 hh and and clorfenamine 1h before the intervention). One hour after the last dose of premedication, followed by anahestetic administration, patient presented facial angioaedema. Further administration of metilprednisolone to treat the adverse event had the effect of increasing symptoms.
Management: Patient was studied for corticosteroids allergy. Patch test, performed with Standard Series, confirmed mentioned sensitization. Skint tests were carried out with metilprednisolone succinate, betametasone fosfate, triamcinolone acetodine. Patient showed immediate local reaction after intradermoreaction with metilprednisolone; no positivity for other CS tested.

Comments: Tixocortol-21-pivalate and prednisone belong to group A corticosteroids, so a possible cross reaction may be suggestive. As CS are poorly soluble in water or saline, they are coupled with esters. In this circumstance patient reacted to both the previous administration of prednisone and then to metilprednisolone; these two preparations are different only for the presence of succinate ester in the second one, so an effect of this additive may reasonably be excluded. Therefore all other excipients included in the tested preparations were commonly existing in both tolerated and untolerated drugs.
Conclusion: Drug adverse reactions appearing altough preventive administration of systemic CS should be reconsidered for possible allergic response to CS. The classification of cross reactivity between CS, with patch testing and intredermal testing, may provide useful support to confirm diagnosis and to identify alternative tolerated molecules.

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Erythema multiforme

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Background: Erythema multiforme is a reaction pattern of blood vessels in the dermis with secondary epidermal changes and manifests clinically as characteristic erythematous iris-shaped papular, little or no mucous membrane involvement; vesicles but no bullae or systemic symptoms. Eruption is usually confined to extremities, face, classic target lesions.
Case: A 56 year-old girl with erythema multiforme minor developed typical lesions involving most of the skin. The eruptions developed after she had taken some medicines for antihypertension. Histopathology result showed focal hyperkeratosis, acantosis, focal spongiosis in epidermis and inflammatory cell infiltrate: lymphocytes and eosinophil in dermis without vacuolar degeneration.
Discussion: Mild forms have appearance of little or no mucous membrane involvement, vesicles but no bullae or systemic symptoms. In this case there were erythematous patchs and vesicles in his skin. This eruption is usually confined to extensor aspects of extremities, and classic target lesions. Clinical manifestation characteristic by classic target lesions that characterized for erythema multiforme but histopathology examination showed appearance like drug eruption without vacuolar degeneration, that mean there is discrepancy of clinical appearance and histopathology. Mild form of erythema multiforme may developed to severe erythema but in this case the lesion improved after treatment with steroid equivalent of dexamethasone 30mg orally a day in divided doses.
Conclusion: There is discrepancy of clinical appearance and histopathology. Erythema multiforme in this patient well responded to corticosteroid, withdrawal of all possible offending drugs, and supportive therapy. Prompt diagnosis and treatment give good result.
Keywords: erythema multiforme, mild forms, classic target lesions, vacuolar degeneration.

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Severe reaction due to fluoroquinolones - Toxic epidermal necrolysis case report

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Background: Toxic epidermal necrolysis (TEN) is an uncommon fulminating mucocutaneous disease associated with high prevalence of mortality. Frequently TEN is associated to drug administration. TEN due to fluoroquinolones are rare, 7 case reports in Medline with TEN due to ciprofloxacin. Allergic reactions due to fluoroquinolones are very rare. Anaphylactoid and/or anaphylactic reactions are estimated to occur in 0.46-1.2 in 100,000 patients. Fluoroquinolones are new antibiotic drugs used mostly in treatment of urinary and respiratory infections. Allergy to fluoroquinolones may have many clinical manifestation: anaphylactic or anaphylactoid reaction, acute urticaria with or without angioedema, fixed drug eruption, serum sick-like illness, nummular eczema. The most frequent used quinolones in Romania are ciprofloxacin and norfloxacin. 2812 patients were evaluated in the department of Allergy, in Cluj Napoca between Jan 2005–Dec 2006. Only 14 (0.49%) cases of allergic reaction due fluoroquinolones were reported.
Case report: A 29-years-old smoking man was admitted in our hospital with malaise, with an eritematous eruption in the whole skin with small areas of denudation of epithelium, ulcers in nasal and oral mucosa. Those phenomena were associated with a severe itching and burning sensation of the skin. The patient began 10 days ago treatment with ciprofloxacin for an acute bronchitis, that was the only drug he took in the last 6 months. After a complete examination the patient presented also hepatocitolisis. After treatment with corticosteroids, antibiotics intense care of the skin the patient recover complete. Evolution in 2 months was "restitutio at integrum".
Conclusion: Toxic epidermal necrolysis (TEN) is a rare, but the increasing rates of reports for this syndrome due to ciprofloxacin could reconsider first choice option for this drug. More studies should be done in order to find the prevalence of TEN to ciprofloxacin.

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Toxic epidermal necrolysis suspected due to nimesulide

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Background: Nimesulide is a new nonsteroidal anti-inflammatory drug with antipyretic and analgesic properties. Pruritus and rash are the most common cutaneous side effects associated with nimesulide. Toxic epidermal necrolysis induced by drugs is a rare, potentially lethal syndrome characterized by the sudden onset of widespread blistering of the skin. To the best of our knowledge there have been no previous reports of toxic epidermal necrolysis induced by the ingestion of nimesulide in the medical literature.
Case: A 68 years old woman, Buginesse. Previous history she ever had TEN six years ago probable caused by paracetamol and metamphiron. The diagnosis was established based on anamnesis and clinical manifestations. The lesions consisted of generalized erythema, vesicles, bullae, Nikolsky sign was positive, erosions, and epidermolisis more than 30%. Treatment was parenteral infusion, dexamethasone intravenous 10 mg, cimetidine 1 ampule, ciprofloxacin injection per drip and topically with silver sulfadiazine and framycetin sulfate for erosion and denuded area.
Discussion: This case was diagnosis as toxic epidermal necrolysis based on anamnesis and clinical manifestations. The most frequent cause is drugs, 80% of cases have strong association with spesific medication. The probable cause was nimesulide. Clinical manifestations toxic epidermal necrolysis are extensive epidermal necrosis, and exfoliation of the skin. There is almost no normal skin seen and in this form over 80% on the skin surface is involved. Day 10th of hospitalization death ensued from sepsis and multiple organ failure.
Conclusion: Toxic epidermal necrolysis is presumed to be a spesific immune response, a recurrent exposure to the trigger should induce a more brisk and intense (and in this case fatal) episode Multiple organ failure are the usual

causes of death. Nimesulide should be added to the list of agents associated with this serious adverse drug reaction.
Key words: toxic epidermal necrolysis, nimesulide

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Stevens Johnson Syndrome in Bandar Lampung, Indonesia

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Background: Stevens Johnson Syndrome (SJS) is severe form of erythema multiforme with related mucocutaneus disorder, often with severe constitutional symptoms and associated high rate of mortaliy and morbidity.
Subject: To find out the incidence, etiology, treatment, lenght of stay and complication.
Methods: A 3 years retrosprctive study (January 1, 2004 through December 31, 2006) of patients admitted to dermatology department of Dr. Abdul Moeloek Hospital Lampung.
Results: Among 24 cases reviewed there were 11 (45,8%) male and 13 (54,2%) female. The youngest patient was a 10 years old girl and the oldest one a 54 years old female. We assume that the caused were antibiotic (penicillin derivate) 10 (41,6%), analgesic/antipyretic 8 (33,3%), anticonvulsant (carbamazepin) 3 (12,5%), and 3 (12,5%) patient unknown. There was a variety in the length of stay of hospitalization from 1 until 27 days with an average of 7,2 days.

Systemic corticosteroid was the of choice against fatality, 19 (79,2%) recovered, 5 (20,8%) patient died, the cause of death were 3 (60%) broncopneumonia, 1 (20 %) septicemia and 1 (20 %) gastrointestinal bleeding.
Conclusion: The incidence of Steven Johnson Syndrome in Dr. Abdul Moeloek Hospital was found female is bigger than male, the highest group of age was 24–44 (54,1%) and the drugs most commonly involved were antibiotics (41,6 %) followed by analgesic/antipyretic (33,3%), broncopnemoni was the mostly cause of death (60%).

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Stevens Johnson syndrome suspected due to sulfasalazine

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Background: Stevens Johnson Syndrome (SJS) is a diffuse, severe, mucocutaneous eruption involving two or more mucosal surfaces with or without visceral involvement most often induced by drugs and occasionally elicited by infections. Sulfasalazine is a sulfa drug. The metabolite of sulfasalazine may trigger an immune-response leading to cutaneous adverse drug reactions.
Case: A 29 years old woman presented symptoms of skin, mucous membrane and eye lesions. She had taken sulfasalazine for several days before the eruptions. Physical examination revealed erythema, and blisters affected on the face, neck, arms, upper trunk, erosions of the lips, and conjunctivitis. Histopathology examination of skin lesion showed Toxic Epidermal Necrolysis (TEN) appearance. The patient was treated with ringer lactate, dexamethasone 5 mg per 6 hours, gentamycin 80 mg per 12 hours intravenously, and topical steroids. After being hospitalized for 14 days, the patient cured without complications.
Discussion: This case is diagnosed as SJS based on anamnesis and clinical examinations, confirmed by histopathology finding subepidermal bullae with necrosis of entire epidermis and inflammatory infiltrate, according to TEN appearances. This case may be a degree of overlap as forms of SJS can evolve into TEN. Sulfasalazine is a sulfa drug, used primarily as an anti-inflammatory agent. SJS due to sulfasalazine is rare.
Conclusion: SJS due to sulfasalazine is rare. SJS is a life threatening cutaneous adverse drug reaction and can evolve into TEN. Early appropriate treatment may produce an excellent prognosis in SJS.
Key words: disparity SSJ-TEN, sulfonamide, corticosteroid

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Analysis of Stevens-Johnson syndrome and toxic epidermal necrolysis in Japan

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Background: (B Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN) are the representative severe cutaneous adverse drug reactions with high mortality, in which the treatnent has not been established.
Methods: To present the current clinical characteristics and treatments of SJS and TEN in Japan, we retrospectively analyzed the 43 cases of SJS (15 males and 28 females, aged 44.8 in average) and 54 cases of TEN (27 males and 27 females, aged 45.5 in average) from reports of SJS and TEN published in medical journals from 2000 to 2005.
Results: Thirty cases of SJS (69.8%) and all cases of TEN were caused by drugs, such as nonsteroidal anti-inflammation drugs (NSAIDs), antibiotics, and anticonvulsant drugs. As the organ involvement, hepatitis was most common in both of SJS and TEN. Renal dysfunction and respiratory disorder were also involved in some cases. Major complication was sepsis. As the treatment, corticosteroid was administered systemically in most cases, and 26 cases (60.5%) of SJS and 32 cases (59.3%) of TEN were treated with corticosteroids alone. Combination of plasmapheresis and/or immunoglobulin therapy with corticosteroid therapy was performed in some cases. The mortality rate of patient with SJS was 2.3%, and with TEN was 7.4%. The mortality in TEN decreased remarkably as compared to 21.6% (58/269) in the previous 17 years from1981 to 1997. The serious patients with TEN were performed plasmapheresis and high-dose immunoglobulin therapy in combination with steroid therapy and they were alive.
Discussion: These results suggest that the combination of the above-mentioned 3 treatments might be useful in the serious patients. More experiences is needed to confirm the effect.

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Adverse effects of DPT immunization in children up to 6 years of age in Tabriz/Iran

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Background: Vaccines, once hailed as "magic bullets" against disease, sometimes fly astray. In fact, shots of common vaccine DPT (diphtheria-pertussis-tetanus may in rare cases trigger serious health problems. In reviewing existing data, the several studies found a link between the pertussis component of the DPT vaccine and anaphylaxis, a life-threatening allergic reaction. Vaccines have been highly effective at eliminating or significantly decreasing the incidence of many once-common diseases. However, the public is now focusing on the potential adverse effects of vaccines. The aim of this study was to identify the adverse effects of DPT immunization in children up to 6 years of age in Tabriz/Iran.
Methods: A descriptive cross-sectional survey was conducted in the city of Tabriz in North-West Iran. Cases were defined as those notified with Adverse

effects of DPT immunization over a 6 months period (between September 23, 2006- to March 20, 2007) in Tabriz health centers. All of the children aged 2 months up to 6 years who recived DPT vaccine in our selected health centers, were carefuly checked up to 72 hours following immunization. A questionnaire was used to elicit information related to Adverse effects of DPT immunization in children. The data was anayzed by software of SPSS/Win.

Results: The findings indicated that fever, limb swelling, unusual crying, irritability, vomiting, erythema were 47.8%, 42%, 37%, 25%, 13%, 11.8% respectively.

Conclusion: As immunizations are usually given to healthy people for prevention of serious diseases, the risks for known adverse reactions to the vaccine must be weighed against the potential benefits. Nurses and health providers should be prepared to answer patients' or parents' parents questions about common as well as alleged vaccine-related adverse events and must utilize the Vaccine Information Statements for vaccines covered under the National Vaccine Injury Compensation Program.

Also health care providers should continue to counsel parentss about the benefits of the vaccine and recommend vaccination for clients.

Key Words: Adverse effects, DPT, Immunization, Children up to 6 years.

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The anticonvulsant hypersensitivity syndrome

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Background: The anticonvulsant hypersensitivity syndrome is a potentially fatal multisystemic reaction to anticonvulsant medications.

Objective: The purpose of this study was to investigate the clinical characteristics of anticonvulsant hypersensitivity syndrome.

Results: A total 32 subjects, aged from 6 to 72 years, diagnosed as having anticonvulsant hypersensitivity syndrome based on clinical and histopathological findings, were included in the study. In 22 of the 32 cases, the anticonvulsants had been administered prophylactically after craniotomy and in 10 cases for epilepsy. When the cases were assessed for skin lesions, maculopapular eruption was registered in 22, Stevens-Johnson syndrome in five, and toxic epidermal necrolysis (TEN) in five. Treatment included suspension of the offending drug and then, except for the cases with toxic epidermal necrolysis, administration of corticosteroids. The 22 cases that required anticonvulsant therapy were treated with valproic acid. In all cases, we observed rapid clinical improvement corroborated by laboratory findings.

Conclusion: It is essential that due importance be given to the development of an eruption in individuals to whom anticonvulsants are administered after craniotomy because anticonvulsant hypersensitivity syndrome is likely to be life-threatening.

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Drug allergic reactions related to the vitally important drugs - medical tactics

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Background: Generally accepted practice of treatment of DA is a primary cancellation of "guilty" drugs. At the same time in some cases, we face with such events, when cancellation of the given drug can bring to unavoidable consequences. Therefore, there is a dilemma between the danger of cancellation syndrome and the development of heavy manifestations of the drug allergy.

Aim: To describe the running tactic of patient with a diabetes mellitus with the concomitant polyvalent DA related to a number of drugs.

Methods and Results: A patient 62 years old is continuously receiving human insulins. This patient has in anamnesis such episodes of allergic reactions related to a number of drugs of different pharmaceutical groups. The patient got a heavy allergic dermatitis to an Isosorbide in a dripping form. The cancellation of this drug and the prescription of antihistamines didn't improve the situation. The patient examination with analyzer UniCap-100 brings to light an existence of specific IgE c73 substances (Insulin Human). The abolition of this drug was impossible due to the danger of decompensation of the diabetes. Tests in vivo were impossible in consequence of the activity of cutaneous process. That's why the client passed the Damage Measure of Neutrophils Test (DMNT) in the modified scheme. The hypersensibility related to a number of drugs was displayed. From such types of insulin that obtained negative results Insulin NPH and Insulin Rapid were administrated. Parallel to this, the patient was prescribed to take a intravenous Prednisolone (120 mg/day for 5 days), after that, the treatment was transferred to steroids per os in the dose of 10 mg/day for one month with the gradual dose decrease and the following cancellation. Glycemic profile had normalized and effects of heavy allergic dermatitis had regressed completely.

Conclusion: In the case of the appearance of the DA on the vitally important drugs, the cancellation of the given drug is contraindicated. It's because, in the case of further necessity of emergency infusion, the risk of anaphylactic shock is increasing. The DMNT (sensitivity is about 95%, specificity- about 80%) in the modified scheme is the effective method due to the impossibility of conduction of more trustworthy tests in vitro and in vivo. Monitoring showed the effectiveness and safety of the prednisolone course for the treatment of DA.

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Connubial systemic cutaneous adverse drug reaction

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Observation: A 86 y.o. women has had for 24 hours, an itchy maculopapular exanthema (MPR) on the trunk and the arms. The daily treatment had not been modified, except concerning the switch from MOPRAL® (omeprazole) to INIPOMP® (pantoprazole) 48 hours before. The biopsy sample was compatible with a drug allergy. There was an eosinophilia (1296/mm3). In her history, 11 years before, cutaneous tests were performed for calcium channel blockers and penicillin sensitizations. Patch tests were positive for diltiazem. The chronology of drug intake was done with his husband. Her treatment was prepared by his husband because she had visual deficiency. A few days after the MPR's onset a member to the family reported that 48 hours before the MPR the patient had taken by error the drugs of her husband put beside those of its wife: that were: trimetazidine, diltiazem, atenolol, lysine acetylsalicylate, domperidone and oxybutynine. The rash vanished within 3 weeks.

Discussion: This observation stresses the importance of the interrogation in the assumption of a CADR in determining the chronology of drug intake. An additional difficulty is to obtain this information from an old person (lack of memory). The interrogation of the other family members, the GP and the pharmacist can be crucial.

Masked drug exposure have been reported by kissing, but not by the misadministration of drug intended for a spouse. The risk could become significant because of the ageing of the population.

Conclusion: 1st case masked drug allergy due to diltiazem taken by mistake (intended for a spouse).

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Drug hypersensitivity syndrome

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Background: Drug hypersensitivity syndrome (DHS) is an idiosyncratic and life-threatening adverse drug reaction that begins acutely in the first 2 months after initiation of drug and is characterized by fever, skin rash, facial edema, or an exfoliative dermatitis, lymphadenopathy, hematological abnormalities (eosinophilia) and multi-organ involvement. In this case, there were no other systemic organ involvement.

Case: A 26-years-old woman presented with fever, maculopapular rash, facial edema and lymph nodes enlarged 2 weeks after taking cefadroxil and mefenamic acid after fine needle aspiration biopsy (FNAB) for suspect thyroid infection. After taking mebhydrolin napadysilate the rash become extended as an exfoliative dermatitis, facial edema, and lymphadenopathy. Laboratory examination including leucocytosis and eosinophilia. There were no other biologic abnormalities including normal liver function test, chest radiograph and cardiac echography. Histology examination showed hyperkeratosis, mild spongiosis, vacuolar degeneration of basal cells with infiltration of lymphocytes, neutrophil and eosinophil.

Discussion: The patient fulfilled the clinical criteria with her presentation of fever, generalized maculopapular rash, facial edema, lymphadenopathy and eosinophilia. The pathogenesis of DHS is not fully understood, including genetic background, altered drug metabolism, and virus infection. The suspect drug was mebhydrolin napadysilate. The treatment was dexamethason intravenously started with 15 mg tid. Topical steroid and emollient were given also. After 3 weeks, dexamethasone intravenously was substitute with methyl prednisolone 24 mg orally a day and tapered. The rash progressively improved.

Conclusion: Drug hypersensitivity syndrome is a severe skin reaction with systemic manifestations. Internal organ involvement can be asymptomatic. The suspect drug was mebhydrolin napadysilate. The treatment of dexamethasone intravenously continued with methylprednisolone oral and withdrawal of possible offending drug gave a good result.

Keywords: drug hypersensitivity syndrome, mebhydrolin napadisylate, corticosteroid

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Drug hypersensitivity

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Background: Drug hypersensitivity reactions are common and can be life threatening. Drug allergies are adverse reactions resulting from immunologic responses to drugs or their metabolites. These reactions result in predictable patterns of organ-specific or systemic hypersensitivity that usually recur on subsequent exposure to the same drug. The objective of this study was to evaluate the incidence of drug hypersensitivity in Sina Hospital in Tabriz University of Medical Sciences March 2002 to March 2007.

Methods: This descriptive study was done retrospectively by reviewing the existing records of 51 drug eruption diagnosed patients admitted in skin diseases ward of Sina Hospital.

Results: 51 patients (17.65 % male and 82.35 % female) have entered our study. Mean age was 36.19 years. Drug eruption was the most common cause of admission (52.94 %) other clinical diagnoses of patients were Angioedema (15.68%) and Stevens Johnson syndrome (21.56%). The most drugs were Co-trimoxazole (89.80 %) and Carbamazepine (17.64%). The following Symptoms are considered: rash, fever, generalized erythema and muculopapular lesions.

Conclusion: Early recognition and appropriate treatment of these underlying causes is recommended in order to prevent drug eruption. With special examination and drug history we can diagnosis this hypersensitivity and with this knowledge we can prevent the dangerous reactions.

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A 63-year-old man with multiple drug allergy and CNS tumor

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Background: Adverse reactions to pharmaceutical and diagnostic products constitute a major hazard in the practice of medicine. Multiple risk factors have been shown to modify the clinical expression of immunologic drug reactions. These can be associated with the drug itself, with the disease state in which the drug is used(chronic disease and infections), or the individual patient receiving the drug. Similarly, some allergic drug reactions are much more likely to occur if the drug is administered to patients with particular disease backgrounds, as when aminopenicillins are given to patients with atypical or abnormal lymphocytes or sulfonamides to patients with AIDS.

Case Report: We represent a 63-year-old man with prolonged history (more than one year)of intermittent headache and use of NSAIDS. He had recurrent history of anaphylaxis and urticaria after getting NSAIDS at first, and with the other analgesics and also antihistamines there after. His laboratory evaluation, includind cell blood count and differentials and complement assay was in normal limits, He consulted with a neurologist, and brain CT-scan was done. It showed CNS tumor. In literature review it was found a few chronic diseses such as cyctic fibrosis or cryogobulinemia (or also immune deficiencies) can cause drug allergy but there was no association between multiple drug allergy and malignancies especially CNS tumors.

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Dapsene hypersensitivity syndrome in leprosy

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Background: Dapsone Hypersensitivity Syndrome (DHS) is a rare well recognized serious adverse effect characterized by fever, skin rashes, generalized lymphadenopathy, hepatitis and hepato-splenomegaly. Hipersensitivity reaction occur during first 6 weeks, but can occur after 6 months.

Objective: To find out the total cases for 2 years of dapson hypersensitivity syndrome in the wahidin sudirohusodo general hospital, Makassar, Indonesia.

Methods: A retrospective study was carried out taken from medical record and registration book. (January 2005- December 2006).

Results: Five patients with dapson hypersensitivity syndrome were reported. All patients received dapsone as a part of MDT for leprosy. Fever, jaundice, skin lesion were detected in 5 cases, but 1 case appearance like morbiliform. Lymphadenopathy (2 cases), splenomegaly and acut psychosis (1 case). Laboratory data on admission included : low hemoglobin, leucocytosis, elevated level of serum liver enzymes, erythrocyte sedimentation, raised bilirubin and hypoproteinemia. The interval between start of dapsone therapy and appearance of symptoms varied from 2–7 weeks. But one case the clinical manifestation occur 8 months after consumption of Multi drug treatment(MDT). All cases were treated with corticosteroids after withdrawing dapsone, MDT was continued by replacing dapsone with minocyclin.

Conclusion: There are significant clinical improvement in all cases, such as complete resolution of skin lesions and other symptoms.

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Drug hypersensitivity in a patient diagnosed with Lemierre's Syndrome

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Background: Lemierre's syndrome is an acute oropharyngeal infection with suppuration of the lateral pharyngeal space, bacteremia and septic thrombophlebitis of the internal jugular vein, causing metastatic infection. It is a rare disease with an estimated incidence of 1 case per million.
Methods: We report a patient diagnosed with Lemierre's syndrome who presented with a rash after 2 weeks of piperacillin-tazobactam, clindamycin and phenytoin.
Results: This 27 year old male with no significant past medical illness presented with a one week history of a dento-alveolar abscess of the right molar, fever, trismus, dysphagia with erythema and swelling of the neck. Work up showed a retropharyngeal extension of the dento-alveolar abscess, pneumonia, pneumomediastinum and necrotizing mediastinitis. HIV enzyme immunoassay was negative. The patient underwent cervical mediastinotomy with subxiphoid drainage and was given piperacillin-tazobactam 4.5g IV q8 and clindamycin 300mg IV q6. During the course of his hospital stay, the patient went into arrest, had post-gliottic seizures and was started on phenytoin 100mg cap q8.

On the 2nd week of piperacillin-tazobactam and clindamycin and on the 8th day of phenytoin, the patient presented with erythematous, pruritic maculopapular rashes initially on the upper extremities later on becoming generalized. Antibiotics were shifted to meropenem and phenytoin was shifted to levetiracetam. The patient received diphenhydramine, ranitidine and prednisone, which resolved the rashes. He also completed 6 weeks of meropenem and was discharged improved.
Conclusion: This is the first reported case of drug hypersensitivity in a patient diagnosed with Lemierre's syndrome. The importance of discontinuing the offending drugs and shifting to structurally unrelated drugs is emphasized.

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Hydrochlothiazide-induced lichenoid eruption: Apropos of 2 cases

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Background: The aetiology of lichen planus is unknown. Stress, autoimmunity and infection have been suggested as possible causes. Many drugs can provoke eruptions that clinically and histologically are similar to lichen. We describe in two patients a probable association between treatment with irbesartan-hydrochlorothiazide and lichenoid eruption.
Case Reports:
Case 1: An 80-year-old woman presented to the dermatology clinic with a 4-month history of acute onset of violaceus papules which started on her arms and spread to her trunk, back and legs. She had been taking amiodarone for auricular fibrillation for 15 months and irbesartan-hydrochlorothiazide for hypertension for 8 months. On physical exam, the patient had accurate violaceus and scaly plaques on the trunk, back and limbs. There were no ocular, buccal or genital lesions and she had no systemic symptoms. Blood count and biochemical profile were normal. Skin biopsy of involved skin showed typical features of lichen planus. Treatment with desonide and cetirizine provided no relief. Irbesrtan-hydrochlothiazide was substituted by irbesartan and the skin eruption and prutitis had resolved within two weeks.
Case 2: A 56-year-old man presented with acute onset of a pruritic rash which stated on his trunk and spread to his whole body. His past medical history included hypertension and Sharp syndrome for which he was taking irbesartan-hydrochlorothiazide and methotrexate. Methotrexate and irbesartan-hydrochlorothiazide were started approximately 2 years and 8 months before skin eruption respectively. On physical exam, the patient had accurate violaceus and scaly plaques on the trunke, back and limbs. There were bilateral shallow erosions of the buccal mucosa surrounded by a network of lacy white striae. Blood count and biochemical profile were normal. Skin biopsy of involved skin showed typical features of lichen planus. Treatment with desonide and cetirizine provided no relief. Irbesrtan-hydrochlothiazide was substituted by irbesartan and the skin eruption and prutitis had resolved within two weeks.
Conclusion: These observations illustrate a rare association between lichen and hydrochlorothiazide (On the basis of the Naranjo algorithm, it is probable that the systemic reaction was due to hydrochlorthiazide therapy). Clinicians should be aware of this potential adverse effect and recommend with-holding the drug intake when temporal relation is evocative.

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Prevalence of allergic contact sensitization among hostel students In Ajman University of United Arab Emirates (UAE)

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Background: It is estimated that a round 20-30% of people all over the world suffer from allergy. Contact with certain materials (allergens) is supposed to cause allergic skin sensitization culminating in development of contact dermatitis in some individuals. Some people are unaware of their sensitization as it shows no or mild symptoms. The prevalence of specific allergens varies by geographic area, population, age and sex.
Objective: The present study aimed at determining the relative frequency of positive reactions to different sensitizers among hostel female students in Ajman University-UAE.
Materials and Methods: Sampling approach was simple and descriptive. 46 female hostel students aged between 19–26 years were tested using the Universal standard patch test series (TRUE test) containing 23 common allergen sensitizers and a negative control. Cross sectional questionnaire was distributed for the same students and those with chronic diseases or pregnancy were excluded.
Results: The results were analyzed by SPSS. In contrast to previous European reports which suggest that nickel sensitization is the most common among females, this study shows that thimerosal is the common sensitizer among female hostel university students (32.6%), followed by nickel 15.2% and colophony 8.69%.
Conclusion: Thimerosal is present in contact lens solutions and cosmetics and most positive students who were unaware of their sensitization have used contact lenses for cosmetic purposes. This study should raise the alarm in using inert materials in contact lenses industry or at least warn females from the consequence of sensitization.

FOOD ALLERGY

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Immunologic comparisons between sesame and perilla seeds

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Background: Sesame and perilla have been a Korean traditional crop and mainly consumed as seasonings or seed oil. However, sesame food allergy is an increasingly common problem in many countries. This study aimed at comparing protein pattern of sesame with that of perilla and analyzing the cross reactivity between sesame and perilla.
Methods: Seeds were placed in 1:1 ethyl ether and extracted with slow stirring at 4°C for 2days in PBS (pH 7.4). To analyze protein patterns and specific IgE to sesame or perilla, crude extracts were separated by SDS-PAGE and then performed with Western blotting using sera diagnosed as allergic to sesame or perilla. Sesame extract with various concentrations is pre-incubated with

pooled sera before inhibition test, and then those sera were added to the ELISA plate pre-coated with perilla extract.
Results: 1. Both sesame and perilla had the higher protein concentration and contained protein bands of 6 – 82.2 kDa. However, after cooked, the proteins with high molecular weight disappeared and only 6 – 14 kDa protein bands existed weakly. 2. The IgE binding bands were also detected at 6 – 14 kDa in sesame and perilla by Western blot analysis. 3. In inhibition test, sesame showed low inhibition rate (<45%) against perilla by inhibition ELISA test.
Conclusion: Sesame and perilla showed different protein pattern and low cross reactivity each other. It suggests that perilla can be the replacement of sesame in consumption. However, because they have the possibility of food allergy, the further study about their major allergens is needed.

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Biochemical and allergenic properties of natural and genetically modified soybeans

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Soybeans are an important source of protein in human's dietary intake today. Some soybean proteins may be allergens and cause allergic reactions. In this connection, the aim of the present study was to investigate the biochemical and allergenic properties of extracts from natural and genetically modified (GM) soybeans.

The protein extracts were obtained from whole natural and GM soybeans, their capsules and cores and analyzed in SDS-PAGE, western blotting and ELISA. The SDS-PAGE analysis has revealed similar results of protein composition of allergenic extracts of core and whole natural and GM soybeans. The main protein band of whole soybeans and soybean core have low a molecular weight (m.w.) from 8,5 kDa to 20 kDa. The protein band with m.w. of 20 kDa is largest and conform a protein group of Kunitz trypsin inhibitor (Hor.v1, CMb, BDR). The protein fraction of capsule of soybeans with m.w. 14 kDa conforming soybean profilin (Gly m 3) predominate natural soy, whereas that with m.w. 34 kDa conforming vacuolar protein (Gly m Bd 30 k) predominate GM soy. We investigated 220 sera of patient's blood different age with atopic dermatitis and food allergy using ELISA. A frequency of revealed IgE-Abs to soy was increasing from 20 % in infant patients to 57,8% in adolescent patients. The increased levels of IgE-and IgG-Abs to capsule of natural soybeans were determined most often. We revealed IgG-Abs to wide protein spectrum of whole natural and GM soybeans using western blotting, where the largest protein fraction had m.w. of 30 kDa. The soybean capsule is known to contain proteins, which are homologous to the major allergens of birch pollen (Bet v1); with this in view high frequency of revealed IgE-Abs to the capsule of natural soybeans is fully explained in the context of cross-reactive antigens. While analyzing the blood serum of the patients with hypersensitivity to soy we revealed increased levels of IgE-Abs to birch pollen (81 % of cases) with high level of IgE-Abs found in 25% of patients. Therefore it was revealed that extracts of natural and GM soybeans contain proteins with identified allergic properties, which may cause the development of allergic diseases related to IgE-Abs and soy enteropathy related to IgG-Abs. Moreover, due to a pronounced homology with allergens of birch pollen, foodstuff containing soybeans may strengthen sensitization and intensify allergic reactions in Pollinosis patients.

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Different proteomic approaches to the identification of wheat flour allergens

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Background: Food antigens penetrating into the gastrointestinal tract do not generally cause clinical symptoms, because most individuals acquire tolerance. However, some food components are recognized as the cause of adverse reactions in susceptible individuals. Wheat belongs among six major foods, which are responsible for approximately 90% of food allergic reactions in children. Although cereals are essential in the daily nutrition, only a few proteins have been identified as cereal allergens on the molecular level.
Methods: To characterize wheat flour allergens, we prepared salt extract from seven of the most frequently used wheat cultivars and compared them by SDS PAGE. Proteins from the selected cultivar Sulamit were separated by 2-dimensional gel electrophoresis (2-DE), analyzed by immunoblotting and IgE-reactive proteins were identified by mass spectrometry (MALDI-TOF, QTOF and LCQDECA nLC-MS/MS ion trap) and tested for the ability to activate patient's basophils (expression of CD63 and CD203c markers was measured by flow cytometry). We developed ELISA to quantify patient's IgE Abs reactive with defined allergens. Wheat extract was also treated with pepsin.
Results: Patient sera can be divided into three groups: IgE Abs of the first one recognizes wheat components <30kDa, the second >30kDa and the third one reacts with components ranging from 6-170kDa. The pattern of wheat components recognized by IgE Abs was significantly influenced by conditions of protein separation and treatment with digestive enzymes. Using SDS PAGE, 2-DE, immunobloting and mass spectrometry we identified 19 potential wheat allergens. Employing newly developed ELISA, levels of IgE Abs against proteins from the Sulamit extract and one of the dominant wheat IgE binding component, α-amylase inhibitors type 1 and 3, were quantified and shown to be significantly elevated in sera of allergic patients compared to those of healthy controls. Furthermore, the salt extract from the cultivar Sulamit as well as α-amylase inhibitors were shown to by able induce significant activation of basophils of allergic patients.
Conclusion: This study characterized the spectrum of wheat components recognized by IgE Abs of allergic patients, analyzed the impact of proteins separation and digestion on immunogenicity of wheat allergens, compared the sensitivity and specificity of newly developed specific IgE ELISA assay and in vitro basophils activation test using identified allergens.

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Heat-induced denaturation, aggregation and changes of immunoreactivity of milk proteins

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Background: Milk for liquid consumption or for processing requires an initial heat treatment to control the growth of microorganisms. However, heat treatment of milk causes the heat denaturable whey proteins to aggregate with κ-casein (κ-CN) via thiol-disulfide bond interchange reactions and other changes. Influence of the soluble macroaggregates presence and other heat-induced changes on immunogenicity of the commercial milks is still a controversial issue. The aim of this study was to contribute to increase knowledge in this area.
Methods: The kinetics of heat-induced precipitation of the main whey proteins β-lactoglobulin (β-LG) and α-lactalbumin (α-LA) were examined in water solutions of individual proteins, in fresh milk, whey, and model mixtures of the both whey proteins with κ-CN at 95°C during 30 min.

At the same time, the kinetics of formation of heat-induced aggregates of the main whey proteins with κ-CN was investigated in fresh milk, whey, and the model mixtures using GP-HPLC method.

Dot blot assays using sera of 19 allergic patiens containing specific IgE antibodies against milk proteins were employed to assess the immunochemical reactivity of the milk proteins containing samples before or after heat treatment. Immunoreactivities of 6 commercial milk samples (pasteurized, UHT and fermented) with different content of the soluble macroaggregates were also assessed by commercial ELISA tests with monoclonal antibodies against individual whey proteins and using the sera of allergic patiens.

Results: The decrease of the main whey proteins β-LG and α-LA was significantly higher in milk or whey than in solutions of the individual proteins. The experiments with the model protein mixtures showed that presence κ-CN is necessary to form the aggregation complexes of the whey proteins at 95°C. The kinetics of formation of the heat-induced aggregates in milk and model protein mixtures was complex with two different maxima during 30 min time interval.

Conclusion: Surprisingly, we have not observed significant changes of the immunochemical reactivities of the milk proteins samples after the heat-induced aggregation under the conditions of the dot blot assays. However, the experiments will be continued to confirm the results.

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Characterization of the putative allergens in nine Chinese adults with food allergy to limpet

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Background: Limpet, a gastropod mollusc, is the second most common cause of IgE-mediated food allergy among adults attending our Clinical Immunology/Allergy clinic. The aim of this study is to characterize the putative allergen(s) and ascertain possible cross-reactivity with common inhalant and food allergens in our patients with limpet allergy.

Methods: Consecutive adults presenting from 1 July 1994 to 31 April 2002 with immediate hypersensitivity following ingestion of limpet were studied. Sera stored at −70°C were assayed for specific IgE to common inhalant and food allergens. IgE western blotting and tandem mass spectrometric analysis were performed to identify the allergens. ELISA inhibition assays were carried out to compare the allergenicity of limpets with other gastropods, as well as to recombinant tropomyosin and arginine kinase allergens from dust mites and cockroaches.

Results: There were 9 patients (6 males) and all were ethnically Chinese. Their mean age was 38 ± 17 (19–60) years. Eight patients had previously ingested limpet, with allergic reactions occurring during a median of 2 occasions. Six patients had history of asthma, allergic rhinitis and/or atopic eczema. Seven patients (2 refused skin testing) tested positive on prick-prick test to canned limpet, 3 were also positive to abalone, and 2 to pacific clam. On invitro specific IgE assay, 77.8% were positive to shellfish mix, 77.8% to *Dermatophagoides pteronyssinus*, 55.6% to *Dermatophagoides farinae*, 44.4% to *Blomia tropicalis*, and 33.3% to cockroach mix. Western blot and protein mass spectrometric analysis showed that the putative limpet allergens were a 37kD tropomyosin and a 40kD arginine kinase. ELISA inhibition studies using recombinant dust mite and cockroach tropomyosin as well as arginine kinase showed moderate cross reactivity between these allergens and the allergens from limpet. There was no significant inhibition of binding by abalone and clam.

Conclusion: The putative limpet allergens in our patients are a 37kD tropomyosin and a 40kD arginine kinase, both of which are major allergens in other gastropods (abalone and pacific clam), crustaceans, American cockroach and house dust mite. Prior sensitisation to house dust mite and cockroach in our patients could have played a role in limpet food allergy. Surprisingly there was limited cross-reactivity with abalone and pacific clams, two closely related gastropod molluscs.

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Cross reactivity among soy, penaut and lupine

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Background: Soy protein is a common dietary protein, the intake is about 1–2 g day in Europe among adults according a multicentre study. Soy based infant formulae are often used in infancy and early childhood in cow's milk allergy and in healthy children instead of cows milk, too.

The prevalence of allergic reactions (gastrointestinal, skin, and pulmonary) elicited by soy protein is around 6% in atopic children. Severe or fatal reactions seem to be rare, but the rate higher in children with peanut allergy. Lupine is a newly used legume in the human diet, which theoretically can give also cross reaction with soy and peanut. Aim of the study was to determine the cross reaction among soy protein, peanut and lupine.

Methods: Immunoblot technique with SDS-PAGE protein separation and ELISA method. Polyclonal antibody and IgE soy positive human sera were used to detect reactions.

Results: Immunological cross reactivity was demonstrated between the major allergens of studied legume proteins. The beta-conglycinin and their oligomeric subunits in soybean have shown cross reactivity with the Ara h1 in peanut and the 7S vicillin allergens in lupine. Furthermore it was observed an immunological cross reaction among the soybean glycinin allergens and the Ara h3 in the peanut and the vicillin 11S allergens in lupine, while the soybean profilin cross reacted with Ara h5 peanut allergen. The 2S allergens of prolamin superfamily in soybean as trypsin inhibitors and lectins shown cross reactive epitopes with Ara h2, Ara h6 and Ara h7 allergens in peanut. Soybean, peanut and lupine protein extracts did not differ in their IgE reactivity with soy positive human sera.

Conclusion: Cross reactivity among the major allergens of soy, peanut and lupine was proved with human soy positive sera in vitro. Results draw attention to the possibility of anaphylaxis as a result of allergic cross reaction in patients with peanut and soy allergy. On this basis, it has to emphasise the need of precise labelling for food components to avoid the anaphylaxis.

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Assessment of IgE reactivity and allergenic activity of cow's milk allergens by microarray and basophil degranulation experiments

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Background: Milk allergy is the most common IgE-mediated food allergy in early infancy. Cow's milk contains about 25 proteins of which eight have been identified as allergens but there are varying results regarding the frequency of IgE recognition and the allergenic activity of the individual allergens. We developed a microarray for milk proteins which allows to determine indiviual patients' IgE reactivity profiles as well as the biological activity of the disease-causing allergens using small amounts of patients' sera.

Methods: We dotted different milk components including whole milk extracts, milk fractions as well as purified and recombinant milk allergens, and recombinant and synthetic fragments of milk allergens onto microarray and determined the IgE reactivity profiles of 78 milk allergic patients. In parallel, allergenic activity of the milk components was determined using rat basophil leukaemia (RBL) cells transfected with human Fcå receptor that had been loaded with the very same sera.

Results: We found that 84.6% of milk allergic patients showed IgE reactivity to whole cow's milk extract. A high number of patients had IgE antibodies to sheep's and goat's milk, 82.1 % and 67.9%, respectively. But only 30.8% of the tested patients showed IgE reactivity to mare's milk. The most frequently recognized cow's milk allergens in our study were alpha-lactalbumin (62.8%), alpha-casein (48.7%), beta-lactoglobulin variant A and B (48.7 and 50.0%, respectively), and beta-casein (43.6%). Lactoferrin and BSA seemed to be of lesser importance since recognition frequencies were below 5 %. The most potent cell activating component in the humanized RBL assay was cow's milk which induced a relevant degranulation in 47.4% of the patients followed by goat's and sheep's milk (35.9 and 28.2%, respectively). Among the single components the caseins showed the highest biological activity (25 to 34.6% of the patients' sera).

Conclusion: Using microarray technology in combination with data from basophil release assays it is possible to test simultaneously the frequency of IgE reactivity and the allergenic activity of a multitude of allergens using just a few drops of patient's serum. This test system will be important for diagnosis and therapy of cow's milk allergy.

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Epitope mapping on gliadins for patient sera reacting to modified gluten

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Background: Wheat gluten can be modified by enzymatic or chemical treatment (wheat isolate) in order to improve its functional properties, increase solubility and thus broaden its application in food industry. However, these modifications were shown to induce IgE mediated allergies in individuals usually tolerant to wheat. Leduc et al. (2003) published the first case of anaphylaxis after ingestion of a reconstituted meat containing a wheat isolate. Laurière et al. (2006) also showed that hydrolysed wheat proteins can induce severe allergic reactions by contact or ingestion. The objective of our work was to identify epitopes detected by IgE from patients with reactions to chemically deamidated gluten (product in which some glutamine residues were modified into glutamic acid).

Methods: Sera from twelve patients with anaphylaxis, urticaria or WDEIA containing IgE antibodies to laboratory or industrial deamidated gluten in ELISA or RAST were analysed by pepscan with overlapping synthetic peptides. IgE-binding epitopes were searched among entire sequences of ã and w2-gliadins with identification of critical animo acid and analysis of effect of substitutions of glutamine into glutamic acid.

Results: Eight sera reacted with repetitive peptides of ã or w2-gliadin containing a common octapeptide rich in proline and glutamine. Five critical positions for IgE binding have been identified on this epitope. Substitution of one of its glutamine into glutamic acid induced its recognition by two additional sera and simultaneous substitution of two glutamine into glutamic acid increased the intensity of responses obtained for the ten positive sera.

Conclusion: Patients with reactions to deamidated gluten possessed IgE antibodies directed to linear sequences of the repetitive domains of ã and w2-gliadins. IgE antibodies from most of the patient sera analysed in this study bound to an immunodominant epitope of 8 residues. Deamidation of two glutamines of this epitope increased significantly its recognition by patient IgE. This epitope is similar to a B epitope evidenced for celiac patients. These results show that some industrial treatments may increase the risk of allergy development.

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Omega-5-gliadin allergen specific IgE antibodies are clinically useful in the diagnosis of food allergy

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Background: The relationship between food-specific IgE titers and the outcome of food challenges is well established for egg and milk through threshold values and probability curves. For wheat or components of wheat no such relationships have been established.

Objective: To evaluate the clinical use of wheat and the wheat component omega-5-gliadin specific IgE titres in relation to the outcome of food challenge.

Methods: Two hundred and forty nine children with suspected wheat intolerance, from 3 clinics in Japan, were included in the study. Age ranged from 6 months to 15.4 years (mean=3.8 years, median=3.0 years). The diagnosis of food allergy was judged through confirming symptom in food challenge tests. In relation to their visit for food challenge, allergen specific IgE antibody levels for wheat and omega-5-gliadin were measured (ImmunoCAP™, Phadia AB, Sweden).

Results: One hundred and eighty five children were challenged. The most frequent symptom for a failed challenge was related to the skin, then cough and lower respiratory systems, 48%, 24% and 13%. Three children had an anaphylactic reaction. Fifty four percent of the children had one symptom, 32% showed 2 symptoms, 13% showed 3 symptoms and 1 child had 4 symptoms. The specific IgE titres for both wheat and omega-5-gliadin were higher for the children who failed the food challenge, Table 1.

In a simple logistic model both wheat and omega-5-gliadin showed a significant relationship between the probability of challenge reaction to the food and the specific IgE antibody concentrations, wheat, OR 1.41, 95%CI:1.17–1.40 and omega-5-gliadin OR 1.97, 95%CI:1.51–2.55. In a multivariate model using both wheat and omega-5-gliadian, omega-5-gliadin showed the strongest association with a failed challenge provocation, OR= 1.95 95%CI:1.41–2.67.

Conclusion: Our results show that omega-5-gliadin specific IgE antibody titres are related to the probability for a failed oral food challenge. Omega-5-gliadin IgE in combination with wheat IgE antibodies are a good predictor of wheat challenge outcome.

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Oral allergy syndrome due to plant-derived foods: a clinical review of 63 patients over a period of 6 years

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Background: $B!'(BThe clinical features of many patients with oral allergy syndrome (OAS) due to plant-derived foods have rarely been reported in Asian countries.

Objective: We aimed to determine the causative foods of OAS due to plant-derived foods based on clinical features and skin prick tests (SPTs).

	Wheat challenge succeeded	Omega-5 challenge succeeded	Wheat challenge failed	Omega5 challenge failed
Mean	18.72	0.75	33.39	5.33
Median	8.00	<0.35	20.85	1.05
No.	75	75	110	110

Furthermore, we aimed to elucidate the association between causative foods and sensitized pollens in patients with OAS due to plant-derived foods.
Methods: SPTs and specific IgE measurements (CAP-FEIA: CAP) were performed in relation to foods and pollens in 118 patients with positive histories of OAS due to plant-derived foods. Patients with positive histories and with positive skin test responses were identified as having type I allergy to the causative foods.
Results: The mean age of 63 patients with positive histories and positive skin test responses was 29.2 years (range, 2-61years), and there were twice as many females as male. The most frequent causative foods were found to be apple, peach, kiwi, and melon in 13, 12, 12, and 11 patients, respectively. CAP frequency was shown to be similar to that of SPT regarding apple, whereas it was less than that of SPT regarding melon, peach, and kiwi. A significant correlation between the frequencies of SPT and CAP was found regarding apple ($R=0.39$, $p<0.05$) but not peach, kiwi, and melon. Forty-one of 63 patients with OAS (66.1%) had pollinosis and/or allergic rhinitis. In patients with OAS due to apple, the positive ratio of CAP response against alder pollen was higher than that in patients with OAS due to melon. In patients with OAS due to melon, the positive ratio of CAP responses against ragweed pollen, grass pollen, and mugwort pollen was higher than that in patients with OAS due to apple.
Conclusion: In this study, positive ratios of SPT and CAP tended to differ according to the causative food, showing a smaller potential for reaction than might be suggested by patient history. Therefore, for the time being it would be more accurate to use a skin test for the diagnosis of OAS due to plant-derived foods.

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Pollen-Food Syndrome (PFS); characteristics, foods involved and predictive factors in a UK cohort

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PFS is a type of Oral Allergy Syndrome (OAS), a common cause of food allergy in adults. To date, there have been no published studies on the prevalence and characteristics of PFS in a UK cohort. This study, designed to test a diagnostic questionnaire, also provided information on the foods involved and and allergy history associated with PFS.

Subjects with reported springtime hay fever completed a PFS diagnostic questionnaire (PFSDQ) and were allocated to either group 1 (has PFS), group 2 (no PFS, ?food allergy) or group 3 (no PFS or food allergy). They then completed a questionnaire (PFSPQ) on their rhinitis, concomitant asthma, medication, symptom severity, reactions to foods, and severity of food symptoms. Diagnosis of PFS was made using clinical history, prick by prick tests (PPT) to fresh foods and oral food challenge.

119 subjects completed the study (mean age 36 years, 85 female); 88 subjects (Group 1 – 58, Group 2 – 30) reported reactions to 73 different foods. A final diagnosis of PFS was made in 60 of the 199 subjects (50%). There were no differences in age or gender between Groups 1–3 or for final diagnosis. Most variables in the PFSPQ did not differ between Groups 1–3 or for final diagnosis. These included family history of atopy, childhood allergy, age of onset of rhinitis, symptom severity scores for rhinitis, the presence or absence of asthma, number of years with a food allergy or onset of food symptoms before, after or at the same time as the onset of their hay fever. Subjects most commonly reported reactions to apple (39 subjects), hazelnuts (22), almonds (21), milk (18), strawberry (16), walnuts (16), peach (14), cherry (14), Kiwi (14) and wheat (13). Subjects in Group 1 reported symptoms to significantly more foods than subjects in Group 2 ($p<0.02$). Apple, hazelnut, peach, almond, kiwi and cherry were the commonest foods to have reported symptoms confirmed by positive PPT. Milk and wheat had the lowest concordance with PPT; only 21% of reported reactions to milk were confirmed and 0% of reactions to wheat.

These data show that 50 % of a cohort of UK subjects with reported springtime hay fever had PFS. The commonest foods to cause reactions were similar to other published studies, although kiwi appeared to be a more significant precipitant of PFS symptoms than previously reported. In this study, age, gender, allergy history, asthma and time of onset of food symptoms were not associated with a diagnosis of PFS.

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Usefulness of skin prick test using bifurcated needle for the diagnosis of food allergy among infantile atopic dermatitis

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Objective: We investigated the usefulness of skin prick test (SPT) for the diagnosis of egg white (EW) CAPRAST-negative egg allergy infants and followed up the EW-CAPRAST in this study.
Subjects and Methods: Data of negative SPT using Bifurcated needle (BF) were analyzed from the data of 202 infantile atopic dermatitis patients, who had received SPT from January in 2001 to April in 2005. From the analysis of SPT results, the average and standard deviation of negative value were obtained to set the range of positive SPT result. Among 202 cases, 89 suspected-egg allergy infants with negative IgE CAPRAST against EW at the time of first visit were recruited to this study to examine the usefulness of SPT. Positive conversion of EW-CAPRAST was checked in 78 cases (65: egg allergy+, 13: egg allergy-) who had been followed up in our outpatient clinic.
Results: Range of negative SPT control value (mean+ 2SD) using BF among infants could be set as less than 2 mm for wheal and 5 mm for erythema. Among 89 suspected-egg allergy infants with negative EW-CAPRAST, 72 infants (80.9%) were diagnosed as egg allergy by the combination of elimination and provocation test, interestingly 39 infants (54.2%) showed positive SPT results in 72 egg allergies. In the follow up study of 78 negative EW-CAPRAST cases, 47 EW-CAPRAST out of 65 egg-allergy cases turned positive later infantile period (mean EW-CAPRAST: 9.6+/−16.7 Ua/ml at 9.9+/−5.6 months old). EW-CAPRAST of 7 cases in 13 non-egg allergies also turned positive in the follow up, however EW-CAPRAST titer was relatively lower compared to that of egg allergies (1.1+/−1.5 Ua/ml at 13.3+/−2.6 months old).
Conclusion: We experienced fairly number of atopic infants with negative EW-CAPRAST at the first outpatient visit, who were later diagnosed as egg allergy. In about half of these cases, SPT was extremely useful for the diagnosis. In the follow up of egg-allergy infants, three quarter of EW-CAPRAST tuned positive around 10 months old. EW-CAPRAST of atopic infants without egg allergy also turned transiently and slightly positive. In the conclusions, SPT seemed to be more useful than EW-CAPRAST for the diagnosis of egg allergy in early infantile period, however provocation test may be required for the definitive diagnosis in suspected-egg allergy infants without any proof of egg-sensitization.

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Optical near-field immunochip biosensor for rapid allergen detection and clinical diagnosis

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Background: An optical immunochip biosensor has been developed as a rapid method for allergen detection and in vitro immunosensing. The optical near-field phenomenon underlying the setup design of the sensor is called resonance-enhanced absorption (REA) utilizing gold nanoparticles as signal transducers in a sensitive interferometry-setup. Combined with visual readout that eliminates complex detection instrumentation, this assay system excellently meets the requirements of a rapid method for allergen detection and provides opportunities for the development of a miniaturised multi-array-sensor with improved sensitivity for allergy testing in clinical diagnosis.
Methods: REA is observed when light-absorbing metal clusters are positioned at a nanodistance from a highly reflective mirror and illuminated with white light of the visible and near-infrared spectrum from the particle side. As a direct detection method based on distance-dependent light interference between the mirror and the resonantly excited metal clusters, a REA based capture assay has the power to link affinity binding with three-dimensional structural information.
Results and Discussion: The immunochip biosensor showed concentration-dependent colour development by capturing biomolecule-functionalised gold nanoparticles on allergen or immunoglobulin coated chips. Semi-quantitative immunochemical responses are directly visible to the 'naked' eye of the analyst with sensitivity comparable to common immunoassay procedures in microtitration plate format (ELISA). Unspecific background signals were minimized to the absolute target specificity of the antibodies and antisera used in the bioassay. We have applied this method to the detection of the egg white allergens ovalbumin and ovomucoid in complex food samples. To achieve improved interpretation and cross-validation relating to the immunochemical characterization of allergenic substances of processed food at the protein and peptide level, complementary investigation (in terms of applicability, reproducibility, and reliability of results) was carried out with alternative immunological methods (performed in ELISA and immunoblot format). In conclusion, the REA-based imunochip sensor is easy to fabricate, is reproducible in its performance, has minimal technical requirements, and will enable high-throughput screening of affinity binding interactions in technological and medical application for determining allergenicity.

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AllergenOnline, a peer-reviewed protein sequence database for assessing the potential allergenicity of genetically modified organisms and novel food proteins

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Background: New genetically modified (GM) organisms and novel processed food proteins are being introduced into commerce. Countries require safety assessments include a bioinformatics (sequence) comparison of the new protein to determine whether the new protein is a known allergen or is so similar to an allergen that currently allergic consumers would be at risk of reactions. If a match is found, the product would have to be tested using sera from appropriately allergic subjects. The quality of the database is a primary determinant of the search efficacy. A peer review process was used to update AllergenOnline and tools for an effective bioinformatics search are provided, free to the public.
Methods: A list of 1758 "allergens" was compiled from NCBI using keyword searches, plus entries in the Allergen Nomenclature list of the International Union of Immunological Societies (IUIS) and AllergenOnline version 6. Sequences were grouped based on taxonomy and sequence identities. Peer reviewed articles were identified for each group. Criteria were developed to evaluate the evidence based on in vivo and in vitro tests using subjects with clinically defined, relevant allergic disease. Published data for each group was compiled and evaluated by primary and secondary reviewers, then the panel to remove groups with insufficient evidence of allergenicity (no IgE binding or in vivo reactivity, with subjects having at least minimal descriptions of allergies to the source of the protein). Free full-length and sliding 80 amino acid alignment tools are provided to allow public searches of the database. Key examples from the literature are used to illustrate the appropriate use of the database.
Results: AllergenOnline version 7 contains 1251 sequences with published data demonstrating at least evidence of specific IgE binding using sera from subjects having some allergic symptoms when exposed to the source of the protein. Many isoforms are listed for some proteins. Others are represented by a single sequence. Bioinformatic comparisons with transgenic proteins from approved products and the literature were evaluated. No approved product exceeded Codex Guideline criteria, but some from the literature (presumably not in commerce) did, indicating a likely need for serum IgE testing.
Conclusion: The curated allergen database will markedly improve the reliability of sequence matches for assessing the potential allergenicity of these new foods.

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The EuroPrevall allergen library: a collection of high quality purified natural and recombinant allergens for in vitro diagnosis

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Background: The quality of in vitro IgE tests used for food allergy diagnosis mainly depends on the analytes used. Usually total extracts of the suspected foods are used. However, in the recent past a rising number of studies about component resolved diagnosis have attracted rising interest. In many cases the use of purified single allergens either of natural origin or applied as a recombinant product has shown a superior performance when compared to total extracts. Within the EC funded IP Europrevall a library of 31 highly purified allergens from 10 foods listed in the new Labelling directive of the European Commission was established for developing novel diagnostics.
Methods: Highly purified allergens either natural or recombinant from 10 foods (apple, peach, hazelnut, peanut, celery, cow's milk, goat's milk, hen's

egg, fish and shrimp) were characterized regarding their structural integrity, biological and allergenic activity. Protein mass was verified by mass spectrometry and sequence integrity by MALDI-TOF and N-terminal sequencing. Presence of isoforms in natural allergen preparations were identified by 2D Gel electrophoresis and secondary structure was evaluated by NMR maps of low molecular weight proteins. Allergenic activity was characterized using a panel of sera from food allergic patients. Results: Purification protocols for natural and recombinant allergens were established and quality assessment of the purified batches performed. Depending on either natural or recombinant origin different quality criteria were defined and met for inclusion into the allergen library. Structural integrity of the purified proteins was verified and IgE binding activity was assessed.

Conclusion: This panel of well characterized allergens will be used to prove the concept of component resolved diagnosis and presents the necessary tools for setting up novel diagnostics. Using a significant number of allergic patients' sera across Europe it will contribute to identify relevant sensitization patterns and to improve dietary recommendations for the food allergic patient in the future.

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Dominant epitopes and allergic cross-reactivity of the thaumatin-like protein act d 2

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Background: Some important food allergens have been identified as pathogenesis-related proteins (PRs). Thaumatin-like proteins (TLPs) belong to the family 5 of PRs. Several structures of plant TLPs have been solved in our group, including the TLPs from sweet cherry (Pru av 2), kiwifruit (Act d 2) and apple (Mal d 2). Comparing different structures belonging to the same protein family can lead to the identification of conformational epitopes.

Methods: ELISA inhibition analysis using the sera of kiwi allergic patients was used for determination of immunological cross-reactivity between Act d 2 and other TLPs (thaumatin, zeamatin, Pru av 2 and Mal d 2). In order to identify putative cross-reactive epitopes virtual epitope mapping was applied. The similarities and differences in surface topology were compared.

Results: ELISA inhibition assays for Act d 2 showed a high cross-reactivity with zeamatin, medium cross-reactivity with Pru av 2 and Mal d 2, and low cross-reactivity with thaumatin. On the other hand, the sequence comparison between Act d 2 and other TLPs yielded a different order of similarity: zeamatin> thaumatin>Pru av 2> Mal d 2. Combining the immunological data and virtuale epitope mapping three putative epitope regions could be determined on the surface of Act d 2.

Conclusion: High sequence homology/identity between two allergens is not necessarily the indicator for high cross-reactivity. We used the comparison of surface features of cross-reactive and non cross-reactive allergens to derive conformational epitope regions.

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Bet v 1 homologous proteins in kiwi fruit- relevant allergens?

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Background: In Central and Northern Europe birch pollen related food allergy is mainly based upon cross reactive IgE to Bet v 1 and homologues present in various plant derived foods. So far, actinidin, kiwellin and the thaumatin-like protein have been identified from kiwi as relevant allergens. However, till now nothing is known about Bet v 1 homologous proteins present in kiwi. Preliminary efforts to purify natural Bet v1 homologues in sufficient quantities from kiwi failed so far.

Methods: Two individual sequences (from green and gold kiwi fruit) homologous to Bet v 1 were identified from an EST cDNA library, subcloned into expression systems pETBlue-2 and pMW 175, respectively and expressed in *E. coli*. The resulting recombinant proteins were purified and characterized according to established methods. IgE binding activity of the purified recombinant Bet v 1 homologous proteins from kiwi fruit was tested by IgE-ELISA, IgE immunoblots, and inhibition assays.

Results: Alignment of recombinant kiwi Bet v 1 homologues and recombinant Bet v 1a revealed an amino acid sequence identity of 50%. Well characterized purified recombinant Bet v 1 homologues from green *(Actinidia deliciosa)* and gold kiwi fruit *(Actinidia chinensis)* were recognized by birch pollen and kiwi sensitized patients in IgE immunoblots and ELISA experiments. The presence of a natural Bet v 1 homologous protein in green and gold kiwi fruit was confirmed by IgE immunoblot inhibition experiments using natural kiwi fruit extracts and immuno tissue prints. Cross-reactivity of the two recombinant proteins with varying intensities to natural Bet v 1 related protein from green kiwi and to rBet v 1a was demonstrated by ELISA and immunoblot inhibition assay.

Conclusion: This is the first evidence of cross reactive Bet v 1 homologous allergens present in gold and green kiwi fruits. The present data point out that Bet v 1 homologues are relevant allergens in kiwi, especially for patients sensitized to tree pollen and kiwi fruits and might have been neglected so far due to low abundancy in the conventional extracts used for diagnosis.

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The evolutionary biology of allergens of the cupin and the Bet v 1 superfamilies

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Background: The massive accumulation of sequence data of plant proteins in recent years has made the classification of allergens into protein families possible. By comparing sequences and structures, related proteins can be grouped together into families, and related families can be grouped together into superfamilies. This much broader picture now allows us to view allergenic proteins in their evolutionary context.

Methods: Representative protein sequences were compiled by manually searching the Pfam database of protein families. Multiple sequence alignments and generation of neighbor-joining trees was performed using ClustalX. Phylogenetic trees were drawn with TreeView.

Results: The cupin superfamily comprises numerous functionally highly diverse protein families from all groups of organisms including archaea. However, allergenicity within the cupins is confined to the vicilin and legumin plant seed storage proteins. The Bet v 1 superfamily contains the PR-10 family, the family of major latex proteins and ripening-related proteins, the norcoclaurine synthases, and the cytokinin-binding proteins. The PR-10 proteins from certain taxa such as the Fagales and Rosaceae are the only allergenic members of this superfamily. The divergence of the allergenic members of the Bet v 1 superfamily from the related major latex proteins, ripening related proteins and norcoclaurine synthases represents an ancient evolutionary event. Apparently the ancestors of modern day allergens were present long before any type of immune system had evolved.

Conclusion: This study of the distribution of allergenic and non-allergenic members of protein families which was funded by the Austrian Science Fund grant SFB F01802 will provide new insights into the evolution of allergenicity and the factors that make proteins allergenic.

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Optimizing allergen components in litchi and peach extracts for in vitro specific IgE assays

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Background: Allergy to fruits is often associated with pollinosis and is then often related to milder symptoms such as oral allergy syndrom (OAS). However, especially in warmer regions more severe symptoms are also common. Fruits are here more prevalent foods and are also often eaten in more sun-ripe forms. Allergy to fruits of the Rosaceae family is strongly associated with Bet v 1 in northern regions but in warmer regions allergic reactions to these fruits are more often related to lipid transfer proteins (LTPs) and sometimes also profilins. The aim with this study was to explore new extraction buffers and different parts of fruits trying to optimize extraction of labile Bet v1 analogues as well as more stable profilins, LTPs and other allergen components.

Methods: Peach was selected as a model for Rosaceae fruits where Bet v 1 homologues, profilin and LTPs are important allergen components. Litchi was selected as a fruit where the exposure to the population in Europe is increasing with changing feeding habits and where profilin is suggested to be an important allergen. Extracts were produced using different buffers and procedures and different parts of the fruits (pulp/peel) were used. Samples from patients from different regions with different symptoms (from mild to severe reactions) were analysed using ImmunoCAP coupled with improved extracts as well as single components. IgE antibodies to components were also assayed with a microarray using only 30 µl of sample.

Results: Optimal extraction performance was achieved by a cold extraction procedure containing polyvinylpyrrolidone. Both pulp and peel were needed for optimal performance. For selected samples, mainly reacting to Bet v1, increased results as measured with ImmunoCAP was seen in 76% (62/81) for Peach f95. In litchi profilin was the dominating allergen component in several patient samples both measured by ImmunoCAP and using the microarray assay. The results were confirmed by inhibition studies.

Conclusion: Improved extraction methods can enhance the content of both Bet v 1 homologues and profilin in fruit extracts used for in vitro assays for specific IgE. Microarrays are useful tools to analyse component profiles in samples using very small volumes of sample. Profilin (Lit c1) is an important allergen in litchi and may cause severe reactions. In peach LTP (Pru p3) and the Bet v 1 homologue Pru p1 were the more important components in the tested patient population.

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Characterization of saffron pollen allergens

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Grass pollen is one of the most important causes of IgE-mediated allergic reactions. Hypersensitivity to saffron pollen is an occupational allergy and is a serious problem especially among people dealing with saffron (farmers). Saffron is a native plant of Iran and this study was performed on saffron pollen in Khorasan province.

Saffron pollen allergenicity was proven using skin prick test. The aim of this study was to characterize IgE-binding components of saffron pollen.

At first saffron pollen was extracted with phosphate-buffered saline (PBS 125mM, pH 7.2–7.4) at 4°C for 5 hours with gentle shaking. After centrifugation and delipidation, the extract was concentrated by polyethyleneglycol (PEG20000) at 4°C. Concentrated saffron pollen extract was fractionated by SDS-PAGE and transferred to nitrocellulose paper by western blotting technique. After western blotting, using sensitive individual sera and anti-human IgE-HRP immunoblot analysis was performed. Finally one specific IgE-binding band was showed with molecular weight of 20-27 KD.

This study indicated that sensitive sera contain specific antibodies directed against allergens present in saffron pollen extract. Also this component can be bind specifically to IgG.

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The role of Interleukin 4 and 5 (IL4, 5) in development of chronic inflammations of stomach in patients with food allergy

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The chronic gastritis can cause protracted iatrogenic effect of certain medicaments, for example non-steroid anti-inflammatory drugs also. Still, to little is known about the extent and role of hypersensitivity in patients with atopic diathesis in the development of chronic gastritis. The aim of the study was to determine serum concentrations of IL-4, IL-5 in patients with chronic gastritis and food allergy, who had been infected with H. pylori. The study was conducted on patients with atopic diathesis, suffering from dyspepsia and abdominal pain, who were monitored in our Outpatient Clinic for Food Allergy. The study group consisted of 71 patients, including 42 females aged 16¡V54 years (mean age 35.5 years) and 29 males aged 18¡V60 years (mean age 36.2 years). The control group was formed of 40 patients with dyspepsia and abdominal pain related to chronic gastritis and confirmed in endoscopy and histopathological investigation. specimens. The patients underwent endoscopy The samples were stained with eosin and hematoxylin (H and E) and with modified Giemza stain. Selected serum cytokines: IL-4, IL-5, nwere determined with ENDOGEN kits(Cambridge). Serum levels of interleukin-4 in patients with food allergy ranged between 0.2 to 291 pg/ml, with mean value of 27.85¡Ó34.89 pg/ml. In the group of non-atopic patients with chronic gastritis, the range of determined values was 1.5 ¡V60.1 pg/ml ¡V mean concentration was 13.26¡Ó7.0 pg/ml. In control patients with functional dyspepsia, the concentration of IL-4 obtained ranged between 0 and 13.2 pg/ml ¡V mean IL-4 concentration was4.31¡Ó2.8 pg/ml. IL-4 concentrations in various groups are presented in table 1. The concentrations of interleukin-5 in patients with food allergy ranged between 0 and 111.3 pg/ml (mean value 7.43¡Ó3.20 pg/ml). In control group of non-atopic subjects with chronic gastritis, IL-5 levels were 0¡V20.6 pg/ml (mean level 4.03¡Ó1.60 pg/ml). The comparative analysis of IL-5 and IL5 levels between patients with food allergy and control group with chronic gastritis showed statistically significant differences ($p<0.05$). Conclusions: 1. Chronic exposure to the sensitising food in patients with food allergy results in the significant elevation of IL-4 and IL-5 levels. 2. Patients with food allergy and chronic gastritis, who had been infected with H. pylori manifest simultaneous increase in IL-4, IL-5, which suggests the participation of allergic factors in the pathogenesis of inflammation.

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Summary of blinded-food challenges against hen's egg and cow's milk allergies in the past 11 years

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Objective: The aim of this study was to analyze the results of 175 single-blinded food challenges (SBFC) against hen's egg(HE) and cow's milk(CM) allergy patients.

Methods: We analyzed the data of the patients who had received the SBFC in Sagamihara National Hospital from 1995 to 2005. The total numbers of

examination was as follows; HE: 92 challenges (male: 65, female: 27, mean age: 72.6 months), CM; 83 challenges (male: 62, female: 21, mean age: 71.0 months). SBFC was performed using dried powder (4g of whole egg powder or 6g of CM powder) with 120g strawberry puree as vehicle and the test was carried out along the AAAAI office procedure manual (JACI 82: 986, 1988).

Results: Most of the subjects received the SBFC prior to the entrance of primary school whether or not they could take school lunch. The positive rate of challenge test was as follows; HE: 70/92 (76%), CM: 37/83 (45%). The most common symptom induced by HE challenge was GI reaction (83%), followed by skin reaction (69%) and lower respiratory symptoms (17%). We further confirmed challenges with heated hen's whole egg against 62 subjects in 70 diagnosed raw egg allergy patients. Interestingly a discrepancy of the result between raw egg challenge and heated egg challenge was seen. Twenty nine percentage (18/62) of raw egg allergy patients could take heated whole egg. Egg white specific IgE level of patients who could not pass the challenge (18.0UA/ml) was higher than that of those who passed (8.0UA/ml) ($p<0.05$). For CM challenge, skin reactions were the most common (89%), followed by lower respiratory symptom (65%) and GI reaction (19%). There was no statistical difference of CM specific IgE level between failed challenges (19.5UA/ml) and passed challenges (12.1UA/ml). Higher SPT positive reaction to CM and higher complication rate of bronchial asthma were recognized in failed cow's milk challenge group.

Conclusion: SBFC for HE and CM was useful for the diagnosis at the time of entrance to primary school. To improve the quality of life of raw egg allergy patients, it is recommended for the patients to receive heated egg challenge. It seems that HE challenge test tended to induce GI reactions more than CM challenge and that persistent CM allergy may have something to do with the complication of bronchial asthma in school age children.

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Cow's milk protein allergy presenting as necrotizing enterocolitis: a case report

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Background: Gastrointestinal symptoms are common manifestation of cow's milk protein allergy. How ever necrotizing enterocolitis resulting from cow's milk protein allergy is rare.

Case report: A 3,300 gm. full term female infant had been fed with cow's milk formula since birth for ten days then switched to breast feeding alone. At twelfth day of life, she gradually developed abdominal distension and vomiting 2-3 times a day and was brought to see the pediatrician. Her physical examination had no other abnormal finding exept abdominal distension. So, she was advised to got some simethicone for abdominal discomfort and continued breast feeding. She still had the same symptoms that was vomiting and progressive abdominal distension. At twenty fourth day of life, she had developed more frequent vomiting, mucous bloody diarrhea, severe abdominal distension, hematochezia and pneumatosis intestinalis was seen on her X-ray finding. Laboratory finding included normal complete blood count, negative blood and stool culture. Necrotizing enterocolitis was diagnosed and successfully tolerated with parenteral antibiotics. She was placed on extensive hydrolysate formula which was well tolerated. The prick skin testing, atopy patch testing and radio-allergosorbent testing were all negative for cow's milk protein. Three months after, she grew very well with normal weight and height. She was challenged with soy-based formula at four month of age and well tolerated. At seventh month of age, she was challenged with cow's milk protein and developed vomiting, abdominal distension within four days. The diagnosis of cow's milk protein allergy was made based on positive symptoms after challenge.

Conclusion: Cow's milk protein allergy should be evaluated in patient presents with necrotizing enterocolitis without obvious risk factors.

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Calcium dependence of IgE reactivity of recombinant alpha-lactalbumin, a major cow's milk allergen

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Background: Cow's milk allergy is one of the most common causes of IgE-mediated hypersensitivity reactions in the first years of life affecting about 2.5 to 6% of infants. Cow's milk contains more than 25 proteins and several of them are known to be allergenic. α-Lactalbumin (α-La) is a major cow's milk allergen of the whey fraction. It is a small (14.2 kDa), acidic (pI 4.8) protein of 123 amino residues with a single calcium binding site.

Methods: We expressed α-La as a recombinant, histidine-tagged protein (rα-La) in *Escherichia coli*, and purified it to homogeneity using Ni-NTA agarose. The molecular weight and the structure of rα-La were studied by mass spectrometry and circular dichroism (CD), respectively. The IgE reactivity of rα-La was analyzed with sera from cow's milk allergic patients. Biological activity was tested using a humanized rat basophil leukemia cell line that expresses the α-chain of the human FcεRI receptor. Calcium dependence of IgE binding of rα-La was analyzed by IgE immunoblotting.

Results: Purified rα-La showed the expected molecular weight of 15.1 kDa in mass spectrometry. The far UV-circular dichroism spectrum revealed that rα-La represents a well structured, mainly α-helical protein. Thermal denaturation experiments using CD showed that the protein retains most of its native-like conformation even at 95°C and thus pointed to an extremely high thermal stability. IgE immunoblots demonstrated that rα-La binds IgE from cow's milk sensitized patients and induces specific basophil degranulation. Furthermore, calcium depletion assays led to a reduction of IgE binding in sera from certain cow's milk allergic patients.

Conclusion: We produced an IgE-reactive, correctly folded and biological active rα-La. Calcium depletion experiments indicated the presence of conformational as well as linear epitopes. This recombinant allergen may be used for diagnosis of cow's milk allergic patients.

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Usefulness of Open Food Challenge (OFC) in the diagnosis of food allergy

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Aim: According to the epidemiologic surveillance in a middle-sized city in Japan, the estimated prevalence rates of food allergy are 5 to 10 % and 1 to 2% among infants and school children, respectively. However, these figures seem exaggerated because the diagnoses are reported by parents and almost based on RAST and/or skin tests. To find out real prevalence rate we

compared the results of RAST and open food challenge tests (OFC) among children who were under elimination therapy because of suspected allergy to some foods.

Patients and Methods: The objects were 218 children, 139 male and 79 female, aged between 3mo and 12yr, suspected of food allergy. From January 2002 to May 2007, they were challenged with foods which seemed to have provoked allergic symptoms previously or which doctors advised not to take because of high scores of RAST tests and eliminated before the challenge. The clinical symptoms after ingestion of foods were observed for 3 days, documented and evaluated thereafter. Symptoms were classified into three categories; none, mild and moderate.

Results: Foods challenged were egg white (61), egg yolk (58), cow milk (27), noodle (13), buckwheat (9), chicken (6), peanuts (5), soybean (4), tofu (3), mayonnaise (2) and others (30)(Figures in the parentheses are numbers of challenges). Positive and negative family history of allergic diseases was 84% and 16%, respectively. Symptoms after ingestion of foods were none in 78%, mild in 14% and moderate in 8%. No life-threatening symptom was noted. Specific IgE titers (RAST scores) against suspected foods were measured in 109 children. There was significant correlation between the symptoms and RAST scores (P$B!a(B0.017). On the other hand, 73% of high score cases (RAST score 5 and 6) were symptom free. These results show that there is discrepancy between Symptoms and RAST score titers.

Conclusion: OFC is a simple and reliable method for clinical diagnosis of food allergy in children.

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The effect of soy-based formula on clinical symptoms and the fatty acid compositions in Infants with atopic dermatitis

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Atopic dermatitis and food allergy commonly occur in early childhood. Especially infants with cow's milk allergy were reported to have the reduced growth and special nutritional needs. Formulations of soy-based infant formula have changed over the years to improve digestibility, the stability and availability of minerals, and protein quality. soy-based formulas are a rich source of essential fatty acids, linoleic acid and α-linolenic acid. This study was intended to investigate whether soy based formula has an effect on clinical symptom and the fatty acid composition in erythrocyte in infants with atopic dermatitis. The infants were recruited from pediatric allergic clinic in Hanyang University Guri hospital from September 2004 to August 2005. One group was supplemented soy based formula (SBF, n=21) and the other was supplemented hydrolyzed cow's milk as controls (HCM, n=20) for 12 weeks. The clinical severity was measured by using SCORAD index. To evaluate nutrition status of the subjects, anthropometric indices compared with Standard Value of Growth and Development for Korean children. Blood samples were collected to analyze fatty acid composition and immune parameters. Specific IgE measurement and Skin prick test was performed. The mean age of the SBF group was 9.0 ± 2.1 month, and the HCM group was 8.8 ± 1.9 month, and their SCORAD scores were mild or moderate states that significantly improved after experimental period (before SBF 40.9 ± 2.4, after SBF 17.2 ± 0.6, before HCM 45.7 ± 24.6 after HCM 13.3 ± 21.7). Although SBF grows consumed more α-linolenic acid (18:3, n-3) intake, eicosapentaenoic acid and docosahexaenoic acid were decreased after SBF. In the other hand, arachidonic acid (C20:4, n-6) was increased after SBF feeding (before SBF 8.3 ± 2.8, after SBF 8.8 ± 0.6), however, decreased after HCM (before HCM 8.6 ± 1.0, after HCM 8.2 ± 0.8). In conclusion, SBF supported to improve symptoms with normal growth and nutritional status in infants with atopic dermatitis but there were no evidences to improve fatty acid compositions although giving a number of ¥á-linolenic acid intakes. It might be suggested that Δ-6-desaturase was reported to decrease in atopic dermatitis, SBF could not show to recover this enzyme deficient condition in this study.

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Egg white-specific IgE levels as predictors of clinical reactivity in the follow-up of egg allergy

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Background: Our objectives were to describe the egg white proteins allergy survival time in our population and analyze the correlation between egg white IgE levels and oral contolled challenge test (OCCT) results.

Methods: We performed a retrospective study including 42 patients diagnosed to egg allergy in the first 2 years of life who were controlled at 3,4 and 5 years of age. In the annual controls skin-prick tests, determination of egg white-specific IgE levels and OCCT were carried out until tolerance developed. In different moments of the follow-up we determined cut-off points of egg white-specific IgE levels that provided a probability ≥ 95 of egg white tolerance, analyzed by Receiver-Operating Caharacteristic (ROC) curve.

Results: The median survival time of egg allergy was 48.7 months. The specific IgE levels cut-off points correlated with OCCT results differ in the different periods of follow-up. The egg white-specific IgE levels which were predictors of the clinical reactivity (positive predictive value (PPV) ≥ 95%) in the different periods were: between 25 and 36 months 1.52 kU/L, between 37 and 48 months 1.35 kU/L and between 49 and 60 months 2.59 kU/L. For these levels the negative predictive value (NPV) was 40%.

Conclusion: The monitorization of egg white-specific IgE levels in allergic children to egg white allows us to predict the clinical reactivity along the egg allergy follow-up. Egg white-specific IgE levels greater than those referred would not be allow the OCCT in our population, while with lower levels we would find tolerant patients.

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The analysis of the severity of sensitization to extensively hydrolyzed cow's milk formula in infants with atopic eczema dermatitis syndrome

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Background: Children with atopic eczema dermatitis syndrome (AEDS) are often sensitized to cow's milk protein. The treatment is based on the extensively hydrolyzed formula (EHF).

The aim of study was to analyse the severity of sensitization to EHF in infants with atopic eczema.

Materials and Methods: 16 children aged 1- 36 months with symptoms of atopic eczema and positive specific IgE to cow's milk proteins were enrolled to the study. The followed diagnostic tools were used to confirm cow's milk allergy: the concentration of total and sera specific IgE antibodies to milk proteins as casein, â-lactoglobulin and á-laktoalbumin, oral food challenge tests to hydrolyzed cow's milk formula performed following standards.

Results: All of children were sensitized to casein, 9/16 of them to á-laktoalbumin and 7/16 to â-laktoglobulin. The serum concentration of total IgE (mean 623,1 ±735,2 vs 88,21 ±124,2; p=0,0001) and SCORAD index (mean 60,01 ±25,14 vs 52,01 ±24,43; p=0,03) was significantly higher in the group of children with allergy to EHF, but no correlation between total IgE and SCORAD index has been stated. The IgE sera concentration to casein and whey proteins was noticed most often in second and third class (0,7-17,5 IU/ml). The number of plasma eosinophils (p=0,006) and white blood cells (p=0,005) was significantly higher.

Conclusion: Children with allergy to EHF demonstrated more severe symptoms of AEDS than those without allergy to EHF. The atopic background of clinical symptoms was more expressed in children with allergy to EHF. Amino-acid based formula should be recommended in therapy of children sensitized to EHF.

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Hypersensitivity to cow's milk protein in a premature infant manifested with feeding intolerance and hypereosinophilia

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Background: Cow's milk protein hypersensitivity is the most common food hypersensitivity in infants. The current diagnostic approaches include a thorough history taking, skin testing and measuring of serum specific IgE to suspected food, elimination of suspected food and food challenge test. Eosinophilia occurs frequently in premature infants and has been associated with total parenteral nutrition and with cow's milk protein allergy, a response to foreign antigen presented to body through gastrointestinal tract, airway or intravenous routes.

Methods: We reported a premature male infant, with gestational age of 33 weeks and birth-weight of 980 gram, who developed feeding intolerance to premature infant formula since 3 days old. He was treated as necrotizing enterocolitis and was stopped feeding for a few days which showed improving of the symptoms. Then he was started feeding again on 7 days old and had feeding intolerance with increasing of peripheral blood eosinophilia from 440 / mcL to 9,640 /mcL at the age of 26 days.

Results: The allergy consultation was appreciated for evaluating the cause of hypereosinophilia. After thoroughly excluding other causes of eosinophilia, the patient was suspected to be allergic to cow's milk protein. He was changed the feeding to extensively casein-hydrolysated formula. The specific IgE to cow's milk was done and revealed a value of 7.77 kUA/L with total IgE of 114.3 IU/mL. His symptom of feeding intolerance was improved with a good weight-gaining. The peripheral blood eosinophilia was gradually decreased to 2,220 /mcL within 5 days. One week later, the infant was reintroduced feeding with cow's milk protein formula for 3 days and developed vomiting, abdominal distension and retention of gastric contents with a striking peripheral blood eosinophilia to 16,592 /mcL. The feeding was changed back to extensively casein-hydrolysated formula and the symptoms of feeding intolerance were improved with decreasing of eosinophil count to 900 /mcL on the age of 60 days and body weight of 2,000 gram.

Conclusion: Herein, we report a case of premature infant with feeding intolerance and hypereosinophilia resulted from cow's milk protein hypersensitivity. Eosinophilia in premature infant may be one response to foreign antigen mediated through gastrointestinal tract resulted in food hypersensitivity.

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A case of cow's milk allergy, which was succesfully treated with an amino acid-based formula but not with breast feeding

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Background: Hematochezia in infancy should be considered as one of the symptoms of cow's milk allergy (CMA). The most effective treatment of CMA is complete avoidance of cow's milk protein.

Objective: We have experienced a male infant with CMA, who tolerated an amino acid-based formula but not breast feeding.

Case: The patient was a full-term baby boy initially raised with breast milk and small amount of CM-based formula for about one month. From one month of age, the patient was exclusively breast-fed. At 3 month of age, a supplement of CM-based formula was started on a pediatrician's recommendation, because of slight poor weight gain. After ingesting a CM-based formula, he developed visible rectal bleeding immediately. A presumed diagnosis of CM allergy was made and mother was instructed to stop CM-based formula and to avoid drinking CM. Because mucus blood mixed stool continued, he was admitted to our hospital for diagnosis and treatment. Physical examination showed he was no apparent abnormal findings. Stool culture was negative for routine pathogens. The number of eosinophil in stool smear was elevated. Serum IgE was 10 IU/ml and specific IgE for milk was positive. Lymphocyte proliferative responses to CM-based formula and breast milk were positive, but to an amino acid-based formula was negative. A skin prick test was positive for CM. Rectocolonoscopy up to the sigmoid colon was performed, showing macroscopic evidence of nodular lymphoid hyperplasia with small erosions in rectum. Histological examination of biopsy specimen revealed colitis with predominant eosinophil-infiltration in the lamina propria. Through these findings we considered that this is a case of CMA and that breast milk contained a small amount of CM. We asked mother to stop breast feeding and CM-based formula and started to an amino acid-based formula. After avoidance of breast feeding, resolution of hematochezia took place with in two days. At the age of 14 months, the patient grows well on an amino acid-based formula and food without CM protein.

Conclusion: It is difficult for mothers to avoid completely milk protein, because a lot of food contain CM as ingredient, and therefore breast milk may contain small amount of CM protein often. In this case, we successfully treated the patient with an amino acid-based formula without breast feeding. And he has no problem for his growth. An amino acid-based formula is a powerful tool of treatment of CMA in infancy.

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A prebiotic oligosaccharide mixture inhibits pathophysiological changes in mice orally sensitized against casein or whey proteins

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Background: Human breast milk is thought to protect the neonate from development of allergies. In particular, oligosaccharides in human milk may be beneficial. These prebiotics may have direct immunomodulatory capacities or exert their effects by enhancing growth of lactobacilli and bifidobacteria. To test the effects of prebiotic oligosaccharides on allergic sensitization, a diet containing a 2% oligosaccharide mixture of fructo-, galacto- and acidic oligosaccharides (GOS:FOS (9:1, 1,8%) and AOS (0,2%)) was tested in mice orally sensitized against casein and whey proteins. The prebiotic mixture mimicks the oligosaccharide composition in human milk.

Methods: For a period of 6 weeks, five week old female C3H-HeOuJ mice were sensitized weekly by gavage (i.g.) with casein or whey (20 mg), using cholera toxin (CT) as an adjuvant. Mice were fed with the control or prebiotic (GFA) diet, starting two weeks prior to the first sensitization. At week 7 the mice were challenged subcutaneously (ear) (10 μg) and orally (100 mg). Serum levels of total IgE and allergen specific IgE, IgG1 and IgG2a were measured. The acute allergic skin reaction was determined by measuring ear swelling. Furthermore, isometric contraction of the colon was determined to get insight into motility changes of the intestine.

Results: In the whey and casein sensitized animals the allergic skin reaction was significantly reduced in the prebiotic group. This was most obvious in the casein group (control vs GFA diet, 202 +/- 23 vs 73 +/- 11 μm; $p<0.01$). Casein and/or whey specific IgG1 and IgE levels were unaltered, while in whey sensitized animals IgG2a levels were enhanced by the diet (control vs GFA

diet; 0.11+/- 0.003 vs 0.40 +/- 0.13 A450; p<0.05). The hypocontractility of the colon of casein sensitized mice was restored to normal levels by the prebiotic diet (control vs GFA diet, 1342 +/- 404 to 3515 +/- 1106 mg; p<0.05).

Conclusion: A mixture of GOS/FOS/AOS oligosaccharides reduces systemic and local allergic symptoms in mice orally sensitized to cow's milk proteins, when provided during the sensitization phase.

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Balb/c mouse model of allergy to wheat gliadins: biochemical and clinical manifestations

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Background: Animal models demonstrating IgE of the same specificity as human IgE and clinical symptoms upon challenge close to those observed in allergic patients are of great interest for the understanding of mechanisms involved in the induction and regulation of allergy. Food allergy to wheat (FAW) affects 0.1 to 0.4% of the global population. Major allergens have been characterized in this pathology. Water/salt soluble albumin/globulin fraction contains major allergens for children with the atopic eczema/dermatitis syndrome (AEDS) and the insoluble w5-gliadins are involved in adults with wheat-dependent exercise-induced anaphylaxis (WDEIA).

Aim: The aim of this study consisted of developing a mouse model of food allergy to a gliadin extract, a wheat fraction containing major allergens by testing different protocols.

Methods: Three strains of mice, Balb/c, C3H/HeJ and B10.A, were immunized by 4 successive intra-peritoneal injections of gliadin extract. The influence of the dose of gliadins (10 and 20 µg per mouse) was evaluated on allergen specific IgE and IgG1 production, on the cytokines production by activated T lymphocytes and on the elicitation of an allergic reaction. The development of the allergic reaction was compared by testing the Th2 type cytokine production and the eosinophil influx in the bronchoalveolar lavage (BAL) fluids 24h after a single intra-nasal stimulation with a total gliadin extract.

Results: Balb/c mice demonstrated higher gliadin specific IgE and IgG1 productions than the other mice, whatever the dose administered (i.e. 10 or 20 µg). This was confirmed by the Th2 type cytokine profile produced by activated splenocytes. The significant eosinophil influx and the Th2 type cytokine produced in the BALs also showed the elicitation of the allergic reaction in Balb/c mice, whatever the sensitisation dose. Low to no elicitation was evidenced in B10.A and C3H/HeJ mice at the 10 µg dose.

Conclusion: This study demonstrates for the first time the interest of Balb/c strain as a mouse model of food allergy to wheat. Our project is to further characterise the fine specificity of induced IgE and to compare the responses obtained in humans and mice.

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The number of IgE-binding cells in stomach mucosa in patients with peptic ulcer disease and food allergy versus without food allergy

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Immunoglobulin E is the main antibody responsible for immediate type reactions. It can bind with FceRI receptors on mast cells and basophiles including those in the alimentary tract.

Aim: The aim of this study was to compare a quantity of cells connected with IgE antibody at stomach mucosa in patients with peptic ulcer disease and food allergy, versus group of patients with peptic ulcer disease and without food allergy The study group consisted of 33 patients with ulcer disease and food allergy. The control group was formed of 21 non-atopic patients with ulcer disease. We evaluate a quantity of cells connected with IgE antibody in gastric mucosa by method of direct immunofluorescence.

Results: The number of IgE-binding cells in stomach in patients with peptic ulcer disease and food allergy was average 15,55 instead in patients with peptic ulcer disease 8,52. We found statistical significant difference of quantity of cells connected with IgE between patients with peptic ulcer disease and food allergy, versus patients with peptic ulcer diseases and without food allergy (p<0,02).

Conclusion: This results confirm the important role allergic mechanisms in pathogenesis of inflammatory changes in stomach mucosa observed in patients with peptic ulcer disease and food allergy.

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Relationship between ulcerative colitis, Irritable Bowel Syndrom (IBS) and food allergy

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Background: Allergic diseases are common diseases that involve many clinical disciplines. Food allergy, the offending food and food additives are concerned problems for doctors.

Objective: to explore the relationship between ulcerative colitis, irritable bowel syndrome(IBS) and food allergy. [Method] During the periods of 2005 to 2007,30 patients with ulcerative colitis or IBS were tested food skin test, serum food specific immunoglobulin G(sIgG), ANA. CIC, C3, C4, CH50, ADNs.

Results: 13 patients have positive result to egg, 8 patients to wheat, 4 patients to tomatoes, 3 patients to beans, 3 patients to fish, 2 patients to shrimp, 2 patients to peanut, 2 patients to corn, 2 patients to beef. Most of them combined with more than two kinds of food. All of them were visited 3 months and 6 months after examination. After avoiding positive food for 3 months, 15 patients with colitis have improvement, abdominal pain relieved, After 6 months, 22 patients recovered.

Conclusion: It is valuable to test food sIgG. According to the elevated sIgG, these patients were asked to change food habits. 73.3% patients have improvement. 26.3% patients have no effect. The study helps us to know the close relationship between digestive disease and food allergy.

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The study of the Chinese herbal medicine-Lonicera extract from water solution on the ovalbumin- sensitized BALB/c mice

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Background: Few studies are available for treating food allergy by used TCMs.

Objective: To investigate the effects of the Chinese herbal medicine Lonicera on the ovalbumin-induced hypersensitivity BALB/c mice.

Methods: BALB/c mice sensitized with OVA were treated 4 hours after the first challenge and then twice daily for 10 days with different Lonicera concentrations extracted from water solution, i.e. 1g/ml as high dosage , 0.5g/ml as middle, and 0.25g/ml as low.

Results: Part I: The conditions of hypersensitivity mice were markedly improved by the high or middle Lonicera doses, such as reduced levels of serum OVA-specific IgE , inhibited the abnormal monocular cell proliferations, alleviated the degranulation and aggregation of intestinal mast cells, increased the percentage of intact mast cells and significantly reduction of

histamine release, as well as downregulated the ratio of IL-4/IFN-γ and promoted the mRNA expression of IL-10 and TGF-β in PLNMC. Part II: The counts of Bifidobacterium and lactobacillus of the stool samples of the sensitized mice were significantly increased by the high or middle Lonicera doses, and meanwill the counts of Enterobacteriaceae and Staphylococcus were markedly declined.

Conclusion: Our data first showed the immunological regulation effects to food allergy from the high or middle Lonicera concentrations extracted from water solution, such as anti-inflammation and immunoregulation of Th1/Th2 responses as well as activation of T regulatory cells. And the inbalance of intestinal microflora in OVA-induced allergic mice were improved by the Lonicera ext, which were helpful to improve the clinical manifestations of food allergy.

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Utility of fast in food allergy: a case report

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We describe the case report of a 29 years old man who experienced an episode of urticaria and angioedema 30 minutes after having taken 100 mg of nimesulide and having drunk a beer.

He was known to suffer from allergic rhinitis and food allergy (skin prick test were positive to: Dermatophagoides Pteronissinus, Dermatophagoides farinae, Grass pollen, olive tree pollen, birch pollen, artemisia, hazel tree pollen, peanuts, nuts). He had had urticaria and angioedema after the ingestion of peanuts and nuts. As regards the last episode, both the drug and the food were suspected to be the cause of the reaction, therefore we performed skin prick test to beer and malt (previously tolerated), which resulted positive; the FastImmune Basophil Activation Assay (Flow-Fast) for beer confirmed the skin prick test positivity; Fast for nimesulide was negative; to confirm this data the patient has been challenged to nimesulide that was tolerated.

According to our clinical and laboratory findings, we concluded that the diagnosis was urticaria due to beer allergy.

Fast reveals the presence of a very specific and sensitive cellular activation marker that permits to diagnose IgE-mediate reactions and pseudoallergies. In our case report has been helpful to confirm the positivity to beer allergen skin prick test and exclude a drug reaction.

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Therapeutic indications to amino acid-based formula in infants with atopic eczema dermatitis syndrome

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Background: Allergy to cow's milk protein is one of the casual factor of atopic eczema dermatitis syndrome (AEDS) in infants. Treatment is based on diet, generally using an extensively hydrolyzed formula (EHF) or amino-acid based formula (AABF).

The aim of study was to establish the therapeutic indications to amino acid-based formula in infants with atopic eczema.

Materials and Methods: 30 children aged 1-36 months with AEDS were qualified to the study. Allergy to cow's milk proteins was established by the measurement of total and specific IgE and verified by oral food challenge following standards. According to the obtained results infants were divided in two groups: 16 children with allergy to EHF and 14 children without allergy to EHF.

Results: In all of children allergy to cow's milk formula was confirmed. In the group of children with allergy to EHF, hypersensitivity to casein hydrolysates was confirmed in all of them, to whey hydrolysates in 9/16 of infants. Severe atopic dermatitis was recognized more often in infants with sensitization to EHF (SCORAD index: mean 60,01 ±25,14 vs 52,01 ±24,43 in the control group; p=0,03). Total IgE was elevated significantly in the group of infants with severe AEDS and allergy to EHF (mean 623,1 ±735,2 vs 88,21 ±124,2; p=0,0001).

Conclusion: Amino acid-based formula should be recommended in children with severe atopic dermatitis and allergy to extensively hydrolyzed cow's milk formula. Allergy to casein is the most common in infants with hypersensitivity to EHF.

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Assessment of birch pollen immunotherapy effect on pollen-food allergy syndrome to apple with in vitro passive sensitization assay

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Rationele: Polllen-apple allergy syndrome affects approximately 50% of birch tree pollen-allergic adults. Allergy to apple results from cross-reactivity between apple and birch pollen proteins. Subcutaneous immunotherapy with birch pollen was reported to improve apple allergy in a subset of adult subjects. Skin tests and serum allergen-specific IgE antibody levels correlate poorly with clinical expression of apple allergy in birch-allergic individuals. Oral food challenge to apple remains the most accurate diagnostic test, however, standardized protocol is not easily available and the procedure is labour-intense. We sought to determine whether RBL passive sensitization assay might be used to evaluate effects of birch pollen immunotherapy in subjects with pollen-food allergy syndrome to apple.

Methods: Birch-allergic individuals had apple allergy confirmed with double-blind placebo-controlled oral food challenge and were randomized to birch pollen immunotherapy arm or to control arm (no immunotherapy, continued standard medical management). RBL-2H3 cells transfected with human Fc? receptor were passively sensitized with sera from birch-allergic individuals with oral allergy to apple at 1:40 dilution and following overnight incubation were stimulated with serial dilutions (10-2-10-8) of in-house freshly prepared raw Granny Smith apple extract. N-hexosaminidase content in the supernatant was used as a marker of RBL degranulation and was expressed as a percentage of total degranulation caused by Triton X.

Results: Eight subjects completed birch immunotherapy course. Median baseline release was 2.6% (range; 1.2-19.5), not different from median post-immunotherapy release 4.7% (range; 1.3-20.5), p=0.9. There was a trend for decreased skin test responses to birch pollen extract (mean 13.1 mm pre-versus 10.3 mm post-immunotherapy; p= 0.21) and fresh apple (mean 4.4 mm pre-versus 3.3 mm post-immunotherapy; p=0.08). There was no significant difference between pre-and post immunotherapy birch IgE and Bet v 1 -IgE levels.

Conclusion: RBL cells passively sensitized with sera from birch- and apple-allergic individuals undergo degranulation upon stimulation with fresh Granny Smith apple extract. Mediator release to fresh apple extract is not different before and after birch pollen subcutaneous immunotherapy. The performance of the in vitro passive sensitization assay is affected by low concentration of major apple allergens in the extract.

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Development of the first multidisciplinary psycho-educational intervention for parents and children with food allergy

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Aim: Development of an evidenced based intervention programme to help children and parents to manage the biopsychosocial impact of food allergy on their everyday lives.

Methods: 6 focus groups with parents of confirmed food allergic children and 10 focus groups with food allergic children aged 5-13 years. Qualitative analysis using grounded theory was used to analyse transcripts.

Results: The transcripts gave rise to categories relating to risk perception, illness cognitions, coping strategies, understanding of health and well-being, meanings given to diet and food. Transcripts also revealed developmental, parent/child, and sex differences vital to creating a targeted and food allergy sensitive intervention. A psycho-education intervention was then developed and is currently being validated with strong initial findings.

Conclusion: Middle childhood is a period when problems with anxiety, low self-esteem, peer comparison, and malconstructive coping strategies emerge, leading to higher risk in adolescence. Our data suggests that an intervention at this critical point in the food allergy developmental perspective is vital to ensuring successful biopsychosocial adjustment in day-to-day life.

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A new questionnaire for food allergy

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Despite clinical expertise suggest that food allergy has a substantial burden on patients' life, little data is now available on its impact and treatment on quality of life.

The aim of our study is to evaluate patients? answers to the preliminary version of a QoL questionnaire specifically addressed to food allergy. During the development phase of the new tool, 32 items were administered to 27 patients (9 M, 18 F, mean age 40, SD 7,2) suffering from food allergy. Patients had to indicate, on a Likert scale with multiple options (1: not at all; 5: very much), how much they were been troubled by each problem.

Results: A relevant percentage of patients reported a important burden in many aspects of their life: Patients live in a state of alert: they are watchful to new symptoms? appearance (21 out of 27) and to what they eat (18/27), they are afraid to become sick when they eat something at risk (20/27) or a new foods (15/27). Patients refer to feel uneasy when they go out for dinner (12/27) and they are troubled because they have to read labels on every product (13/27). Another relevant problem is the diet that, due to avoiding the food, could be unbalanced (10/27). So food is a cause of concern for patients (14/27), and they can't enjoy food (10/27).

Conclusion: on the basis of our preliminary data, food allergy appears as a problem that deeply interfere with patients life.

918

Sensitization to latex in patients with food allergy

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The increased prevalence of latex allergy is associated with the increased usage of the latex materials and with the cross reactivity to latex and food allergens, due to the common determinants of the plants.

Goal: to reveal the cross sensitization to the latex allergen in the patients with the food allergy using the test of allergic alteration of leucocytes (TAAL).

Materials and Methods: 37 patients with the food allergy to fruit and 18 health care workers with glove-related skin symptoms (the itching, redness

and dryness, sometimes an oedema, of the hand skin) were included in the study. The diagnosis of latex allergy in the patients of the second group was confirmed by the positive use test (in 10 from 10 ones), the negative results of Contact Patch Test with rubber chemicals for type IV hypersensitivity and by revealing of IgE antibodies to latex in 6 from 7 sera with ELISA method. TAAL and skin testing with latex and fruit allergens (carrot, potato, apple, tomato, kiwi, mango, melon, avocado, apricote, banana, perch - "Stallergenes", France) were performed in all patients.

TAAL: 100 mkl of the each blood sample were incubated at 37oC during 30 min with 10 mkl of the standard solution of the ammoniated latex (Stallergenes), then micropreparates were prepared, stained with acrydine orange for luminescent microscopy and a percent of the alterated granulocytes was counted. The result was considered to be positive if the number of the alterated cells was more 10%. A sample of the same blood, incubated with 0.9% NaCl, was examined as a negative control.

Results: the positive results of TAAL and skin test with a battery of fruit allergens were observed in the both groups of patients. In the latex allergy patients the positive results of the TAAL and skin test with latex allergen were observed in 77.8 and 83.3%, consequently. The positive results of the TAAL with the same allergen were observed in 70.5% of patients with the food allergy, and 76.4% of these patients had positive skin tests to the latex. The high percentage of the sensitization to latex in the food allergy patients without clinical symptoms to latex, established by two methods, may be the evidence of the cross reactivity and the risk for development of latex allergy.

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Latex recombinant hevein b IgE antibody in atopic children sensitized to fruit, latex and pollen

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Rationale: The associations of latex and fruit oral allergy syndrome (OAS) with pollinosis have been reported in adult allergic patients. In childhood fruit allergy, however, no significant data had been collected on clinical features and frequency of cross-reacting IgE antibodies to latex. Latex antigen is clarified to hevein b1 to 13, and primary allergen to be related to clinical manifestation has been elucidated by IgE antibody determination for these recombinant hevein (rHev) b proteins.

Objective: The aim of this study was to identify the specific IgE antibodies to latex rHev b protein in fruit OAS children and latex allergy.

Methods: Twenty-six atopic children (aged 2-11 years, 5.6̆2.6 y.o.) with fruit OAS sensitized to latex and 3 latex allergy children (3-8 y.o.) with having operation history from early stage of age were enrolled in this study. Specific IgE Antibodies to food, pollen, latex, rHev b 5, 6.01, 6.02, 8, 11 were measured by CAP-FEIA (Phadia KK).

Results: The clinical manifestations of fruit OAS were due to banana (67%), melon (63%) and kiwi (20%). With a fruit specific IgE antibody of 26 cases, banana 80%, melon 78%, kiwi 54% were positive. All patients had IgE antibodies to latex (0.6~31 UA/ml) and Japanese cedar pollen. In 6 fruit OAS with clear latex elicitation history, latex IgE antibody levels were high and IgE antibody to rHev b6.01 were positive in 3 cases and rHev b6.02 in 2 cases. In 14 of 20 cases without latex allergic provocative medical history, IgE antibody to rHev b 8 (profiln) increased remarkably. Specific IgE to rHev b 5, 6.02 and 6.01 were clearly positive in 3 cases of latex allergy with operation history.

Conclusion: The results provide evidence that food allergy infants with fruit OAS have the risk of early sensitization to latex and pollen. Measurement of specific IgE to latex rHev b6.01 and 6.02 was useful as a prediction of latex anaphylaxis in fruits OAS children with latex IgE antibody.

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Turnip/oilseed rape and mustard allergy in Finnish and French children with atopic dermatitis

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Background: Finnish children with atopic dermatitis (AD) are frequently sensitized to turnip/oilseed rape and French children to mustard. These plants belong to the same *Brassicaceae* family and their seeds contain homologous 2S albumin allergens. We examined the relationship between the oilseed plant and mustard allergy in the two patient populations.

Methods: 14 Finnish and 14 French AD children (mean ages 3.8 and 4.7 years) with positive skin prick test (SPT) to turnip rape were challenged with seeds of turnip rape and mustard. IgE antibodies were measured by ImmunoCAP and ELISA, and cross-reactivity examined by ELISA inhibition. 28 age- and sex-matched children with negative SPT to turnip/oilseed rape served as controls.

Results: Labial or oral challenge to turnip rape was positive in all (100%) Finnish and five (36%) French children. Mustard challenge was positive in five Finnish and five French children. IgE antibodies to oilseed rape and mustard were frequent (71% - 100%) in the Finnish and French children but uncommon (4%) in the controls. IgE levels to 2S albumins were increased in most Finnish and French children, and IgE inhibition experiments showed similar cross-wise inhibition patterns.

Conclusion: The present results in children from Finland and France show that turnip/oilseed rape, mustard or both are potential sensitizing allergens due to highly cross-reactive 2S albumins. Vegetable oils made from the seeds of oilseed/turnip rape are, in contrast to mustard, often added to foods but whether these contain the 2 S albumin allergens are at present not known.

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Nationwide survey of immediate type food allergy in Japan

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Objective: The food labeling system for food allergens was introduced from April 2002 in Japan. To confirm the effectiveness of the system, we regularly conduct a nationwide food allergy survey every three years.

Methods: The survey was conducted in cooperation with 1190 volunteer allergists in Japan form January to December in 2005. We sent questionnaire to contributing doctors every three months based on the past survey system, and contributing doctors were asked to report immediate type food allergy cases seen by those doctors. In this survey, immediate type food allergy was defined as the patients who had developed symptoms due to food allergic reaction within 60 minutes after intake of offending food. The details of questionnaire consisted of age, sex, cause of food allergy, symptoms, IgE CAPRAST, and type of onset.

Results: A total of 2283 immediate type food allergy cases were reported by the doctors. The most common offending foods were hen's egg (39.5%), milk products (18.1%), wheat (8.7%), salmon roe (4.5%), peanuts (4.2%), shrimp (3.2%), buckwheat (3.2%), kiwi fruit (1.8%), soy bean (1.7%) and crab (1.4%). The most common clinical symptom was observed on skin (91.2%) followed by respiratory system (31.3%). Interestingly, the causes of food allergy were completely different from infancy (egg, milk, and wheat) to adulthood (crustacean, wheat and fruits). Anaphylactic shock was observed in 10.1% of the total reported cases. The cases of anaphylactic shock were due to hen's egg (26.1%), milk products (20.0%), wheat (18.7%), peanuts (6.1%), buckwheat (3.5%) and shrimp (3.5%). Ten percentages of patients had been hospitalized and 38.9% of the patients had developed food allergic symptoms by accidental intake of offending foods.

Conclusion: We revealed the current condition of the immediate type food allergy cases seen in Japan in 2005. Based on these data, countermeasures

against food allergy are on going in collaboration with the Ministry of Health, Labour, and Welfare in Japan in order to improve quality of life of patients with food allergy.

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Investigation of peanut processing and its effect on allergenecity in Asian and North American populations

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Aim: Peanuts are cultivated over almost the whole world. Despite the widespread consumption of peanut products, the prevalence of peanut allergy is varying between different areas. Peanut is, prior to consumption, subjected to different kind of processing in different parts of the world. In this study the effect of different treatments, on a variety of peanut cultivated in North and South America, trying to mimic different eating habits was investigated. Finally the IgE-binding patterns to untreated and roasted peanuts were studied in two different populations from US and Asia.

Methods: Peanut source materials, Jumbo Runner, from US and Argentina were used. The peanuts were treated in three different ways; heated (90C, 20 min), boiled (20 min), or roasted (180C, 20 min) followed by grinding and extraction. Untreated peanut was used as control. The proteins were separated on both conventional SDS PAGE and 2D gel electrophoresis. Some spots from the 2D electrophoresis gel were excised and analyzed by MALDI-TOF MS. Immunoblotting analyzes were performed with sera from US and Asia from individuals with peanut sensitization.

Results: Differences in protein patterns between the two source materials were detected on SDS-PAGE electrophoresis. One prominent protein band of approx 40 kD, in material from US was missing in the peanuts from Argentina. As expected a decrease in protein amount dependent on treatment and temperature was detected. Distinct spots in 2D electrophoresis at approx 60 kD from untreated materials, were not visible after roasting. These spots were identified as major peanut allergen Ara h 1 by MALDI-TOF MS. No obvious differences in IgE-binding in Immunoblotting between the two populations could be observed. For both populations there was a decrease in IgE-binding to roasted compared to untreated peanuts, especially at approx 60 kD (Ara h 1) and at 30-40 kD. Other differences in IgE-binding patterns were due to individual differences.

Conclusion: Peanuts cultivated in different parts of the world may contain different proteins and allergens. Different treatment and cooking habits can influence the allergenicity. In this study no obvious differences in IgE-binding patterns between the two populations were observed even though the populations were chosen to reflect the different eating habits of peanuts. The clinical relevance of the missing protein in peanut source materials from Argentina need to be further investigated.

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The outcome of food allergy among infants and children in Israel

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Background: Food allergy is common among infants and children. Milk, eggs and soy allergy tends to resolve early in life while allergy to peanuts, tree nuts and fish usually stay for life. The knowledge about the natural history of sesame allergy is scarce. We did a survey among the children that were treated in the allergy clinic in E. Wolfson medical center.

Methods: The survey was done based on the clinical records of children that were diagnosed with food allergy. A phone survey and a detailed questioner were filled. The emphasis was on the outcome of the child's food allergy.

Results: We found 234 children that were suspected to have food allergy. 54 (23%) of them were found not to have food allergy. The 180 (77%) patients with food allergy were found to be allergic to several foods: Milk-125, eggs-71, sesame-30, soy-23, peanuts-14, tree nuts-9, fish-3, beef and chicken-2, other foods-6. The children were followed up for an average period of 4.7 years. In 139 cases the allergy had resolved, in 93 it was still existing, and in 51 cases it was not clear whether the allergy had been resolved or not.

Among those with milk allergy, 86/125 (69%) were IgE-mediated (group I) and 39/125 (31%) were non IgE-mediated (group II). In group I there was clearly a more atopic background then in group II (P=0.0032). The allergy had been resolved in 35/39 (90%) among group II patients, as compared to only 32/86 (37%) in group I (P<0.0001).

Allergy to soy was found in 23 children, 21 of them were also allergic to milk and only 2 were allergic just to soy. The number of children with soy allergy among group II patients was higher than in group I. In 19/23 (83%) the allergy resolved. 30 children had allergy to sesame. In third of them the allergy presented as anaphylaxis. Most of the children with sesame allergy were found to have atopic background. 5 of them have a first degree relative with sesame allergy. In 9/30 (30%) the allergy had resolved in an average of 2.8 years.

Conclusion: The distribution of food allergens in Israel differs from other countries. We found that the atopic status and the outcome of IgE-mediated milk allergy.is significantly different compared to non IgE-mediated patients. This survey may help to understand many aspects of food allergy among children in Israel, and may help us provide appropriate guidance for the children and their families.

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Personal experiences of teenagers with food-hypersensitivity

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Background: Teenagers are a high-risk group for food-hypersensitivity (FHS) fatalities and engage in risk-taking in managing their condition. Existing research has identified anxiety and impaired quality of life in children with FHS and their parents. To date, however, research has not addressed what the experience of living with FHS is like for teenagers. This study therefore aimed to describe the lived experiences of teenagers with FHS.

Methods: Individual semi-structured in-depth interviews were conducted with 21 UK teenagers (aged 13-18 years) with a pre-existing diagnosis of FHS. Participants were hypersensitive to a range of foods and experienced a variety of reactions. Participants were excluded if they had a concomitant non-allergic disease. Interviews explored the lives of the teenagers generally with particular attention paid to the experience of living with FHS. Interviews were audio-taped, transcribed verbatim and were then analysed using a phenomenological approach.

Results: Four main themes were identified: *FHS as a way of life, managing FHS as an assessment of acceptable risk versus acceptable burden, living with FHS as coping with necessary burden, and alleviation/exacerbation of the burden of living with FHS*. For participants the process of managing FHS was not described as problematic but as *a way of life*. Managing FHS was described as negotiable rather than prescriptive and was based on *an assessment of acceptable risk versus acceptable burden*. Negotiating management did not however completely relieve the burden felt and participants felt that a necessary part of living with FHS was *coping with necessary burden* and a variety of coping strategies were employed to this effect. Participants also described ways in which the *burden of living with FHS is alleviated or exacerbated* both by them personally and by others whose behaviour affects their life with FHS.

Conclusion: The themes provide some explanations for why teenagers with FHS engage in risky behaviours and describe how teenagers cope with

managing FHS and what factors make living with FHS easier or more difficult. The themes indicate ways in which the lives of teenagers with FHS can be improved and therefore provide new information about living with FHS from the perspective of teenagers that will be of use to healthcare practitioners working with teenagers with FHS.

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Cow milk allergy fo associated to rhinitis atopic

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Background: The Allergy to the cow milk (CM)is one of the causes of manifestations allergic but common in the childhood. It has an excellent one I foretell since most of the cases they are solved from the 4 years of age. The associated clinical disorders to the allergy to cow milk in these children include atœpics conditions such as atœpics dermatitis, allergic rhinitis, asthma and other manifestations corresponding to foods allergies.

Objective: To determine the association of allergy to cow milk and persistent allergic rhinitis in smaller children of 4 years. Material and Method: 5 smaller children of 4 years with diagnosis of allergic rhinitis and allergy to cow milk through Clinical History evaluated themselves. They were made Prick Test with cow milk standardized protein extracts (Casein, A-lactoglobulin and B-lactoglobulin); Concentrated of Histamina 1mg/ml considering as positive control the one presence papula of 5 mm, and saline solution as negative control without presence of pápula. One quantified Ig and EspecÚfic to cow milk in International Units mililiter by means of enzimoinmunoensayo. To the clinical aims, the specific concentrations of IgE were evaluated according to the following data: 5 Ul/ml:nonsignificant ;0.51 - 1.0 Ul/ml:Low;1,1 - 5.0 Ul/ml: Moderate ;5,1 - 25.0 Ul/ml:High;25,1 - 75.0 Ul/ml: Very elevated;> 75.0 Ul/ml:Highest.

Results: Of the 5 studied children, the totality I present/display direct atopicos familiar antecedents; and manifestation of rhinitic symptoms from the incorporation of the cow milk like dietario supplement (Three children from birth and two as of the 2 months of life). Were levels of moderate specific IgE in 3 of the 5 studied children (two with supplement of CM from birth and one as of both months of life) presenting/displaying these prick positive for alpha and beta lactoglobulin and negative test for casein; with serum of IgE a superior to 100 UI/m. In other two patients of the five were levels of specific IgE to nonsignificant CM with low serum values of IgE to 50 UI/ml and negative prick test for cow milk proteins.

Conclusion: According to these results it is possible to be determined that the cow milk is present in the allergic rinitis of smaller children to the four years of age, and that his replacement could improve the clinical sintomatology of the patients with medication saving for such.

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Prevalence of peanut and tree-nut allergy in Singapore teenagers - estimates from a questionnaire survey, allergy testing and food challenges

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Introduction: Peanut and tree nut allergy can potentially be life threatening. Hence, awareness of its prevalence in the population would allow physicians to better address the issues of diagnosis, management and education.

Objective: To estimate the true prevalence of peanut and tree-nut allergy in Singaporean teenagers.

Methods: The prevalence of food allergy in 14 to 15 year old Singaporean children was evaluated using a standardized questionnaire which was distributed to students of randomly selected schools. Students who responded positively to these food allergies were administered an extended questionnaire to determine presence of convincing food allergy. A portion of those with convincing allergy symptoms were recalled for skin prick testing (SPT) as well as underwent a graded food challenge for those who consented.

Results: From a cohort of 8072 students, 6765 (83.78%) responded. The crude prevalence of self-reported allergy to peanuts was 0.90% and tree-nuts 0.63%. However, using the criteria for convincing food allergy, the prevalence fell to 0.33% for peanuts and 0.20% for tree-nuts. A proportion of subjects with convincing allergy symptoms agreed to skin prick testing, which showed low frequency of skin test positivity peanut 4.17% (1/24) and tree-nuts 13.3% (2/15). Finally, all food challenges (5 for peanuts and 2 for tree-nuts) carried out on 7 subjects with either a convincing history plus negative SPT or convincing history plus positive SPT, resulted in a negative outcome.

Conclusion: Our results indicate that the prevalence of peanut and tree-nut allergy in Singapore adolescents is low. Evaluation based on questionnaire survey alone overestimated the prevalence of peanut and tree-nut allergy in our population.

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IgE-mediated food allergies in Swiss infants and children

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Background: IgE-mediated food allergy is the most common type of adverse reaction to food in infants and young children. It presents with a wide spectrum of clinical reactions including life-threatening allergic symptoms. The study has been performed to determine the most prevalent food allergens inducing immediate clinical reactions in different age groups of Swiss infants and children. Furthermore we investigated if there are specific patterns of allergic reactions to individual food allergens.

Methods: Prospective analysis of patients referred from 2004-2006 for assessment of immediate type allergic reaction to food. The study has been carried out at the Food Allergy Clinic of the Children's Hospitals Aarau and Lucerne. Individuals presenting with atopic dermatitis and evidence of delayed reaction to food were not included. Diagnosis of food allergy was based on clinical history, skin Prick tests (with commercial extracts), Prick-to-Prick testing using native food and determination of specific IgE antibodies to food proteins by CAP-FEIA system. Food challenges were performed in patients without history of life-threatening anaphylaxis when the causative antigen could not be conclusively determined.

Results: 278 food allergies were identified in 151 children (67% male, 33% female) with a median age of 1.9 years (range 3 months - 15 years). Overall, the most prevalent food allergen is hen's egg (23.7%), followed by cow's milk (20.1%), peanut (14.0%), hazelnut (10.4%), wheat (6.1%), fish (4.3%), kiwi and soy (2.2% each). In infancy, the most common food allergens are cow's milk, hen's egg and wheat. In the second and third year of life, hen's egg, cow's milk and peanut are predominant whereas above the age of 3 years, peanut, hen's egg and fish are the most prevalent. Urticaria is the most frequent reaction to food allergens. However, beyond urticaria, there are different patterns of clinical reactions to individual food. Cow's milk and hen's egg allergies are mainly presented with gastrointestinal symptoms. Peanut and hazelnut with angioedema. Fish with both angioedema and anaphylaxis and wheat with both angioedema and asthma.

Conclusion: 8 allergens account for 83% of IgE-mediated food allergies in Swiss infants and children. In each age group there is a specific order of most

prevalent offending food proteins. Furthermore, there is a specific spectrum of allergic symptoms to individual food allergens.

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Immediate hypersensitivity to common food allergens–An investigation on respiratory allergic patients in Kolkata (Calcutta), India

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Food allergy may be defined as an IgE-mediated immune response to food proteins. Such studies have not been done systematically from Calcutta, India. The present study was therefore undertaken to record the sensitivity to commonly consumed food in patients of allergic rhinitis and asthma. Affected individuals belonging to the age group of 5-60 years were evaluated using a standard questionnaire and skin prick test results (SPT). A survey of 800 patients (410 males & 390 females) reporting to the Allergy Unit of the Institute of Child Health, Kolkata were selected for the study, conducted from May, 2006 to April, 2007. Out of them 684 patients (85.5%) with a history of food allergy, most of them (49.41%) are in 16-40 years, followed by 41-60 years (28.07%). 22.51% belonged to the age group of 5-16 years. Most of the patients with food allergy suffered with asthma (65.05 %), rhinitis and asthma (20.03 %) and skin allergies (4.97%) such as itching, eczema and urticaria. The foodstuffs that were found to elicit symptoms of hypersensitivity were egg, milk, wheat, pulses, vegetables, fishes and fruits. The patients between 17-40 years (Male: Female-1:1.19) were sensitive to prawn (61.5%), brinjal (43.42%), banana (40.57%), lady's finger (35.6%), papaya (33.34%), wheat (33.3%), egg (31%), cauliflower (26.3%), spinach (23.8%), beef (20.6%) milk (18.42%), lentil-*Phaseolus mungo* (11.76%), *Lens culinaris* (11.26%) and cabbage (11.53%). The age group of 41-60 years (Male: Female- 1:1.04) had skin reactivity to brinjal (55%), egg (41.6%), banana (40.54%) fish (38%), *Phaseolus mungo* (33.33%), cabbage (28.5%), wheat (23.63%), beef (22.2%) *Lens culinaris* (21.8%), milk (20.75%), spinach (20%) and tomato (17.95%) Patients below the age of 16 years (Male: Female- 1:1.33) were sensitized to brinjal (45.16%), prawn (44.4%), banana (40.67), spinach (39.1%), egg (37.3%), papaya (28.95%), lady's finger (28.8%), cauliflower (25%), milk (25%), wheat (21.73%), *Lens culinaris* (18.75%), *Phaseolus mungo* (18.1%), and cabbage (13.6%). Although the percentage of allergy to beef is quite high, it is restricted only among the Muslim and Christian communities of Kolkata. Thus, we see food hypersensitivity reflects different genetic factors, variations in cultural and dietary habit of each individual.

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Sensitization to nut allergens in Zimbabweans

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Background: Peanuts, *Arachis hypogea*, are the prominent sources of Immunoglobulin E (IgE) mediated allergic reactions to food items. They are highly recognised for their nutritional value of proteins and oils. The major allergens are Ara h 1 and Ara h 2 , which result in allergenic manifestations such as eczema, urticaria, angiodema, asthma and sometimes fatal anaphylactic reactions.

Objective: The aim of the pilot study was to establish whether Zimbabweans had nut allergy in general and peanut allergy in particular as well as to identify clinical entities associated with sensitization.

Methods: Patients for the study were selected upon review of sensitization to peanut allergens. Sensitization was diagosed using the Skin Prick Test (SPT) and the serological evidence of allergen specific IgE antibodies with the Euroimmune assay. Most patients who came to the allergy clinic had eczema, dermatitis and urticaria.

Results: 63 patients had nut allergies, peanut and hazelnut.16 patients reacted to peanut only with 6 children of 2-8 years and 10 adults of 21-72 years of age.22 patients reacted to hazelnut only, with 4 children of 1-7 years and 18 adults of 18-58 years. Cross-reactivity of peanut and hazelnut was noted in 25 patients with 10 children of 3-13 years and 15 adults of 17-68 years of age.

Conclusion: Allergen specific antibodies to peanut, hazelnut ,or both peanuts and hazelnuts were demonstrated. We observed overlap in reactivity of biologically different nuts one being a groundnut (peanut, *Arachis hypogea*) and the other one being a tree nut (hazelnut ,*Coryllus avellana*) in 40% of the patients, hence our interest in further investigations to establish whether it was cross-reactivity or that it was a mere co-sensitization between the two nuts.

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Peculiarities of a food sensitization of children with atopic dermatitis in Ukraine

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Background: A food allergy affects the considerable quantity of children because of their gastrointestinal tract immaturity and low immunologic tolerance. According to the recent data of investigations from 1/3 to 1/2 children with an atopic dermatitis have a food sensitization. But the food sensitization is different in different countries because of the national food tradition and genetic factors. For example, there are not allergy reactions for peanuts and shellfishes in Ukraine but they are characteristic for American countries. From other hand, some food has changed its allergic properties. For example, all over the world it is marked increasing allergy to soy.

Objective: To find out peculiarities of a food sensitization for early aged and late school aged children with atopic dermatitis in Ukraine.

Methods: Sixty seven patients with atopic dermatitis were observed. The patients were divided into two groups: the first one (28 patients) aged from 3 month to 3 year and the second one (39 children) aged 12-18 year. The food sensitization was determined by the enzyme immunoassay method for the measurements of the level of allergen specific human IgE-antibodies in the blood for the first group and by a skin prick testing for the second group.

Results: The food sensitization was revealed in 92,9% and 71,8% patients of the first and the second group accordingly, including the high and very high reactions for 92,9% and 21,4%, monoallergy for 7,1% and 5,1% children accordingly. The high and very high levels of specific IgE were detected for antigens of a corn (42,3 %), a chicken egg protein (39,3 %), a fish (30,8 %), a rye (28,6 %), a chicken egg (26,9 %), oats (25 %), a cow's milk (23,1 %), a chicken egg yolk (21,4 %), rice (21,4 %), a tangerine (21,4 %), a lemon (21,4 %), and an orange (21,4 %) in children of the early age. The food tolerances for a corn, a rye, a cow's milk, oats, rice, a chicken egg protein, a tangerine are formed to late childhood. Certain types of food are more likely than others to be allergen for children of late school period: a chocolate (70 %), a tomato (26,3 %), a pear (23,3 %), a carrot (20,6 %), a potato (20,5 %). Nobody from 24 examined patients has sensitization to peanuts.

Conclusion: The established peculiarities of a food sensitization should be taken into account at initial preventive maintenance of atopi, and also they should be used for prescription of the empirical hypoallergic diet in Ukraine and the East Europe.

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Sensitization to parvalbumin and collagen in 6 Japanese patients with fish allergy

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Background: Fish has been reported to be one of the most important allergenic foods causing severe allergic reactions. Furthermore, parvalbumin, a small calcium-binding protein, and collagen have been identified as important cross-reactive allergens in fish.

Objective: To clarify the clinical and laboratory features of allergy to fish and to detect the cross-reactive allergens by ELISA in Japanese patients.

Methods: Six patients with suspected hypersensitivity to fish were subjected to skin prick tests and measurement of specific IgE antibody to fish. Additionally, specific IgE against purified parvalbumin and collagen were investigated by ELISA.

Results: The patients'(1 male, 5 females) age ranged from 8 months to 22 years. All of them also had other atopic diseases, such as atopic dermatitis, bronchial asthma and pollinosis. The diagnosis based on symptoms after fish ingestion and results of skin prick tests, was anaphylaxis and oral allergy syndrome (OAS) accompanied with symptoms other than oropharyngeal symptoms in 2 and 4 patients, respectively. The causative foods were saury, yellowtail, and flatfish in 3 patients; and horse mackerel, mackerel, tuna, salmon, Atka mackerel and flatfish in 2 patients. The SPT was positive for more than three kinds of fish in all patients. The immuno CAP-RAST test showed that 5 of 6 patients were positive for cod; 3 for flatfish; 4, for horse mackerel; 5, for salmon; 4, mackerel. Furthermore, specific IgE against the purified parvalbumin and collagen was detected in 4 patients each.

Discussion: The results indicated that our patients with fish allergy tended to develop severe symptoms after ingestion of different kinds of fish due to sensitization to cross-reactive allergens.

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Induction of oral tolerance by breastfeeding

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Background: We found that significant amounts of food proteins were secreted into human and rat breast milk as immune complexes with sIgAs. To clarify a physiological consequence of this phenomenon, animal experiments have been carried out in view of allergy prevention.

Results: SD rats were divided into two groups, one group (E-group) fed only egg white proteins and the other (M-group) fed only cow's milk proteins as a dietary protein source. When infant rats fed their own mother's milk were immunized with egg white proteins by using Freund's complete adjuvant, serum concentration of anti-ovomuciod IgG was lower in E-infants than in M-infants. When infant rats were immunized with cow's milk proteins in turn, serum concentration of anti-casein IgG was lower in M-infants than in E-infants. Thus, the immune tolerance against food proteins was acquired via breastfeeding from mothers fed the food containing the relevant proteins. The acquired tolerance against ovomucoid diminished spontaneously after weaning while the infants did not feed egg white proteins. Similar immune tolerance via breastfeeding were also observed in BALB/c mice.

Conclusion: We propose the hypothesis that breastfeeding, probably through the immune complexes of food proteins and sIgAs, serves as the natural drinkable vaccine against food allergy.

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Oral desensitisation in children with immunoglobulin e-mediated hen's egg allergy

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Background: Basic treatment for food allergy has been to avoid the offending item, to date. This approach is difficult in the case of common foods, and where there is a risk of severe reaction, due to child's and child's family concern of consuming the offending food, even inadvertently. In this study preliminary data regarding 3 children allergic to hen's egg (HE) orally desensitised with increasing doses of this food, are shown.

Objective: To desensitise children with severe IgE-mediated HE allergy in a period of 6 months by introducing increasing daily doses of HE in order to enable the child to assume 25 ml of HE daily (corresponding to 3.2 protein grams), or to induce tolerance of the highest possible HE dose.

Study Design: These are preliminary data of a larger study concerning children at least 6 years old with severe IgE-mediated HE allergy. A positive DBPCFC with HE confirmed the diagnosis. Oral desensitisation was done with increasing doses starting from 0.06 mg of HE proteins. Specific IgE to HE were evaluated through allergenic molecule-based approach with a proteomic technology (VBC Genomics microArray).

Results: The 3 children achieved the daily intake of 25 ml of HE during a 7 month mean period. During this period a reduction of IgE values (kUA/L) for Gal d 1 (Ovomucoid) was observed in all cases (in one case with negative result after desensitization). No significative differences were seen comparing the result for Gal d 2 (Ovalbumin), Gal d 3 (Conalbumin) and Gal d 4 (Lysozim)(see table).

Conclusion: We successfully desensitised 3/3 children with IgE-mediated HE allergy in a mean period of 7 months. Moreover, we dramatically reduced the risk of severe reactions after accidental or unnoticed introduction of low quantities of HE. We observed a parallel reduction of specific HE IgE against Gal d 1. We do not propose generalizing this method beyond trained staff.

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Systemic anaphylaxis induced by lupin ingestion: two case reports

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Background: IgE-mediated food allergy is an important cause of anaphylaxis, approximately 6% of young children and 3.7% of adults have a food allergy. The most frequent finding in food allergic patients are: peanuts, tree nuts, milk, egg, crustaceans and fish.

	Child 1		Child 2		Child 3	
	Start	End	Start	End	Start	End
Gal d1 (Ovomucoid)	0,34	0	5,81	2,51	2,89	1,45
Gal d2 (Ovalbumin)	0,3	0,31	0,92	0,9	0,77	0,45
Gal d3 (Conalbumin)	0	0	0	0	0	0
Gal d4 (Lysozim)	0	0	0,4	0,39	0	0

Case Reports: We report the cases of two young women referred to our department from emergency room for food induced anaphylaxis. The first case occurred in a 42 year-old woman after few minutes from eating lupins, with the development of cyanosis, inspiratory dyspnoea and sudden lack of consciousness. She was immediately treated with epinephrine and systemic steroids with remission of symptoms. The second case occurred to a 30 year-old woman showing anxiety, general itching, urticaria, vomit, facial angioedema, hypotension and syncope after ten minutes from assumption of gluten free cookies (patient was not celiac). The sudden administration of systemic steroids and antihistamines induced patient's recovery. Both patients underwent prick test for inhalant allergens showing positivity only in the second patient for dermatophagoides, birch, parietaria and grass pollens. No positivity was observed for common food allergens mentioned above. The two patients showed marked positivity to the prick-prick test with lupin. A deep analysis of cookies-label showed the presence of lupin flour. Previous reports described the sensitivity to lupin most of all in patients with professional exposition to this legume flour. In our case reports no specific risk factors to lupin allergy nor particular predisposition to food allergens sensitisation were evident.

Discussion: Lupin products are gradually being introduced into more and more foods including beverages, bread, biscuits and pasta as an additive to wheat flour substitutive of soy flour often present in food for people affected by celiac disease. In this contest allergy to lupin may be considered as an emerging condition, related to the hidden presence of this allergen or contamination, potentially dangerous for celiac patients.

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A case of food protein-induced enteropathy with a polypoid lesion

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Food protein-induced enteropathy (FPIE) is a cell-mediated hypersensitivity disorder commonly seen in the first several months of life, presenting with diarrhea and failure to thrive caused by ingestion of a certain food. A case of FPIE with a polypoid lesion has not been reported yet. We examined a case of FPIE by allergy-related blood and skin tests and endoscopy.

A two month old Japanese female patient showed repeated blood streaks in her stool. She was exclusively breast-fed and did not show any other symptoms. Food protein-induced proctocolitis was considered and the symptom was resolved temporarily by eliminating cow's milk from her mother's diet. However, she developed diarrhea at 3 months of age and began failing to thrive. At 4 months of age, gross bleeding in the stool was observed 16 hours after her mother ate cake and breast-fed. The stimulation index (SI) of the lymphocyte stimulation test (LST) for cow's milk was extremely high (783 %) and her mother stopped all dairy product intake. The patient experienced gross bleeding in her stool 24 hours after eating tofu at 6 months old. Her body weight did not increase for two months. The SI of the LST for soy beans was also high at 407%. She showed positive results in patch tests for cow's milk, soy beans, wheat and pacific cod, but did not show any positive results in skin prick tests. Also, antigen specific serum IgE levels did not rise. Fecal examinations were repeated but eosinophils and neutrophils were not found and microbiological results were all negative.

For further investigation, we performed a colon endoscopy under general anesthesia. Lymphoid nodules and mucosal edema were found in the colon. A 7 mm polypoid lesion was found on the ileocecal valve and juvenile polyposis was considered. However, the biopsy specimens showed non-specific inflammation with massive lymphocyte infiltration and did not indicate a solid tumor. Food protein-induced enteropathy was diagnosed. She stopped breast-feeding and started to take highly hydrated cow's milk (MA-1, Morinaga Japan) and rice and vegetables as solid foods. She has not shown gross bloody stool since, and her body weight has increased dramatically. The endoscopic findings improved after 2 months of dietary treatment.

In infants with intestinal bleeding and failure to thrive, the influence of food allergies should be considered and the LST, patch test and an endoscopic examination should be performed.

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The investigation of cases which performed provocation test on Food-dependent exercise induced anaphylaxis for the last 8 years

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Background: Food-dependent exercise induced anaphylaxis (FDEIAn) is a severe systemic syndrome induced by physical exercise after food ingestion. In recent years there has been an increasing number of reports of FDEIAn, but the actual situation is not known well.

Objective: To investigate the symptom of FDEIAn and grasp the actual situation of provocation test we investigated about 19 cases that was doubted FDEIAn, and were admitted for the purpose of detailed examination for the last eight years.

Methods: Nineteen children (16 boys and 3 girls) 19 cases were enrolled in this study. Age at onset was from 8 to 16 years old (average age 11 years old). IgE was measured by CAP-FEIA (Pharmacia) and skin prick test was examined in need. Provocation test by combination of food and exercise test was performed for 18 patients and 4 of them with preadministration of aspirin.

Results: As for the symptom at the onset, facialangioedema or blepharedema was present in 12 cases and urticaria in 10 cases, respiratory symptom in 13 cases ,hypotension in 5 cases, unconsciousness in 4 cases. Intramuscular epinephrine injection was medicated for 4 cases. The causative food allergen was as followed, wheat in 8 cases, crustacea in 7 cases, vegetables / cone / milk / cheese for each 1 case, unknown in 4 cases. We could be able to estimate food antigen by medical history in 17 cases. Related food specific IgE was positive in 5 of them. Twelve cases were negative CAP-RAST ,and 8 of them showed positive SPT. Two cases were not be able to estimate antigen by medical history and negative CAP-RAST. And one of them showed positive SPT. Four cases in 15 were positive by provocation tests with combined foods and exercise. We performed administration of aspirin before ingestion of food and exercise for 4 examples, symptom was provocated in only one case.

Conclusion: Wheat or Crustacea was a major food allergen of FDEIAn, but few case were caused by vegetables or fruit. Combination of CAP-RAST and skin prick test was useful to specify the allergen. In our cases, a positive rate was 50% by combination of food and exercise test or with preadministration of aspirin. It is need to establish about laboratory methods on children with FDEIAn such as food loading dose, exercise methods, and premedication of aspirin in future.

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IgE mediated anaphylaxis to sesame

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A 70-year-old Caucasian man presented with suspected food allergy. In the past emergent medical help was needed, because of an acute systemic anaphylactic reactions, timely associated with sesame ingestion. 15 minutes after consumption of sesame-poppy-raisin sweets suffered the patient from generalised urticaria, facial and laryngeal oedema, severe hypotension and unconsciousness. Emergency treatment with intramuscular administration of epinephrine and high doses corticosteroids and antihistamines intravenously was needed. Previous allergic reactions were denied. The patient stated occasional contact to sesame throughout his life span and non-contributory further own and familial anamnesis. Skin prick tests to common inhalant allergens, preservatives and food allergens were negative. Prick-to-prick tests with poppy seed, raisins and wheat flour remained negative as well. Surprisingly, skin prick-to-prick test with sesame seed and sesame oil revealed a pronounced positive reaction. The specific IgE for sesame was highly positive (8,37 kUA/l) whereas total serum IgE (93,5 kUA/l) and basal serum tryptase level (3,1 µg/l) remained in the normal range. A double blind, placebo-controlled food challenge with incremental doses of sesame seeds was conducted. 15 minutes after distributing 100 mg backed sesame seed, acute anaphylaxis occurred. The IgE mediated mast cell activation was proven by increase of the serum tryptase level (17,5µg/l) two hours after the food challenge.

Prevalence of allergy to sesame (Sesamum indicum) with its major allergens Ses i 1-7 becomes more significant due to its increasing industrial importance. Sesame represents an insidious "hidden allergen", found not only in alimentary products, but also in cosmetics and medical products, especially sun-protection creams and solvents for intramuscular injections.

The incidence of food allergy is age-dependent, particularly affecting children and young patients. In elderly, anaphylactic reactions tend to be less severe and a primary manifestation is unusual.

Nevertheless we were able to diagnose the rare case of classic IgE mediated anaphylaxis to sesame with primary manifestation in a 70-year-old patient. This shows that the potential life-threatening severity of food anaphylaxis, especially to foods with pronounced allergenic properties like sesame, should not be underestimated, irrespective of the patient's age.

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Assessment of quality of life in children with food allergy

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Objective: The dietary life of children with food allergy (FA) is restricted very much, and their quality of life (QOL) of diet would be affected. Therefore we have to perform dietary management for such children, but we have not yet known how they are restricted in their dietary life. We conducted current study to clarify the problems of dietary life in children with FA in our outpatient clinic.

Methods: By questionnaire we examined QOL of diet (burden of the dietary life at home and outside, problems of nourishment, and parents' psychological burden) against the parents of children with FA or without FA (control group). In addition, only to parents of children with FA, we interviewed to qualify the troubles they had in the dietary life just after the diagnosis of FA and at the some time after starting elimination diet.

Results: A total of 544 patients was surveyed. In the QOL survey, 40.1% of the children with FA had different menu from the family's menu, whereas only 21.7% of the control subjects did. In addition, 69.0% of parents of children with FA had difficulty in making menu, and 96.8% of the parents paid attention to food labeling, and 40.1% of those felt an economic burden for purchasing foods for FA children. Furthermore, 54.1% of the children with FA were not satisfied with eating-out, whereas only 35.4% of control was not satisfied. Children with FA were satisfied with the quality of school lunch by 71.9% but 95.4% for control. The most prominent problem just after the diagnosis of FA was about "making menu / cooking" in all age group. It was also a problem for the mothers to wean FA babies during infancy. However, a tendency to diversify troubles by age was recognized after starting elimination diet, such as problems concerning "purchase foods and eating-out", or "school lunch".

Conclusion: The study revealed that children with FA were restricted in terms of the dietary life, and that their QOL was deteriorated compared with control group. Since we could obtain information of the dietetics of which children with FA and their parents desired, it is important to pursue appropriate dietary management for the children with FA.

LATE BREAKING ABSTRACTS - BASIC

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Imatinib decreases duodenal mast cell density and degranulation in epicutaneously-sensitized food allergy model of mouse

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Background: Balb/c mice, when sensitized epicutaneously with ovalbumin, had increased serum ovalbumin-specific IgE as well as IgG1. After repeated intragastric challenges (ig) with ovalbumin, mice developed acute diarrhea within 60 min of the 6. ig dose, whereas there was no diarrhea nor increased IgE or IgG1 in the control groups. Since the role of small intestinal mast cells was undisputable in this model, we wanted to evaluate the effect of imatinib on these cells. Imatinib (GlivecR) is a potent inhibitor of the most important growth factor receptor of mast cells, the c-kit.

Methods: Imatinib (100 mg/mouse) was given orally to Balb/c female mice (20-25 g) as a pre-treatment during each 4 sensitizations and every second day from the day 39 at the same time with the 10 ig ovalbumin (50 mg/mouse) challenges. Ovalbumin-specific IgE, IgG2A, and mucosal mast cell protease 1 (MMCP-1) were measured from the serum and histamine from the plasma with standard ELISA methods. Rectal temperatures were measured and diarrhea evaluated before and after the 6.-10. ig challenge of ovalbumin. Chloroacetate esterase (CAE) was used for general mast cell staining, for immunohistochemical staining MMCP-1 antibody was used. For double staining fluorescent method with MMCP-1 antibody and TUNEL methods were used. The total amount of stained cells was calculated from the whole cross-sectional specimens of duodenum.

Results: There were no significant differences in the levels of ovalbumin-specific IgE or IgG1 between the imatinib-treated and the vehicle treated ovalbumin-sensitized mice indicating that the treatment did not prevent the sensitization of the mice. The serum MMCP-1 levels at the sacrification were lower in the imatinib-treated mice than in the vehicle treated group (10100 ± 10967 and 14173 ± 11246 ng/ml, respectively). Imatinib decreased the number of CAE+ cells (59 ± 32 and 126 ± 53 cells/mm^2, $p < 0.05$) and MMCP-1+ cells (13 ± 15 and 65 ± 47 cells/mm^2), respectively. The combined MMCP-1 and TUNEL staining indicated that the duodenal mast cells did not go into apoptosis during the treatment.

Conclusion: Imatinib treatment indicates the important role of the intestinal mast cell population, CAE+ and MMCP-1+ cells, of which MMCP-1+ cells seems to represent the majority of intestinal mast cells in the mouse food allergy model.

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Identification and immunochemical characterization of a common allergenic pollen Cycas circinalis

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Objective: Cycas circinalis is a common garden plant. The pollen grains of Cycas circinalis are airborne and remain suspended in the air from May to June with a peak concentration at the end of June. The allergenicity of the pollen grains is very common among the local inhabitants, but has not been reported so far.

Materials and Methods: The pollen extract showed high (25.6%) skin reaction on 172 respiratory allergic patients. Sera from the subjects were tested directly by ELISA for estimating the allergen specific IgE. The results were expressed as P to N value (ratio of patient's sera in respect to normal one). The allergic fraction was separated in 12% SDS-PAGE.

Results: The results showed the presence of 14 components in the range between 14-100 kDa. Sera of eight patients showing P to N value greater than 3.5 were used for immunoblotting to identify the specific IgE binding protein. The proteins from the gel were transferred to PVDF membrane. Immunoblotting was performed using eight patients sera, where anti human IgE-AP conjugate was used as second antibody and NBT and BCIP as substrate.

Conclusion: The results showed two components of 20 kDa and 39kDa. Thus, the Cycas circinalis pollen grain is an important aeroallergen in India. The 20 kDa and 39kDa components are the major allergens present in the pollen extract.

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The correlation between SNPs of GATA-3 and Chinese with allergic rhinitis

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Objective: To discuss the correlation between SNPs of GATA-3 and Chinese with allergic rhinitis.

Methods: 109 patients and 112 normal control people were enrolled in this study. We detect SNPs of two sites of rs1269486 and rs2229360 with RFLP.

Results: We detected SNPs in both of the two sites. The genotypes of rs1269486 are G/G, G/A and A/A, and the genotypes of rs2229360 are C/C, C/T and T/T. The distribution of G/G in patients was significantly higher than that in control group (P□*f*0.01). The haplotype frequency of GC in patient group was significantly higher than that in control group (P□*f*0.05), and the haplotype frequency of AC in patient group was significantly lower than that in control group (P□*f*0.01).

Conclusion: This is the first study to evaluate the association between SNPs of GATA-3 and allergic rhinitis, and we found that the SNPs of rs1269486 was significantly different between Chinese with allergic rhinitis and normal control people. The SNPs of GATA-3 may influence the phenotype of allergic rhinitis in Chinese.

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A novel approach to IgE epitope mapping – virtual epitope mapping

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Background: The determination of cross-reactive IgE epitopes on allergens is a major challenge. Mapping of discontinuous (=structural) IgE epitopes by experimental methods is extremely time consuming and possible for only few allergens and epitopes. Most structural methods for epitope determination (e.g., co-crystallization of Allergen-Fab complexes, NMR-based hydrogen exchange or MS-based methods) work only with monoclonal antibodies and therefore epitopes can only be determined one at the time and never represent the polyclonal situation found in reality. The mapping of allergen specific sequence motives onto the surface has been used or prediction of putative epitopes. However, sequence homology (alone) may be a weak predictor for cross-reactivity, as structural conservation is bound to be a major determinant of cross-reactivity.

In order to improve the accuracy of the prediction of structural epitopes, we have developed a new approach combining the 3D-structural information and surface topology with the information on sequence conservation.

Methods and Results: The computational methods for structure based epitope mapping are integrated in a suite of programs which combine public domain programs for sequence and structure alignment with topological comparison algorithms and evaluation of chemical properties of the solvent exposed surface. A program suite consisting of several *Perl* scripts, embedded into a graphical user interface written in *Tcl / Tk*, provides automated surface comparison. The following steps are executed by the program:

- •Superposition of two or more user selected atomic models
- •Calculation of solvent-accessible surfaces of the aligned structures
- •Surface comparison (calculation of differences)
- •Assign similarity scores to surface elements (including topology, chemical similarity, H-bonding capacity, etc.)
- •Averaging of similarity scores for each amino acid residue
- •Graphical display of results

The program was tested with two families of cross-reactive allergens, where the structures of three or more members have been determined.

Conclusion: We developed a novel method of IgE epitope mapping based on the structural information of two ore more cross-reactive allergens. The method uses sequence conservation as well as topological similarity for the delineation of putative IgE epitopes. The graphical user interface allows for the interactive visualization of the epitopes.

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Exosomes in human blood plasma contain RNA – a genetic signal between cells at a distance?

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Background: Exosomes are vesicles of endocytic origin produced by many cell types and released in the extracellular environment as 30?100 nm large membrane vesicles. Exosomes are involved in different signalling events including antigen presentation to T-cells and development of tolerance. It has recently been shown that mast cell exosomes contain a substantial amount of functional RNA (Valdi et al., Nat Cell Biol 2007). The aim of this study was to investigate weather exosomes in human blood plasma contain RNA and/or DNA.

Methods: Peripheral blood plasma was derived from healthy blood donors and exosomes were isolated by a series of ultracentrifugations and filtrations. Purified exosomes were attached to anti-MCH class II coated dynabeads and analysed using FACS. The tetraspanins CD 63, CD 9 and CD81, known to be enriched in exosomes, were used as markers for exosome detection. We also performed electron microscopy (EM) to detect exosomes in the sample. RNA was precipitated using TrizolÛ according to the manufacturer?s protocol. To magnify the signal, the RNA was then converted to DNA using a random primer, and separated by agarose gel. We also confirmed the presence of mRNA in exosomes by using oligo d(t) primers for conversion of mRNA to cDNA.

Results: The isolated vesicles from plasma were identified as exosomes by both EM and flow cytometric analysis (FACS). The results from the EM showed exosomal structures with a size of 30?100 nm in diameter. The FACS

analysis showed the presence of MHC II, CD63, CD9 and CD81, all markers of exosomes. The exosomes also proved to contain significant amounts of RNA, but no DNA. Some of the exosomal RNA was identified as mRNA.
Conclusion: Our results show the presence of RNA-containing exosomes in human plasma. Since exosomes have the ability to fuse with cells and deliver their RNA to that cell, it is possible that the RNA in plasma exosomes constitute a genetic signal between cells at a distance.

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Identification of Cladosporium herbarum allergen by immunoblotting technique

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Background: Many studies on C. herbarum antigens has shown that it has an important role to produce of specific IgE in atopic individuals and exacerbate of clinical signs of these patients such as atopic dermatitis. The purpose of this study was to identify C. herbarum allergen by immunoblotting technique.
Methods: C. herbarum was cultured in Sabouraud?s dextrose agar. The grown C. herbarum were harvested and ruptured by liquid nitrogen and glass beads. Samples were centrifuged at 3000rpm in 15 minutes and then at 15500rpm (4C) in 2 hours and then supernatant were collected as crude extract. The crude extract was separated by Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). The separated proteins transferred to nitrocellulose filter and then socked with atopic dermatitis patient's sera. The responsive bands to IgE were revealed by antihuman IgE antibodies conjugated with enzyme in chromogenic substrate.
Results: In SDS-PAGE, the crude extract of C. herbarum showed 16 different protein bands with molecular weight between 15.1-110 kDa. The bands with 15.1, 18.4, 25.1, 36.3, 45 and 54 kDa identified as strong bands. In immunoblotting, the bands with molecular weights 15.1, 18.4, 42, 110 kDa were reacted to sera IgE from patients with atopic dermatitis.
Conclusion: The results of this study showed that the strong bands in SDS-PAGE had the most reaction with anti-C. herbarum IgE antibody in immunoblotting technique. So we speculated the intensity of bands in SDS-PAGE can affect on IgE response. In accordance to other studies we think the C. herbarum antigen can trigger allergic reaction in atopic dermatitis patients.
Key words: Cladosporium herbarum, Atopic dermatitis, IgE, Immunoblotting.

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Increased expression of nerve growth factor in allergic rhinitis

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Background: Allergic airway diseases are characterized by inflammation and hyperresponsiveness. There is evidence that, in the upper airways, hyperresponsiveness can be attributed to increased neural activity. Nerve growth factor (NGF) is a neurotrophin that has been shown to be an important mediator of neural responsiveness in the airways of bronchial asthma patients. Recent studies suggest that NGF may also be an important inflammatory mediator. Therefore, NGF could be a link between allergic inflammation and neural responsiveness However, expression of NGF has not been reported in the nasal mucosa of allergic rhinitis patients. This study was performed to investigate the expression of nerve growth factor (NGF) messenger RNA and protein and to localize the NGF protein in the nasal mucosa of patients with perennial allergic rhinitis and control subjects.

Methods: Inferior turbinate mucosa samples were obtained from 20 patients with perennial allergic rhinitis and 20 matched non-allergic patients. NGF messenger RNA was extracted from the inferior turbinate mucosae, and then reverse transcription-polymerase chain reaction was performed. Western blotting was used to analyze differences in expression levels of NGF protein between patients with perennial allergic rhinitis and normal controls, and the NGF protein was localized immunohistochemically.

Results: The expression levels of NGF messenger RNA and protein in the nasal mucosa were significantly increased in patients with perennial allergic rhinitis compared with those in controls. NGF protein was expressed in the epithelium, infiltrating inflammatory cells, and submucosal glands.

Conclusion: This study provides evidence of NGF expression in the human nasal mucosa and its increased expression in perennial allergic rhinitis. These results suggest that NGF may contribute to the pathogenesis of perennial allergic rhinitis.

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Upregulation of angiogenin in nasal polyps

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Background: The exact etiologic mechanisms leading to the formation of nasal polyps have remained largely obscure. However, the formation of new blood vessels (angiogenesis) may play an important role in the development of nasal polyp. Angiogenin, a heparin-binding 14 kDa plasma protein with angiogenic activity, plays an important role in inducing angiogenesis. The aim of the present study was to investigate the expression and distribution of angiogenin in normal nasal mucosa and in nasal polyps and to evaluate the implication of such expression as regards the development of nasal polyps.

Methods: Normal inferior turbinate mucosa was obtained from 20 patients undergoing surgery for augmentation rhinoplasty. Nasal polyps were obtained from 20 patients undergoing endoscopic sinus surgery for chronic polypoid sinusitis. Semi-quantitative reverse transcriptase-polymerase chain reaction was performed for angiogenin mRNA. Sections were immunostained by using specific antibodies for localization of angiogenin and quantitatively analyzed using computer-based image analysis. Western blot analysis was done.

Results: Using immunohistochemistry, moderate to high levels of angiogenin were mainly localized in the infiltrating inflammatory cells of nasal polyps, and faint staining was found in normal turbinate mucosa. Semiquantitative reverse-transcriptase polymerase chain reaction and Western blot analysis showed that angiogenin expression was increased in nasal polyps compared with that in normal turbinate mucosa.

Conclusion: The markedly increased expression of angiogenin in nasal polyp compared with normal nasal mucosa suggests that angiogenin may play a significant role in the formation of nasal polyp.

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Proteomic identification of known and novel allergens from house dust mite *Dermatophagoides pteronyssinus*

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Background: *Dermatophagoides pteronyssinus* is the most predominant house dust mite (HDM) species and the most important source of indoor allergens in Thailand. Currently, there is no subunit vaccine available for successful prevention of the allergy to HDM. Characterizations of all potential HDM allergens that cause IgE production in the allergic patients are needed for development of a proper vaccine. The present study was therefore conducted to globally identify potential allergens from *D. pteronyssinus* using a proteomics approach.

Methods: Proteins derived from *D. pteronyssinus* cells were resolved by two-dimensional gel electrophoresis (2-DE), transferred onto a nitrocellulose membrane, and subjected to immunoblot analysis using pooled HDM-sensitive patients' sera (n = 39) or pooled normal control sera (n = 5) as the source of primary antibody. The membrane was then probed with mouse anti-human IgE (α- chain specific) conjugated with horseradish peroxidase and immunoreactive proteins were then visualized using a chemiluminescence substrate. The corresponding spots in another parallel 2-DE gel stained with Coomassie Brilliant Blue that matched to these immunoreactive spots were excised and identified by quadrupole time-of-flight (Q-TOF) mass spectrometry (MS) and/or tandem MS (MS/MS).

Results: A total of 33 immunoreactive spots were observed using the pooled patients' sera, whereas none was detected with the pooled control sera. Among these, 23 were successfully identified by Q-TOF MS and/or MS/MS analyses, including Der p 1, Der p 2, Der p 8, Der p 14 and other proteins that had not previously been recognized as the HDM allergens.

Conclusions: Combining the conventional immunological method to the recent advanced proteomic technology allowed identification of a number of known and novel allergens of *D. pteronyssinus*. Further characterizations of these allergens may lead to successful development of an effective vaccine for prevention of the allergy to HDM.

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Immunomodulation by Salvia mirzayanii: Effects on the cellular and humoral immune response and the induction of apoptosis in lymphocytes

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This study was performed to examine the possible immunomodulatory effect of Salvia mirzayanii, a native herbal plant to Iran. Human peripheral blood lymphocytes were used as target cells, and cell proliferation was determined by [3H]-thymidine uptake. The results indicated a dose related change of proliferation in phytohemagglutinin activated cells. The extract at 0.1 to 10 μg/ml was stimulatory and at 100 to 200 μg/ml was inhibitory ($p<0.01$). Immunization of the extract-treated mice with the antigen using delayed hypersensitivity (DTH) skin test and antibody measurement showed an increase in the footpad thickness which the amount reached to the level of the control when mice treated with more doses of the extract. The antibody titer in immunized mice treated with 1 mg/kg of the extract was higher than the non-treated mice, whereas treating mice with 100 mg/kg of the extract decreased the titer to under the level of the control in both primary and secondary response ($p<0.007$). In flow cytometer analysis the presence of apoptotic cells in sub-G1 phase at higher concentrations of the extract in mitogen-treated human lymphocytes was observed. DNA fragmentation analysis confirmed the result of flow cytometry. In conclusion, these results indicates that immunomodulatory agents are present in the extract of S. mirzayanii and that the induction of apoptosis in lymphocytes might be the mechanism responsible for the inhibitory effect of the extract observed at higher concentrations.

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IgE-binding epitope analysis of Bla g 5, German cockroach allergen

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Background: Several allergens from cockroach have been identified. The information on their B cell and T cell epitopes are limited, which are important for diagnostic and immunotherapeutic approaches. This study was conducted to analyze the linear IgE-binding epitopes of Bla g 5, a glutathione S-transferase homologous allergen, using recombinant proteins.

Method: Full-length and five fragmented or truncated recombinant Bla g 5 were expressed (A, 1-100 amino acid residue; B, 91-201; Ba, 1-125; Bb, 1-150; Bc, 1-175). IgE reactivities of the recombinant proteins to the cockroach-sensitized subjects were assessed by ELISA.

Results: Twelve (37.5%) of 32 sera tested showed positive IgE reactivity to the full-length Bla g 5. Six serum samples were selected for the epitope study. Any recombinant proteins containing 1-175 amino acid residues were unable to bind human IgE while the full-length and B fragment were recognized by 6 sera tested. These results suggests that the C-terminal region between 176–201 amino acid residues contains most of the IgE-binding sites.

Conclusion: These findings will advance the understanding of the allergenicity of cockroach allergen, thereby contributing to the development of strategies for allergen specific immunotherapies.

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IgE binding capacity of peptide fragments of Bla g 2, German cockroach allergen

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Background: Cockroaches cause various respiratory allergic diseases such as asthma. Although Bla g 2 is one of the most important cockroach allergens, researches on B cell and T cell epitopes of Bla g 2 have not been performed yet. This study was conducted to identify IgE-binding epitope of Bla g 2 by using recombinant proteins.

Method: The full-length and five peptide fragments (A, aa 1-75; B, aa 71-150; C, aa 146-225; D, aa 221-300; E, aa 296-352) of Bla g 2 were generated by PCR and over-expressed in E. coli. IgE binding reactivity was compared with Pichia-expressed Bla g 2 (PrBla g 2) and E. coli-expressed Bla g 2 (rBla g 2) by enzyme linked immunosorbent assay (ELISA). And the IgE binding reactivities of the full-length rBla g 2 and peptide fragments were measured by ELISA using 38 serum samples of from patients with cockroach allergies.

Results: Half of 38 sera showed IgE reactivities to PrBla 2 and rBla g 2. And only two (5.3%) serum samples among them showed stronger reactivities to PrBla g 2 than rBla g 2, indicating that they are reactive to conformational or carbohydrate epitopes. All fragments showed IgE-binding reactivities (80% to A fragment, 50% to B, 100% to C, 30% to D, 40% to E). Especially, amino acid residues 1 to 75 and 146 to 225 are shown to be important for IgE-binding.

Conclusion: Various IgE-binding patterns to Bla g 2 in individual patients' sera were observed. This information on IgE-binding epitope of Bla g 2 could be helpful for precise diagnosis and treatment of cockroach allergy.

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Production of interleukin-5 and interleukin-13 by stimulated CD4+ T cells from children with wheezing

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Background: Interleukin (IL)-5 and IL-13 have been recognized as proinflammatory cytokines involved in allergic inflammation. However, the role of these cytokines in children with wheezing is unclear. Thus, we determined IL-5 and IL-13 production by CD4+ T cells in children with wheezing in order to better understand the role of T lymphocytes in the pathogenesis of wheezing disorders.

Methods: This study was conducted on hospitalized wheezing patients. Wheezers may be atopic or nonatopic depending on the presence of immunoglobulin E specific to several common allergens. The control group comprised patients who were admitted for noninfectious and noninflammatory diseases. CD4+ T cells were isolated from the peripheral blood of the patients and incubated with phytohemagglutinin (PHA) for 72 hours. The levels of IL-5 and IL-13 in the cell culture supernatant were measured.

Results: After stimulation with PHA, the CD4+ T cells of the atopic wheezers produced increased amounts of IL-5 and IL-13 with statistical significance ($P < 0.017$ and $P < 0.002$, respectively), while those of the nonatopic wheezers and controls did not.

Conclusion: IL-5 and IL-13 production by the CD4+ T cells of wheezers could play a role in the pathogenesis of wheezing disorders in children.

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Quantification of FcεRI-bindable human IgE using chimeric EGF receptor

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Background: Quantification of human IgE (hIgE) in the patients' sera is important for the diagnosis of allergy. Most of conventional assay methods of hIgE depend on the binding of anti-hIgE antibody and hIgE in the sera. However, the hIgE concentration in the sera quantified by such methods is not necessarily correlated to the activation of mast cells. One possible reason is anti-hIgE autoantibody in the sera, which may interfere binding of hIgE to high-affinity IgE receptor (FcεRI). Therefore, it is important to develop a new method which can detect hIgE actually contributed to the sensitization of mast cells. In this study, we have tried to establish new quantification method of surface-bound hIgE using chimeric receptor bearing the extracellular region of FcεRI α subunit and intracellular region of EGF receptor (EGFR).

Methods: The cDNAs encoding chimeric receptors with or without intracellular EGFR region were made by fusing cDNAs of FcεRIα and EGFR based on PCR. The transmembrane domains of the chimeric receptors were from those of both receptors. The cDNAs were inserted to a mammalian expression vector, and the plasmids were transfected to HLR-Elk1 cells, which are HeLa cell-derived cell line containing Elk1-transactivable luciferase gene. The expression of the chimeric receptors were determined by confocal microscopy and Western blotting. The phosphorylation of intracellular signaling molecules induced by hIgE sensitization and crosslinking with anti-hIgE was investigated by Western blotting. The crosslinking of the chimeric receptors were measured by luciferase activity.

Results: Confocal microscopic analysis revealed that the chimeric receptors with transmenbrane domain of EGFR were efficiently expressed on the plasma membrane of HLR-Elk1 cells, however, those of FcεRIα were less expressed on the cell. Addition of hIgE to the chimeric receptor-expressing HLR-Elk1 cells caused significant phosphorylation of the chimeric receptor and MAPK, and the phosphorylation was increased when anti-hIgE was applied. However, crosslinking of hIgE with anti-hIgE did not augment luciferase expression induced by hIgE addition.

Conclusion: The cells expressing FcεRI-EGFR chimeric receptors could be a useful diagnostic tool that detects binding of hIgE to the cell surface FcεRI. However, a new method to detect crosslinking of FcεRI should be developed.

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AllFam – the database of protein families of allergens

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Background: Along with the identification of a large number of allergenic proteins from diverse sources, it has become clear that most allergens belong to a limited set of protein families. Most currently available allergen databases such as the official list of allergens (www.allergen.org) group allergens mainly by source organisms and do not take into account evolutionary relationships among allergens. Hence, we established AllFam, a database of protein families of allergens.

Methods: Allergen data were downloaded from the Allergome (www.allergome.org) database. Protein sequences from the Uniprot (www.expasy.uniprot.org) database were compared to protein family definitions from the Pfam (www.sanger.ac.uk/Software/Pfam). The AllFam database combines these protein family data with exposure and source data from the Allergome. AllFam contains a fact sheet for each allergen family with information on biochemical properties and allergological significance.

Results: The Allergome database contained 831 allergen sequences not including isoallergens. The sequences were classified into 182 protein families. However, only 94 families contained more than one allergen. The most abundant allergen families were the prolamin superfamily that comprised several subfamilies, and the profilins, a ubiquitous group of cross-reactive plant allergens. The family of EF hand containing calcium-binding proteins included polcalcins from pollen and parvalbumins from fish and amphibians. Allergenic tropomyosins were found as food allergens in crustaceans and molluscs and as inhalative allergens in mites and cockroaches. A classification of allergens by source revealed a similar number of protein families among plant and animal allergens (72 and 79 families) and a considerably lower number of fungal allergen families (50). Grouping of allergens by route of exposure showed that the distribution of food allergens is biased towards a few abundant protein families. The database contained 64 families of food allergens with the seven most abundant families accounting for 55% of all allergens. In contrast, there are 125 families of inhalative allergens with the first seven families making up only 27% of all allergens.

Conclusion: The Allfam allows a quick overview of protein families of allergens and their distributions among allergens from different sources and with different routes of exposure.

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Aspergillus germ tubes induce higher cytokine responses in human bronchial epithelial cells in comparison with spores

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Background: Aspergillus species are ubiquitous saprophytic fungi that cause a variety of diseases, ranging from hypersensitivity reactions to flu-like pneumonia and life-threatening invasive aspergillosis. As the lung is the primary site of initial infection with airborne conidia, we investigated the innate immune responses of bronchial epithelial cells against different forms of Aspergillus.

Methods: Human bronchial epithelial cells were treated with equal numbers of killed spores or germ tubes of either *Aspergillus fumigatus* or *Aspergillus flavus*. Transcriptional activation of the cytokine genes in the presence of germ tubes or spores was determined using real-time PCR analysis. Secreted cytokines following treatment with the mentioned fungi were also measured by enzyme linked immunosorbant assay.

Results: Analysis by real-time PCR showed that inflammatory cytokines such as IL-8 and IL-6 as well as the proinflammatory protease, caspase-5 were strongly upregulated by both treatments in a dose-dependent manner. Consistently, germ tubes induced a stronger response than spores. TNF-α and β-2-defensin were induced by high a concentration of germ tubes, but not by spores. Furthermore, IL1 pretreatment highly induced the expression of β-2-defensin, IL8 and IL12.

Conclusion: Taken together, our results show that germ tubes of *Aspergillus fumigatus* and *flavus* are potent inducers of innate immune responses in human airway cells. Considering the presence of Aspergillus spores in the air, differentiation between transient spore contact and invasion, as represented by germ tube formation, is important in order to determine proper immunological response. Moreover, these results can also provide additional data in understanding pathophysiology of hypersensitivity reactions due to the aspergilli.

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Detection of fungal aeroallergens from wards of a teaching hospital

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Background: The importance of indoor bioaerosols has been emphasized in the last decades. It is the fact that hospital environment is often contaminated with fungal spores and control of the air quality in hospital wards has assumed great importance. Exposure to the spore floating on the air in hospital wards leading to appearance of several diseases that ranges from allergies to sever fungal infections in hospitalized patients. The aim of this study was to identify and evaluation of fungal air contamination rates in high risk wards of a teaching hospital in Shiraz, southern Iran.

Materials and Methods: A settle plate assay was used for air sampling. Petri dishes containing Sabouraud and Malt extract agar were opened and exposed in high risk wards of a teaching hospital and incubated for five to ten days. The colonies are then identified, enumerated, and reported as the number depositing in one square meter of surface rooms.

Results: A total of 528 microbial colonies were detected which 223 (42.23%) colonies yielded fungi. Fungal isolates belongs to 12 genera as follows: *Cladosporium, Aspergillus, Penicillium, Alternaria, Trichosporon, Aureobasidium, Mucor, Rhizopus, Chrysosporium, Curvularia, Trichothecium, Stachybotrys, Stemphylium, Acremonium, Bipolaris* and Yeast. 4.25 percent of rooms were as clean room and *Cladosporium* was the predominant fungal isolates. The highest and lowest fungal spore concentration were 1015 and zero colony forming unite in emergency and transplantation rooms, respectively.

Conclusion: *Cladosporium, Aspergillus* and many other fungi are the major aeroallergens and causative agent of nosocomial infections. Detection of these fungi from hospital area has a potential risk of infection to these diseases.

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TH1, Treg and TH2 immunoregulation through TLR2 and TLR4 by *Lactobacillus plantarum IS-10506* probiotics in reducing allergic reaction. Experimental study in mice subjects

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The purpose of this study is to clarify the mechanisms and involvement of Toll-like Receptor (TLR) 2 and 4 in reducing allergic reaction by *Lactobacillus plantarum IS-10506* (LIS) probiotics.

In the factorial design study, Balb/c mice subjects were randomized into 6 experimental groups and were sensitized with OVA. Probiotics (LIS and placebo) and TLR inhibitors (PD98059, LY294002 and placebo) are independent variable. The expression of innate immune response (TLR2 and 4, NFκBp65, NFκBp105/p50); the activation of adaptive immune response (IFNγ, IL-2, IL-4, IL-5, TGFβ and IL-10, sera OVA-specific IgG2a, IgA, IgE, ileum OVA-specific IgA and total IgE); and the allergic reaction (histamine and symptoms score) are dependent variable.

The TLR2 and 4, NFκBp65, NFκBp105/p50, IFNγ, IL-2, IL-4, IL-5, TGFβ and IL-10-positive cells were measured on histological slices from the small intestine of mice in all groups. Levels of sera OVA-specific IgA, IgE, IgG2a and ileum OVA-specific IgA were determined with an indirect ELISA. Measurement of total IgE antibodies in sera was done with Sandwich ELISA. Histamine levels were determined with a competitive ELISA.

After final OVA sensitization, the LIS groups did not experience the severe and fatal allergic reaction, however most of mice in control group did experience. There were significant modulation of the innate and adaptive immune response in the LIS group compared with the control group, except IL-4 and total IgE. In control group, the low level of innate immune response (TLR2 and 4, NFκBp65, NFκBp105/p60) was in conformity with TH2-skewed (TH1 < Treg < TH2) and IgE-mediated mast cell degranulation which produce histamine and clinical symptoms. LIS probiotics enhanced the innate immune response (TLR2 and 4, NFκBp65, NFκBp105/p60), formed the TH1-skewed (TH1 > Treg > TH2), and reduced the mast cell degranulation (decreasing the level of histamine and symptoms score). The involvement of TLR2 and 4 in the effect of the LIS probiotics on decreasing of allergic reaction was proved by the interaction effect between those probiotics and inhibitor TLR in reducing allergic reaction.

In conclusion, LIS probiotics is involving TLR2 and 4 in reducing allergic reaction through enhancement of the TH1 and Treg immune responses and forming a new TH1-TH2 equilibrium without reducing the level of TH2.

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Identification of interferon-inducible gene expression in fibroblasts derived from allergic and nonallergic patients

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Rationale: Recently, asthmatic tissue-derived cells are reported to retain their characteristic features such as steroid insensitivity even when cultured for a long time. In our previous study, we identified many differences in gene expression patterns containing interferon-inducible CXC chemokine genes in fibroblasts derived from allergic (AF) and nonallergic (NF) patients, although the fibroblasts were cultured and stimulated under the same conditions. Therefore, we determined differences in gene expression of IP-10/CXCL10 and I-TAC/CXCL11 between AF and NF.

Methods: Nasal fibroblasts were isolated from inferior turbinate mucosa from allergic (AF) and nonallergic (NF) patients. Cells were stimulated with IL-4, TNF-α, IFN-γ, IFN-β, poly I:C or their combination. After mRNA was extracted, the expression of IP-10/CXCL10 and I-TAC/CXCL11 was examined using real-time PCR.

Results: IP-10/CXCL10 and I-TAC/CXCL11 were more down-regulated when stimulated with IL-4 and TNF-α in AF compared to NF, whereas their expression levels were increased when treated with TNF-α alone in AF. In addition, we found mRNA expression of these genes was significantly more strongly up-regulated by TNF-α in AF than in NF.

Conclusion: We identified specific gene expression profiles containing interferon-inducible CXC chemokine genes in AF and NF. The expression of IP-10/CXCL10 and I-TAC/CXCL11 was up-regulated in AF compared to NF even after cultured for several weeks in the same culture condition, suggesting genetic or epigenetic modification may be involved in its mechanism. Additional studies are needed to clarify the exact mechanism underlying this finding.

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Novel two series of benzo[b]furan derivatives having potent and selective LTB4 (BLT1and/or BLT2) antagonist

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Leukotriene B4 is a dihydroxy fatty acid produced mainly by macrophages and neutrophils. LTB4 plays important physiological roles on leukocytes trafficking to the site of infection and clearance of invaded microorganisms. However, overproduction of LTB4 is reported to cause inflammatory diseases including bronchial asthma and inflammatory bowel diseases. Thus, much work has been done to prepare LTB4 antagonists for clinical use as anti-inflammatory drugs. Unfortunately, no antagonist has yet

been developed for clinical medicinal application. Recently, a novel cell surface receptor (BLT2) for LTB4 has been isolated and its molecular cloning has also been established. Current studies on LTB4 antagonists and its receptors (BLT1, BLT2) suggest the possibility of the development of new clinical drugs for treating arteriosclerosis, immunosuppression of allograft rejection in organ transplantation, psoriasis, cancer, and rheumatoid arthritis. Stable compounds with definite conformation should be favorable as antagonists. Thus, we prepared two series of benzo[b]furan derivatives, 2-alkylcarbamoyl-1-methylvinylbenzo[b]furans (1) and 2-(4-thiazolyl)-3-phenylbenzo[b]furans (2), and evaluated their inhibitory activity for BLT1 and /or BLT2. Some of them showed more potent and more BLT2 selective inhibitory activity than positive standard compound ZK158252. Especially, 2-(2-N,N-diethylcarbamoyl-1-methylvinyl)-7-(1-phenylethyl)benzo[b]furan ((E)-1c) and 2-[2-[(dimethylamino)methyleneamino]-5-formylthiazol-4-yl]benzo[b]-furans (2a) showed potent and BLT2 selective inhibitory activities. These compounds will be assay for treatment ability concerning immflamatory and asthma in vivo screening.

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Relation of IgE antibody to antigen 5 of echinococcus granulosus

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Anaphylactic reactions, such as urticaria, edema, respiratory symptoms, and anaphylactic shock often complicate the course of Cystic Echinococcosis (CE). To investigate the role of the IgE immunoreactive Antigen 5 (Ag 5) in the sero-positive patients with CE, we determined N-terminal of 57 kDa subunit of Ag5 responsible for IgE and C-terminal of this active antigen related to induction of IgG specifically. Immunoblotting analysis showed that specific IgE to 57-kDa subunit related to inter-chain disulphide band of two 22 kDa and 38-kDa component of Ag5 and conformational epitope on this subunits. In addition, IgE specifically recognized N-terminal of 22 kDa subunit which remain bounds to the other component, whereas IgG reacted with C-terminal of 38 kDa component of Ag5. Recognation of the specific binding site on the 57 kDa subunit of Ag5 could leads to understanding the mechanism regulating IgE/IgG production in some immune circumstances that IgE tends to some dominate, whereas in other IgG predominates.

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Effect of the complex of beta-glucan and Zataria multiflora on oxidative burst of neutrophils in BALB/c mice

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Background: Natural products, beta-glucan and herbal essences, have been found to enhance immune functions. The aims of this study were to purify beta-glucan from the cell wall of Saccharomyces cerevisiae and to investigate its conjugate effect with Zataria multiflora essence on oxidative burst in BALB/c mice.

Methods: After preparing the cell walls from the yeast, beta-glucan was extracted from wall by alkaline-acid treatment (glucan-S1). This preparation contained protein and mannan, which were removed by DEAE sephacel chromatography (glucan-S2) and con-A sepharose chromatography (glucan-S3), respectively. Glucan-S3, Zataria multiflora essence, and their conjugate were injected into BALB/c mice intraperitoneally. Blood collected at days 4 and 7 after injection and oxidative burst was assayed by chemiluminescence method. Results: The findings showed that intraperitoneally administration of glucan-S3, Zataria multiflora essence and their conjugate significantly increased oxidative burst of neutrophils both at days 4 and 7 when compared to control group ($P<0.05$).

Conclusion: These results suggest that the complex of à-glucan and Zataria multiflora essence can be used as adjuvant agents to stimulate immune functions in immunocompromised subjects.

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Effect of staphylococcal enterotoxin on inducible nitric oxide synthase in the mice with allergic rhinitis

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Background: According to recent reports, inducible form (type II) of nitric oxide synthase is expressed at higher levels in the nasal mucosa of allergic rhinitis patients, and these patients show a higher carriage rate of Staphylococcus aureus than normal adults.

Aim: Are to investigate the role of staphylococcal enterotoxin B in the development of allergic rhinitis and to determine its relevance with inducible nitric oxide synthase overexpression.

Methods: The nasal mucosa and serum were obtained from the sensitized and negative control groups, and the frequencies of allergic symptoms, such as sneezing, were compared. Eosinophil counts and inducible nitric oxide synthase expression in the nasal mucosa were examined. The serum levels of ovalbumin-specific IgE were measured by ELISA. Differences between the sensitized and control groups were statistically analyzed using the Kruskal-Wallis test and the Mann-Whitney U test.

Results: The frequencies of sneezing, serum levels of ovalbumin-specific IgE, and degrees of inducible nitric oxide synthase expression were significantly higher in the groups locally sensitized with a mixture of ovalbumin and staphylococcal enterotoxin B or ovalbumin alone than in the negative control group or in the group locally sensitized with staphylococcal enterotoxin B alone.

Conclusion: Inducible nitric oxide synthase may play a crucial role in the development of allergic rhinitis whereas staphylococcal enterotoxin B may not participate in this process.

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The anti-allergic effect of lactobacillus probiotics in mite-allergen sensitized murine model

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Background: Clinical trails have demonstrated that probiotics can improve the allergic symptoms but the mechanisms responsible for these benefits are still unclear. We sought to investigate the effects of probiotic organisms in a mouse model of allergic airway inflammation.

Methods: First, Balb/c mice were treated with two probiotic organisms Lactobacillus rhamnosus and Lactobacillus paracasei via gavaging needle for two weeks daily before Dermatophoides pteronyssinus (Der p) allergen sensitization and for another two weeks during sensitization and allergen challenge. Second, to know the dose effect of Lactobacillus paracasei, we further investigated the ability of three doses of Lactobacillus paracasei, $5 \times$ 107CFU/ml, 107CFU/ml and $3 \times$ 106CFU/ml, in the prevention of allergic airway inflammation.

Results: First, we found that treatment with Lactobacillus paracasei significantly elevated Th1 cytokine, IL-12 in bronchoaveolar lavage fluid (BAL) and decreased Der p-specific IgG1 in serum but there were no changes in airway hypersensitivity, influx of inflammatory cells to the lung and serum IgE level. Treatment with Lactobacillus rhamnosus did not have any significant effects. In the second part, treatment with medium dose (107CFU/ml) of

Lactobacillus paracasei significantly attenuated the influx of total leukocytes, monocytes and lymphocytes to the airway and elevated IL-12 in BAL, Der p-specific IgG2a in serum. Medium dose of Lactobacillus paracasei also decreased airway hyperresponsiveness and increased IFN-γ production in Der p stimulated splenocytes. The high dose of Lactobacillus paracasei reduced the influx of total leukocytes, monocytes in BAL and IL-4 production in Der p stimulated splenocytes, increased Der p-specific IgG2a in serum and IFN-γ production in Der p stimulated splenocytes. The low dose of Lactobacillus paracasei only elevated Der p-specific IgG2a in serum, IFN-γ production in Der p stimulated splenocytes and reduced the splenocyte proliferation activity.
Conclusion: Our results show that oral treatment with probiotics can attenuate allergic airway inflammation by increasing Th1 immune response and these effects are strain specific and dose dependent.

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Aerobiological and Immunochemical studies in Domjur, Howrah District, West Bengal, India

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Allergic diseases, like bronchial asthma, allergic rhinitis, conjunctivitis due to exposure of various airborne biological particles are very common. Pollen grains among them are known as a causative agent which cause respiratory allergy in susceptible individuals. In this program, a rural area at Howrah district, Domjur Block near Kolkata was selected for our aero-biological and clinico-immunological studies. The area was selected as it is rich in vegetation and a high density of population suffering from respiratory allergy. Aerobiological Study was done using Burkard sampler. Preparation of allergenic extracts was done using pure bulk of pollen grains using 0.1 M phosphate buffered saline (PBS). Skin tests (SPT) with different pollen extracts were performed following the method of Stytis (1982). Sensitive sera were collected from the patients visiting the outpatients' Department of the Allergy Clinic of the Institute of Child Health, Calcutta. Normal sera were obtained from healthy donors with no history or symptoms of atopy. Electrophoresis and Western blotting was analyzed by SDS-PAGE according to the method of Laemmli (1970). In order to prepare a pollen calendar, at Domjur, rich in vegetation with wide agricultural farming aerobiological survey was done. 47 air borne pollen types were recorded and identified from the air of Domjur. The dominant types were, Areca catechu, Cocos nucifera, Phoenix sylvesrtis, Riccinus communis, Lantana camara, Perthenium hysterophorus, Trema orientalis, Moringa olifera, Carica pa paya, Catharanthus roseus, Casuarina equisetifolia, Albizzia lebbeck, Cyperus rotundus etc. Among the dominant pollen types, some were subjected to Skin Prick Test. Areca catechu, Carica papaya, Catharanthus roseus, Cocos nucifera, Amaranthus spinosus, Borassus flabellifer, Lantana Camara were found to be highly positive. Carica papaya pollen was selected for the biochemical and clinical investigations. SPT reaction was studied with the relevant pollen extract on 320 respiratory allergic patients and 30% of them showed a positive response. The native polyacrylamide gel electrophoresis of Carica pollen and fruit showed the presence of a number of protein bands . In IgE specific dot immunoblotting of Carica pollen showed remarkable cross reaction with papain, an enzyme present in the latex, fruit and other plant parts.

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Nasal mucosal infiltration of eosinophil after nasal provocation

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The Background of the study: Eosinophils are found in the nasal lavage fluid and nasal biopsies of patients with allergic rhinitis after nasal antigen challenge. Numerous studies have been carried out to clarify the paticipation of eosinophils in a late-phase allergic reaction or airway hyperresponsiveness. However, there has been a few report describing in detail the role of eosnophils during an early-phase allergic reaction. The aim of this study was to investigate the degree of nasal eosinophil infiltration before and after nasal provocation test and to evaluate the changes of degree of nasal eosinophil infiltration according to the duration of allergic rhinitis.
Methods: One hundreds and forty-five allergic rhinitis patients who were diagnosed by allergic work-up were included. The patients were divided into three groups by duration of symptoms. Group 1 was the patients whose duration of symptoms was less than 2 years (n = 40), group 2, 2 to 9 years (n = 58), and group 3, more than 10 years(n = 47). Symptom scores of the patients were checked for nasal obstruction, watery rhinorrhea, sneezing, and itching with visual analogue scale. Nasal mucosal scraping and smear was performed with curette at before and 30 min after nasal allergen provocation. Degree of eosinophil infiltration was evaluated in each slide by 6-point scale. And degree of eosinophil infiltration was compared between before and after provocation. The degree of eosinophil infiltration was evaluated and compared according to the duration of symptoms.
Results: Prominent nasal symptoms of all 3 groups was nasal obstruction and it was increased by duration of symptoms. But other symptoms were more severe in group 2 than in other group. Group 2 and 3 showed higher degree of eosinophil infiltration compared with Group 1. And degree of eosinophil infiltration was increased after provocation in group 2 and 3.
Conclusions: The eosinophil infiltration is increased even in early stage of allergen provocation and increase of eosinphil infiltration in the nasal mucosa after provocation is more significantly in chronic state. Eosinophil may be involved in early reaction of allergy, especially in chronic state.

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Association of cytochrome P450 (2C9, 2C19) gene polymorphism with susceptibility to Ankylosing Spondylitis in a population group from Algeria

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Background: Cytochrome P450 (CYP) superfamily members CYP2D6, CYP2C9 and CYP2C19 are polymorphically expressed enzymes that are involved in the metabolic inactivation of several drugs, including, anti-epileptics, NSAIDs, oral hypoglycemics, and anticoagulants.

Many of these drugs have a narrow therapeutic index, and several studies indicates a prominent role of these enzymes polymorphisms in the therapeutic efficacy and in the development of adverse effects among patients treated with drugs that are CYP2D6, CYP2C9 or CYP2C9 substrates. Previous linkage and association studies has suggested the presence of a susceptibility gene for ankylosing spondylitis (AS) close to, or within, the cytochrome P450 2D6 gene which could play an key role in pathophysiology of AS. In our study, we examined whether common mutations of cytochrome P450 (CYP) [CYP2C9*2,*3, CYP2C19*2, and CYP2D6*3,*4,*6,*7 and *8] are involved in outcome of AS in Algerian patients and, find out and determine genotype frequency in Algerian population.
Methods: CYP genotype was determined in 160 AS patients [fulfilling the modified Naw York criteria] and 204 healthy non-related subjects. DNA isolated from blood samples was used for the analysis of CYP2D6, CYP2C9 and CYP2C19 allelic variants by allele-specific real-time PCR. Thus, with respect to allelic variant frequencies, genotype distributions or predicted phenotypes were deduced from genotype combinations.
Results: i) no significant difference was found in CYP2D6 allelic variants between AS patients and control subjects; ii) the CYP2C9*3 allelic variant is more frequent in AS patients than healthy subjects (pc=0,015,OR=2,98); iii)

CYP2C19*2 allelic variant is more frequent in AS patients than healthy subjects (pc=0,009, OR=2,06) ; iiii) and the frequency of the "intermediate metabolizers" genotype and/or phenotype for substrates of CYP2C19 is 2-fold more important in AS patients than controls (pc=0.0009).
Conclusion: It would exist a possible role of CYP2C9 and C19 polymorphisms in the risk for development of AS and probably on the metabolism of some drugs used in treating AS patients.

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Exosomes from bronchoalveolar fluid of tolerized mice prevent allergen-specific allergic reaction

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Background: Exosomes are nanovesicles originating from multivesicular bodies that are secreted by a variety of cell types. The dual capability of exosomes to promote immunity or to induce tolerance has prompted their clinical use as vehicles for vaccination against different human diseases. In the present study, the effect of allergen-specific exosomes from tolerized mice on the development of allergen-induced allergic response was determined using a mouse model.
Methods: Mice were tolerized by respiratory exposure to the olive pollen allergen Ole e 1. Exosome-like vesicles were isolated from bronchoalveolar lavage fluid of the animals by the well-established filtration and ultracentrifugation procedure, characterized by electron microscopy, western blot and FACS analyses, and assessed in a prophylactic protocol. To this end BALB/c mice were intranasally treated with tolerogenic exosomes or naïve exosomes as a control, one week prior to sensitization/challenge to Ole e 1. Blood, lungs and spleen were collected and analyzed for immune responses.
Results: Intranasal pretreatment with tolerogenic exosomes inhibited IgE response along with a marked reduction of lymphoproliferative response and Th2 cytokine production. Moreover, the pathologic alterations of the lung associated with airway inflammation, such as eosinofilia, were significantly suppressed.
Conclusion: These results demonstrate that exosomes can induce tolerance and protection against allergic sensitization in mice. Thus, exosome-based vaccines could represent an alternative to conventional therapy for allergic diseases in humans.

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Primary and secondary infection with Respiratory Syncytial Virus (RSV) before allergen challenge enhanced airway hyperresponsiveness in murine model

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Rationale: Respiratory viral infections have been implicated in both the development and exacerbation of asthma. Although repeated RSV infection and allergen sensitization can occur throughout life, the effects of respiratory infection and subsequent allergen sensitization in the development of airway hyperresponsiveness (AHR) are not well known.
Objective: To determine whether neonatal infection with RSV plays a critical role in the development of AHR and pathophysiological response to subsequent allergen sensitization in adult mice.
Methods: Mice were infected initially at 5-7 days of age with RSV and were re-infected 5 weeks later. RSV-infected mice were exposed to ovalbumin (OVA) at 1 or 3 weeks after infection via airway followed by assessment of airway function by invasive method in changes of lung resistance to inhaled methacholine (Mch), cytokine levels in bronchoalveolar lavage fluid (BALF), and lung histopathology.
Results: RSV infection and subsequent allergen sensitization resulted in development of AHR and inflammation, which is associated with increased eosinophils influx into the lungs, increased IL-5, and mucus production. Although enhanced AHR to Mch and inflammations are observed in both RSV-infected mice at neonate and subsequently re-infected with RSV after weanling, these response were more exaggerated in mice with early exposed to ova than later after RSV infection.
Conclusion: Neonatal RSV infections predispose the adult to develop enhanced airway responses and inflammation upon allergen exposure.

LATE BREAKING ABSTRACTS-CLINICAL

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Food allergy in Chinese preschool children: epidemiology and disease spectrum

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Background: Food allergy is an early and potentially life-threatening manifestation of 'atopy march'. There is limited published epidemiological data of food allergy in Chinese children. To plan for possible intervention to prevent the development of food allergy, it is important to identify its disease spectrum and risk factors in our population. This study aims to determine the prevalence and clinical features for self-reported food allergy in Hong Kong preschool children.
Methods: Children aged 2-6 years living in Hong Kong were recruited through randomly selected nurseries and kindergartens for this study to ascertain the presence of self-reported symptoms of food allergy and other atopic disorders. The parental questionnaire was developed from the Chinese version of International Study of Asthma and Allergies in Childhood questionnaire, with addition of validated items on details of food allergy.
Results: 3827 children from 21 local nurseries and kindergartens returned our study questionnaires, of which 3677 (96.1%) children were evaluable for food allergy. Two hundred and fifty-three (6.9%) children were born in mainland China and subsequently migrated to Hong Kong. Four hundred and twenty-two (11.5%) children were currently avoiding any food. The prevalence rates of self-reported food allergy and self-reported, doctor-diagnosed food allergy were 8.1% and 4.6%, respectively. The figures for ever wheeze, doctor-diagnosed asthma and current flexural eczema were 14.0%, 5.0% and 31.9%. The six leading causes of self-reported food allergy in our Chinese children were shellfish (15.8%), egg (9.1%), peanut (8.1%), beef (6.4%), cow's milk (5.7%) and nuts (5.0%). When compared with children born and raised in Hong Kong, children born in mainland China had significantly lower prevalence of self-reported food allergy (4.0% versus 6.7%; $P=0.016$), doctor-diagnosed asthma (1.7% versus 5.4%; $P=0.006$) and current rhinoconjunctivitis (13.9% versus 25.4%; $P<0.001$) but not current flexural eczema ($P=0.257$). The occurrence of food allergy was strongly associated with current wheeze, rhinoconjunctivitis and flexural eczema ($P<0.001$ for all).
Conclusion: Food allergy is a significant atopic disorder in Hong Kong preschool children, and coexisting airway and skin allergies are also commonly seen in these children. Further studies to identify early life feeding and environmental factors for food allergy are needed.

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A patient with tracheobronchial amyloidosis and complete lung collapse

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Introduction: Tracheobronchial amyloidosis is a rare condition, characterised by the deposition of amyloid plaques in the upper respiratory tract.
Background: A 65 year old lady with pre-existing tracheobronchial amyloidosis presented to the emergency department with dyspnoea, cough and coryzal symptoms. On the second hospital day, she developed stridor and worsening hypoxia. Clinically there was no air entry in the right lung, and chest radiography confirmed collapse. The patient was intubated and ventilated and underwent emergency bronchoscopy. This demonstrated mucus plugging and blood in the right main bronchus, which was aspirated and washed out. The underlying mucosa was grossly oedematous and friable. After bronchoscopy the patient required ventilatory support but air entry in the right lung had increased significantly, and re-expansion was confirmed by chest radiography. This was accompanied by a marked improvement in oxygenation, and the patient was extubated 3 days later.
Discussion: Amyloidosis can be defined as a diverse group of disorders that share a common aetiology–the extracellular deposition of protein in a pathological fibrillar, insoluble form. Localised AL amyloidosis has a predeliction for the respiratory, gastrointestinal and genitourinary tracts, and is frequently nodular in character. Systemic AL amyloidosis is associated with underlying monoclonal plasma cell dyscrasia, and features widespread organ involvement. Gilmore et al outlined the main respiratory amyloid syndromes: parenchymal (subclassified into nodular and diffuse alveolar septal) laryngeal intrathoracic lymphadenopathy tracheobronchial. Tracheobronchial amyloidosis refers to the deposition of amyloid plaques in the upper respiratory tract, and is relatively rare. It occurs most frequently in a diffusely infiltrative form, although a nodular form also exists. Reduction in airway calibre is the main feature, with consequent atelectasis and recurrent pneumonia. Successful treatment of endobronchial lesions using Nd-YAG laser irradiation has been reported. External beam radiation is a relatively recent technique, and results in improvement in spirometry, symptoms and bronchoscopic appearance. Other treatment modalities include bronchoscopic resection, surgical resection, carbon dioxide laser ablation and stenting.

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Detection of specific anti-Alternaria alternata IgE in asthmatic patients

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Background: Alternaria alternata (A. alternata) is well-known as source of allergenic components in the cell wall and cytoplasm of conidia and hyphae that cause allergic asthma.
Purpose: Purpose of this study was to purify the A. alternata antigens to evaluate the skin allergic reactions and specific anti-A. alternata IgE in asthmatic patients.
Methods: Eighty-eight patients with asthma (46 male, 42 female) and 40 non-atopic cases were enrolled with sequential trials. Alternaria alternata antigens were prepared and then skin prick test (SPT) and dot-blotting assay were performed for all patients.
Results: Positive SPT reactions were obtained on 70.5% of patients ($P < 0.05$). The males had more positive SPT reactions than female ($P < 0.05$). Using dot-blotting test, specific anti-A. alternata IgE antibody was detected in 46 (52.3%) of patients and had strong positive responses to A. alternata antigens, but no specific IgE antibody was detected in control group ($P < 0.05$). However, 16 (18.2%) sera of patients showed weak positive IgE-dot blotting results, in respect to sex and age, no significant differences were observed.
Conclusion: The results confirmed that different allergenic components of A. alternata may play an important role in producing signs of asthma in the sensitive patients.

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Nonatopic asthma have more hazardous effects on the course of chronic rhinosinusitis compared with atopic asthma

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Background: We wanted to investigate the clinical characteristics of CRS patients with atopic and nonatopic asthma.
Methods: Twenty-one atopic and 25 nonatopic asthma with CRS and 30 nonasthmatic CRS patients were enrolled in this study. The severity of asthma was evaluated and clinical parameters including the degree of nasal polyposis and disease extension, pre- and postoperative subjective symptoms, and surgical outcome evaluated with endoscopy.
Results: The severity of asthma was about the same between the two asthma groups. While all groups showed improved rhinosinusitis symptoms, significantly more severe disease extension and poorer endoscopic surgical outcome was noticed in the nonatopic asthma group. No differences were found in other parameters.
Conclusion: In this study, nonatopic asthma is supposed to make more hazardous effects on CRS disease extension and surgical outcome in patients with both asthma and CRS.
Clinical implications: CRS patients with nonatopic patients have more possibilities of severe disease extension and poor surgical outcome, which should be kept in mind when interviewing the CRS patients.

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The changes of allergic sensitization with age in children with allergic rhinitis

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Objectives: Allergic disease in children has diverse characteristics & sensitization with age because of allergen, pathophysiologic and immunologic differences. There has been few reports regarding the changes of allergic sensitization with age in children with of allergic rhinitis in Korea, so we aimed to find out these changes by CAP test.
Methods: A total of 464 children with allergic rhinitis were subjected to this study. They were classified by age groups into less than 3, 4~6, 7~9, 10~12, 13~15 and 16~18 year-old. All children underwent the CAP test(Fluroenzyme immunoassay) about two subtype of house dust mite, cat, dog, and grass, tree, and weed mixture allergen. Also we analyzed the total IgE.
Results: Children with 10~12 ages had significantly higher specific IgE to house dust mite (D. pteronyssinus = 31.50 U/ml, D. farinae = 53.64 U/ml) and reactivity to multiple allergens than other age groups. There were positive correlation between age and ratio of sensitization to weed and tree mixture allergen but no correlations were not found in cat, dog, grass and mold allergen. Also Total IgE was significant difference between sex (Male: 451.05 U/ml, Female: 252.46 U/ml) but no significant changes with age.
Conclusion: The sensitization in children with allergic rhinitis was changed with age in some allergens by CAP test.

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Etoricoxib role in hypersensitivity reactions to NSAIDs, a tolerability evaluation

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Background: Adverse reactions to nonsteroidal anti-inflammatory drugs (NSAIDs) are a frequent reported problem due to the fact that these molecules are often used for pain and phlogosis control. Coxibs represent a new NSAIDs class with a specific trigger action, involving the inhibition of the isoform 2 of the cyclo-oxigenase (COX2) enzyme, without inhibiting the COX1 activity and the endothelial and epithelial prostaglandin synthesis. These drugs,

however, if taken for a very long time, can have cardiac side effects. Etoricoxib is less cardio-toxic.

Methods: We have tested the drug on 65 patients with previous adverse reactions to NSAIDs: 13 to salycilates, 18 to arylpropionics, 10 to arylacetic acid, 12 to oxicam and derivates, 8 to pyrazolones and 4 to acetaminophen (paracetamol-P). The reported symptomatology was urticaria and/or angioedema in 69%, rhinitis reactions in 3% and one case of anaphylactic shock (1,5%). Peroral challenges were performed at random controlled with placebo, starting with a 10 mg dosage up to a total dosage of 90 mg. in the third day. The challenge has been done in Day Hospital and the patients were cardio-respiratory monitored for 4-6 hours.

Results: The tolerance test was fully carried out and well tolerated in 97% of the patients. Only two systemic reactions and a slight tiredness along with stomach-ache were reported during the first day.

Conclusion: Etoricoxib can be considered a safe molecule for those patients with previous adverse reactions to NSAIDs.

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Positive prevention by people living with HIV among injecting drug users, mashhad positive club: A joint project with UNAIDS Iran

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Background: Mashhad locates in North-East of Iran and so close to the borders of Afghanistan and Turkmenistan, also considered as a migrated city due to religious reasons. Injecting Drug Users are increasing due to its lower cost and other interests especially in marginalized regions. Sharing the contaminated syringes and injecting tools is the most important and common way of HIV transmission in the country and city. Peer education with PLHIVs among their peers and most at risk groups such as IDUs is a strategy of positive prevention which also helps as psychosocial support for PLHIVs. Mashhad Positive Club established to empower PLHIVs based on the national AIDS program and positive prevention through UNAIDS, MOH CDC and Family Planning Association support.

Method: After preparing PE Protocol and call for action, we trained 7 PLHAs including 3 of them were IDU. They were going to the ruins, places for group injection (blood sharing) and some in Drop In Centers in Daravey and Nodeh, two marginalized regions of Mashhad. They have trained their peers, IDUs and other positives, by face to face talking and distributing harm reduction packages including syringes and condoms for safer sex came out through FGD. The process monitoring was being done by writing a daily and weekly report, going with another peer educator and officer direct visit. The other training program is peer group sessions including movie display titled "AIDS Borne" then a discussion between trained educators and beneficiaries.

Results and Conclusion: Based on one month evaluation (1-31 July 2007) 455 IDUs (431 Male and 24 Female),%88 of them were 20-30 years old, have been trained by PLHAs and referring to DIC to exchange syringe program and getting condoms or start MMT increased about 42 percent. An indirect finding is health care workers in DIC have changed their attitude to PLHAs and see them not as a problem also as key of problem solving which would be decreasing social stigma.

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Gene polymorphisms of tumor necrosis factor α-308 and interleukin 10-1082 among asthmatic Egyptian children

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Background: Tumor necrosis factor (TNF) α–308 and interleukin (IL) 10-1082 have potent in inflammatory response that may including bronchial asthma.

Objective: Of this study was to check for association of polymorphisms related to cytokine genes with susceptibility and severity of bronchial asthma in Egyptian children.

Methods: Blood samples of 69 asthmatic children receiving treatment and follow up at Allergy and Respiratory Medicine Unit, Mansoura University Children Hospital, Egypt were subjected to DNA extraction and amplification using PCR with sequence- specific primers for detection of single nucleotide polymorphisms in the promoter regions of cytokine genes TNF-α-308(G→A), IL-10-1082 (G→A).

Results: Compared to normal controls Egyptian asthmatic children showed significant higher frequency of IL-10-1082 G/G homozygosity genotype ($P<0.0001$, OR=7) with lower frequency of G/A heterozygosity genotype among cases. This finding was also detected in cases with persistent asthma and eczema. Whereas these cases showed significant lower frequency of TNF-α-308 G/A heterozygosity ($P<0.05$, OR=0.44). Male cases and cases with positive family history and persistent type of asthma showed higher frequency of G/G homozygosity.

Conclusion: These specific Egyptian cytokine gene polymorphisms may contribute to asthma susceptibility and severity of asthma among children. Separate studies should be specified relating these cytokine genotypes to response to various modalities in asthma therapy.

Key words: Gene Polymorphisms, TNF α-308 and IL 10-1082, Asthma, Egypt, Children.

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A two-year aerobiological study and immunotherapy follow-up with Phoenix sylvestris pollen to seasonal allergic patients from West Bengal, India

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Phoenix sylvestris or date sugar palm commonly occur in India in both wild and cultivated forms. The tree is economically important as it yields fruit, sugar, alcoholic drinks etc. But this pollen was reported to be allergenic also. So after aerobiological studies we tried to do immunotherapy to the susceptible patients. A 2-year aerobiological suuvey in the northern suburb of Greater Calcutta was carried out with the help of a Burkard Volumetric Sampler. SPT was carried out with P. sylvestris pollen extract on adult respiratory allergic patients. 35 subjects with typical seasonal allergic rhinitis with/without bronchial asthma were selected. Symptom-medication score (based on questionnaire and patient's diary) was correlated with pollen count as recorded in Burkard sampler. 18 subjects were recruited in immunotherapy (IT) group receiving regular injections containing standardized allergen extract and 17 were recruited to placebo-controlled (PC) group. Changes in the level of specific IgE, IgG1 and IgG4 were recorded at three months intervals. Measurement of skin wheal diameter, total IgE level and FEV1 were performed prior starting and after one month of finishing therapy schedule. The pollen grains of P. sylvestris were found to be airborne from January to March. Although the occurrence of the pollen in the air was seasonal, during the peak month, i.e., in February, it contributed 15.97% for the first year and 14.84% in the second year of the total aeropollen load. 44.07% of the total patients showed positive reaction in SPT. The IT group showed 33.5% and 57% decrease in symptom-medication score during the first and second treatment season respectively. This group showed a significant decrease of skin-reactivity to PS pollen extract & specific IgE, significant increase in FEV1, specific IgG1 (1.95–3.2 times) & IgG4 (21.24–30.83 times) and no significant changes in total IgE level. In case of PC group, no significant changes were recorded for all the mentioned parameters excluding the development of new sensitization in two cases against Saccharum officinarum (Poaceae) pollen grain and Alternaria spore respectively. Phoenix sylvestris pollen are airborne and they cause respiratory allergy to the susceptible patients. Allergen immunotherapy with standardized PS pollen extract is effective in seasonal respiratory allergic subjects susceptible to PS pollen with narrow range of sensitization.

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Fungal spore, a potential source of occupational health hazard among the workers of potato cold stores in west Bengal, India

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Intense occupational exposure to a number of mould spores or their fragments for brief or prolonged periods may be responsible for serious health hazards. This had been the case with the workers of the Potato Cold Stores in West Bengal. So the basic purpose of the present investigation was first to assess the qualitative and quantitative estimation of the aeromycoflora of the cold stores, to determine their allergenic potentialities to incite respiratory diseases among the workers and also deterioration of the food grains. Burkard personal Sampler and Andersen two stage sampler was used for this investigation. Diagnosis of respiratory allergic disorders was based on detailed medical history followed by skin-Prick test (SPT). A total of 20 fungal spore types were recorded during the survey period of two years (July'2005 to June'2007) with the dominance of Aspergillus niger (21.73%), Curvularia lunata (19.13%), Echinobotryum (15.96%), Alternaria alternata (13.06%), Fusarium solani (5.78%), and Rhizopus nigricans (2.45%) etc.,from the store houses. Clinical investigation of the susceptible workers with the antigenic extracts of the six dominant spore types using Skin Prick Test method clearly demonstrated their variable allergic potency. Among the 126 patients tested the highest positive reaction (2+ or more intensity) was noted in Aspergillus niger (26.98%) followed by Rhizopus nigricans (21.42%), Alternaria alternata (19.84%), Curvularia lunata (19.04%), Fusarium solani (18.25%), etc. Allergenic symptoms were pronounced during monsoon and summer season or whenever, there was prolonged disruption of power supply. High frequency of positive response to SPT was due to an increased prevalence of allergic fungal forms in the working environment, which can be attributed to poor post-harvest storage and scientific management of the store houses. The results obtained showed that the work environment was considerably polluted with the spores of fungi of allergenic and immunotoxic properties, which cause a high degree of health risk to people employed in that area.

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Determination of total and specific IgE response in allergic rhinitis patients of Kashmir Valley-India

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Allergy accounts for a substantial number of human diseases with significant morbidity to patients. Kashmir valley has been witnessing an increase in allergy related disorders usually due to aeroallergens present in the environment mostly during spring and autumn seasons. The present study was aimed at finding total and specific IgE responses in serum sample of 250 patients, reporting to various health centers across the valley with symptoms of seasonal allergy. Samples were first screened for the total IgE levels by a sandwich ELISA method. All the samples showed presence of high levels of total IgE (650–1200 iu/ml). Specific IgE levels were determined using grass and tree mix antigens by a two step capture ELISA method. After recording the concentration of total and specific IgE levels of 250 serum samples, 97 patients having severe symptoms of allergic rhinitis complaints were tested clinically by intra-dermal antigen skin test method for different allergens like pollen (18 types), insect (1 type), dust (5 types), fungi (5 types) and epithelia (2 types). 187 samples (74.80 %) out of 250 were reactive for the allergens used in the present study. Specific IgE ranged from 1.1–75.0 iu/ml representing moderate to high specific IgE levels. 63 samples not reactive to the allergens used above but had high total IgE levels were probably due to other allergens which are further being evaluated. The skin test reactions were interpreted and graded at 15 – 20 minutes as per the criteria of Shivpuri (1974).

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A study on association of pet ownership history and risk of asthma among Iranian children

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Background: The identification, isolation, and elimination of allergen(s) causing bronchial asthma are the most efficient aspect of treatment. The pet industry has diversified recently, increasing the risk of pet owners' exposure to many unknown antigens.

Results: Of population studies have been contradictory and some epidemiological studies showed the risk of pet keeping, some even suggesting that keeping pets decreased the risk of sensitization and asthma.

Purpose: This study was to determine the association between pet ownership and asthma.

Methods: A case-control study was conducted among 215 asthmatic participants referred to Children Medical Center in Tehran in a 2 year period and were asked to reply a questionnaire in concerning to kind of pet, its sex and puberty, the place of keeping it, duration of keeping and the aim of pet keeping. Cases were recruited and matched (age and sex) with 215 healthy controls. Statistical analysis performed to calculate Odds Ratio (OR) of asthma morbidity in individuals who had kept pets.

Results: Odds ratio of asthma morbidity in patients who had kept pets was 2.59, CI=1.60–4.21 and P>0.001. Financial aim was the most reason of pet keeping (18.1%) and the most of pets were mature and were kept outdoor (41.7%). Pet keeping duration was less than 6 months (87.6%). No difference between genders was observed.

Conclusion: This study provides evidence that pet ownership is an important risk factor for asthma. We suggest that individuals who are at the risk of asthma (Atopic individuals) must avoid contacting with pets. However more research in this field in Iran is necessary.

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Health hazards in potato cold stores. Manas Ranjan Majumdar

Manas Ranjan Majumdar. *HALDIA GOVERNMENT COLLEGE, BIOLOGY, HALDIA, India.*

Occupational exposure to a number of mould spores and fragments for a brief or prolonged periods may be responsible for health hazards. Such has been the csae with the workers of potato cold stores in West Bengal, India. The investigation was carried out to assess the qualitative and quantitative composition of air aeromycoflora of the store houses and then to determine their allergenic potentialities to incite respiratory or other allergic disease amonfg the workers. A total of 22 fungal types were recorded during the investigation period for years (from August 2004 to July 2006)with the dominance of *Aspergillus niger* (21.73%), *Curvularia lunata*, (19.13%)*Echinobotrynum* (15.96%), *Alternaris alternata* (21.73%), *Fusariun solani* (5.78%). and *Rhizopus nigricans* (2.45%). Cnincal investigation of the susceptibel workers with the antigeniic extracts of the six dominat spore types using Skin Prick test method clearly demonstrated their variable allergic potency. Among the 100 patients tested a high positive reaction (2+ or more intensity) was noted in *Aspergillus niger* (26.98%) followed by *Rhizopus nigricans* (2.45%), *Alternaria alternata (19.84)*, *Curvularis lunatat*, *Fusarium solani (18.25%)*, etc. Allergenic symptoms were pronunced during monosoon ans summer season or wheneever there were prolonged disruption of power supply. High frequency of positive response to SPT was due to an increased prevalence of allergic fungal forms in the working environment which can be attributed to poor post-harvest storage and scientific management of the store houses.

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Data inconsistencies in abstracts of research articles of three allergy journals

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Background: Recent studies have shown data inconsistencies between the abstract and full text of published papers.

Objective: The study was to determine data inconsistencies between the abstract and full text of articles published in three leading allergy journals.

Methods: All articles published in Clinical and Experimental Allergy, Journal of Allergy and Clinical Immunology and Allergy consecutively over six months from January 2005 that contained data in the abstract were checked against corresponding data in the body of the article.

Results: Of 96 articles in Clinical and Experimental Allergy, 33 contained data in the abstract inconsistent with those reported in, or absent from the body of the articles, giving an abstract data inconsistency rate of 35.4% (95% CI: 25.8–45.0). Corresponding abstract data inconsistency rates for the Journal of Allergy and Clinical Immunology and Allergy were 29.9% (95% CI: 20.3–39.5; 87 articles) and 24.3% (95% CI: 16.3–32.3; 111 articles) respectively.

Conclusion: About one-quarter to one-third of research articles in three leading allergy journals contained data in abstracts that were inconsistent with corresponding data in the article, or were not in the article.

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Prevention of signs and symptoms of cold urticaria by ebastine

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Cold urticaria is a frequent subtype of physical urticaria that is caused by the release of proinflammatory mast cell mediators after cold exposure, resulting in wheals and itching and sometimes in general systemic

complications. Cold urticaria may be potentially life threatening, in any case it has a profound impact on patient quality-of-life. Second-generation antihistamines are recommended as the first-line treatment, but to date only a few have been scientifically tested for this condition. To assess the safety and efficacy of ebastine in preventing ACU symptoms, we examined 22 adult patients suffering from cold urticaria within the scope of a double-blind cross-over trial administering a single dose of 20 mg ebastine or placebo before local cold challenge. The safety of ebastine was sensitively assessed with a psychometric battery testing cognitive performance and mood. After cold challenge, wheal and erythema were assessed by the investigator and the intensities of pruritus and burning were rated by the subject. We could show that Ebastine had no negative impact on any of the parameters of cognitive performance or mood. Most importantly, cold urticaria symptoms were markedly reduced after challenge in the verum group, indicating that ebastine can effectively protect patients from cold urticaria symptoms. These results demonstrate the safety of ebastine and reveal its efficacy in treating patients with cold urticaria.

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Dermographic urticaria symptoms can be safely prevented by ebastine

Markus Magerl, Jan Schmolke, Frank Siebenhaar, Torsten Zuberbier, Martin Metz, and Marcus Maurer. *Charité - Universitätsmedizin Berlin, Dermatology, Berlin, Germany.*

Physical urticaria includes a heterogeneous group of disorders characterized by the development of urticarial lesions and/or angioedema after exposure to certain physical stimuli. The most frequent physical urticaria is the dermographic urticaria (DU), in which shearing forces on the skin result in wheals and itching. Subsequent scratching aggravates the symptoms. Second-generation antihistamines are recommended as the first-line treatment, but to date only one has ever been tested for this condition. The aim of this study was to assess the safety and efficacy of ebastine in preventing DU symptoms. We administered 20 mg ebastine to seven adult DU patients in the scope of a double-blind cross-over trial. The safety of ebastine was sensitively assessed with a psychometric battery testing cognitive performance and mood. After challenge by shearing forces, wheal and erythema were assessed by the investigator and the intensities of pruritus and burning were rated by the subject. We could show that Ebastine had no negative impact on any of the parameters of cognitive performance or mood. Wheals, pruritus, and burning were greatly reduced for the majority of subjects. These results underline the safety of ebastine and reveal its efficacy in cold urticaria. and should be tested on a larger scale.

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Sensitization to environmental allergens among the inhabitants of Calcutta city and suburbs with special emphasis to inhalant fungal allergy: an aerobiological and allergological study

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Background: Exposure to inhalant bio-particles including mold spore, pollen, dander, dust mite may incite allergic reaction in pre-sensitized individuals. Present study was designed to find out the prevalence of allergic diseases among Calcutta population and to explore the role of mold spore as inhalant allergen.

Methods: A Burkard sampler, installed in an agricultural field in suburban Calcutta, operated from Nov 2002-Oct 2004 for catching the mold spores and a mold calendar was constructed.

Subjects: (n = 1251, male-47.48%; female-52.52%, mean age = 37.97iÃ13.13 year) were selected from individuals attending the Allergy unit of the Institute of Child Health, Calcutta. A questionnaire was used to assess symptoms and personal factors (current smoking, pet keeping, family history of asthma, and atopy). A 17 point scale (0 to 17) was constructed for calculating symptom score. Skin prick test was performed with sterile antigenic extract and the reaction was considered positive if the wheal diameter was î? 3 mm. The values of lung function variables like FVC, FEV1, FEV1% FVC and FEF 25–75% were recorded by RMS Medspiror spirometer. Relationship between symptom score and various personal, and environmental factors was explored by linear regression at 0.05 probability level (p).

Results: A total of 37 mold spore types were identified and counted. The total air-spora (cumulative concentration varies from 824.69–3921.72 spore/m3) attained a peak in December.About 67.80% of study population reported symptoms of respiratory allergy (allergic rhinitis, bronchial asthma or both). Subjects showed variable skin sensitization to different allergens: 46.03% to molds, 93.65% to pollens, 65.08% to dust mite, 49.21% to cotton fibre and 3.17% to animal dander. About 40.28% subjects exhibited airflow limitation in peripheral bronchi whereas 4.88% showed airflow limitation in both central and peripheral bronchi. Symptom score was best predicted by family history of atopy (adjusted beta 1.34 at 95%CI) at $p<0.05$ whereas sensitivity to Azadirachta pollen was a significant risk factor for mold sensitization (adjusted beta 0.27 at 95% CI) at $p< 0.01$. The predictors for FEV1%FVC included female sex, age, sensitization to mold, and animal dander ($p<0.05$).

Conclusion: Prevalence of respiratory allergy was found in Calcutta population. Family history of atopy, female sex, age, sensitization to mold, and animal dander were risk factors for it.

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Eosinophil cationic protein in sputum of wheezing infants

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Background: Considering the role of eosinophilic inflammation in asthma it has been suggested that eosinophils or its granule proteins may be applicable in the diagnosis of asthma.

Purpose: The study was to assess the level of eosinophilic inflammation of the lower airways, eosinophils and eosinophil cationic protein (ECP) by sampling induced sputum from the pharyngeal region of their oral cavities.

Methods: Fifty four infants with a mean age 9 months (2–22 months), who had been hospitalized in the Pediatric Department of the Alexandrovska Hospital with acute wheezing episodes from October 2005 to March 2006 year, were included in the study. The wheezy children were divided into three clinical groups according to the number of previous wheezy episodes: group 1-first wheeze (n = 29), group 2-with recurrent wheeze (n = 15) and group 3-those with persistent episodes of wheeze diagnosed clinically as asthma (n = 10).

Results: Neither sputum eosinophils nor ECP turned out to be of discriminative value for the three groups of children outlined on the basis of clinical criteria. Still the median sputum ECP levels in the asthmatic group tended to be higher (966 ìg/L) than those in the recurrent wheezy group (575 ìg/L) and in the first wheezy group (123 ìg/L), with a considerable overlap between the groups.

Conclusion: We documented very early presence of eosinophilic inflammation in infants with recurrent respiratory symptoms and suspected asthma.

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Defining asthma phenotypes based on seasonal distribution of exacerbation in children with bronchial asthma

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Background: Nowadays asthma is not considered as a homogenous disorder. Several asthmatic phenotypes could be distinguished, especially in childhood. No standardized method for determination of these phenotypes is available.

Aim: To define asthma phenotypes in asthmatic children based on the seasonal distribution and some clinical features of asthma exacerbations.
Methods: Object of research were 326 children with 1407 acute episodes treated at the Clinic of Pediatrics in the period between 2004 and 2006. Based on distribution of the exacerbation frequency throughout the year with two well-marked peaks in December-January and in June-July three phenotypes were defined: "winter" phenotype (WP) /presumptive virus-induced obstruction/ in 145 children, "summer" /atopic/ phenotype (SP) in 82 children, and "non-seasonal" /persistent/ phenotype (NP) in 99 children.
Results: Children with WP showed significantly lower average age (6.8 years for WP versus 11.3 years for SP), dominating symptoms for respiratory infection (88 % for WP versus 31 % for SP), and lower frequency of atopy (in 32 % for WP versus 78 % for SP). The asthma onset in SP group of children was later (average age of 6.5 years) while in WP the onset was at average age of 4.1 years. WP and NP showed slower improvement of the obstructive syndrome and more manifested functional impairments after the treatment (FVC 61 % of the expected for WP and FCV 67 % of the expected for NP).
Conclusion: Based on seasonal distribution of asthma exacerbations three asthma phenotypes with different clinical features, course and treatment results were defined.

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Risk factors leading to hospital admission in Iranian asthmatic children

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Background: Asthma is one of the most common chronic diseases in the world, leading to an increased rate of hospitalization. We performed this study to better understand the factors leading to admission among asthmatic children.
Methods: We performed a study among asthmatic children in a referral hospital for asthma and allergy in Tehran. Sixtythree cases were selected from asthmatic children admitted to the emergency room (ER) who still had an indication for ward or intensive care unit admission after primary treatment. Our control group was the asthmatic children discharged after primary treatment and patients who were referred to the asthma and allergy clinic (63 patients). Data were obtained by structured questionnaires filled out during clinical interviews.
Results: There was a significant difference in mean age (5 years for cases vs. 6years for controls; $p = 0.049$), personal and familial allergic history (69.8 and 57.1% for cases vs. 34.9 and 36.5% for controls; $p < 0.01$ and $p = 0.02$, respectively), history of recent respiratory infections (79.4% for cases vs. 49.2% for controls; $p < 0.01$), hospitalization history due to asthma (57.1% for cases vs. 23.8% for controls; $p < 0.01$) and regular use of inhaled corticosteroid (66.7% for cases vs. 33.3% for controls; $p < 0.01$).
Conclusion: Our findings confirm most previous observations, suggesting that recent respiratory infections, hospitalization, personal or familial allergy, disease severity and lower ages are important factors leading to hospitalization. We also found that regular clinical follow-up, regular use of inhaled corticosteroids, higher IgE levels and O 2 saturation may lower the probability of hospitalization during asthmatic attacks.

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11Polymorphisms impairing histamine degradation moderate behavioural responses to food additive challenge

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Background: The relationship between artificial food colouring and benzoate perservative (AFCP) intake and behaviour has hitherto been contentious. We have previously shown in a population based DBPCFC study, an adverse effect of AFCP on parentally-rated behaviour of 3 year olds and have now confirmed the observations in 144 8–9 year olds and 153 3 year olds using objective measures of observed behaviours in a school setting. One potential mechanism is IgE independent histamine release from circulating basophils.
Methods: From the second challenge study we have genotyped the children using buccal cells and related findings to the magnitude of the behavioural response to challenge.
Results: Polymorphisms of catecholamine genes, COMT Val108Met and ADRA2A C1291G, previously associated with ADHD, had no impact on responses but T939C and Thr105Ile polymorphisms of the histamine N-methyltransferase gene (HNMT) significantly ($p=0.02$ and 0.04 respectively) adversely affected responses to AFCP challenge.
Conclusion: HNMT polymorphisms impair histamine clearance and AFCP cause histamine release. The presence of Histamine 3 receptors in the brain provides a potential mechanism (and therapeutic target) to explain the effect we observed. Many environmental factors increase histamine including infections and many foods. This would explain the frequent claim that food intolerance and infections adversely affect behaviour in some children. This gene by environment interaction should be investigated in relation to AFCP induced urticaria, asthma and other atopic conditions.

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Serum specific IgE - is result enough?

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Background: In vitro determination of serum specific IgE (sIgE) is an essential diagnostic tool in allergy. Clinical analysis laboratories offer different methods / technologies to determine sIgE, in some cases with highly variable and poorly interchangeable results, concerning the different allergens. Rotation between different methods may compromise allergic patients follow-up, namely in those situations that may benefit from sequential sIgE determinations, as food allergy and specific immunotherapy efficacy evaluation.
Aim: To verify the availability of written information concerning the method used in sIgE determination in lab result reports from clinical analysis laboratories in Lisbon District.
Methods: During 2 months (April and May 2007), 3 Immunoallergologists collected serum sIgE result reports from the patients they observed. Reports from 50 different clinical analysis laboratories within Lisbon district were obtained and analysed. Labs that did not mention the method that had been used to sIgE determination were further inquired by phone, in order to get that information.
Results: Among the 50 reports analysed, only 13/50 (26%) specified the method that had been used, namely the immunoenzimatic (IE) method in 9 and the chemiluminescent (QL) method in 4. Among the remaining 37 labs, when later contacted by phone, we verified that 17/37 (46%) were using IE method, 14/37 (38%) QL and 6/37 (16%) were using both methods, according to the different allergens. In 27/50 laboratories (54%), "RAST" was used as synonym of sIgE.
Conclusion: We intend to alert to the generalized lack of written information concerning the methods used in serum sIgE determinations in the lab reports from Lisbon district laboratories. This lack of written information, the persistence of the incorrect use of the term "RAST" and the simultaneous use

of both methods for different allergens reveal the unawareness of most laboratories concerning this important issue, which is urgent to amend.

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Chronic urticaria - why are patients dissatisfied with treatment?

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Background: Chronic urticaria (CU) is a common skin disorder characterized by recurrent spontaneous outbreaks of itchy wheals and/or angioedema. Chronic urticaria has been shown to have substantial impact on patient quality of life, but little else is known about patient perspectives on CU and its treatment. In particular, it is not known whether patients actually follow guideline treatment, and if not, why not.

Methods: To address these questions, a survey was conducted via an online panel with 321 randomly selected, representative adults in Germany and France who were diagnosed with CU. The survey included the Skindex-29 questionnaire on quality of life and questions about treatment and patients' relation to their physician. Treatment was compared with recommendations from the current GA^{2}LEN/EAACI/EDF guidelines.

Results: The survey confirmed that CU has substantial impact on patient quality of life. Notably, half the respondents were only "somewhat satisfied" with their treatment of their CU and considered their physician only "somewhat knowledgeable." Only 64% of the respondents were using prescription medications as recommended by guidelines. Notably, 62% of respondents reported that they avoid seeing their physician unless absolutely necessary.

Conclusion: This survey implies that physicians either do not fully understand the experience of many of their patients with CU or are not building trustworthy supporting relations with them. Physicians need to talk with these patients more about how the condition impacts their lives and feelings in order to improve their compliance with treatment guidelines.

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Lidocaine/prilocaine-treated skin modulate allergic inflammation induced by skin prick test

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Background: There are evidences that peripheral nerves contribute to the pathophysiology of many dermatologic diseases. The effects of neurogenic inflammation in allergen-induced skin prick test reaction are not clear and no study has been measured the effect of inhibition of peripheral nerves on all inflammatory skin signals induced by skin prick tests.

Objective: The purpose of this study was to investigate the anti-inflammatory effects of lidocaine/prilocaine cream on histamine and allergen-induced skin inflammation.

Methods: Skin prick tests using histamine and allergens were performed in 48 subjects on lidocaine/prilocaine and vehicle-treated skin. Volume, temperature, erythema size and pruritus on skin test area have been determined and compared.

Results: The absence of functional cutaneous nerves has significant and similar effects on all qualities assessed (rubor, calor, tumour, pruritus) in both allergic and histamine-induced inflammation skin responses. Effects on hyperthermia were found to be mild, i.e. similar to those on wheal formation. Pruritus responses were unaltered or even enhanced in denervated skin.

Conclusion: Lidocaine/prilocaine-treated skin modulate allergic inflammation induced by skin prick tests.

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A differential effect of two probiotics in the prevention of eczema and atopy

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Background: The role of probiotics in the prevention of allergic disease has not been clearly established, despite early reports that *Lactobacillus GG* halves the risk of atopic eczema at 2 years. This study aimed to determine whether probiotic supplementation in early life can prevent the development of eczema and atopy at 2 years.

Methods: Randomized double blind placebo controlled trial of infants at risk of allergic disease and their mothers. From 35 weeks gestation, pregnant women were randomized to take *Lactobacillus rhamnosus*, *Bifidobacterium lactis* or placebo daily until 6 months if breastfeeding, and their infants from birth to 2 years (n = 474). The infant's period prevalence of eczema and point prevalence of atopy, using skin prick tests to common food and environmental allergens, was assessed at 2 years (n = 466).

Results: Compared to infants in the placebo group, those receiving *Lactobacillus rhamnosus* had a significantly (p=0.01) reduced risk of eczema (OR=0.47, 95% confidence intervals (CI) 0.26–0.84), but this was not the case for infants taking *Bifidobacterium lactis* (OR=0.88, 95% CI 0.53–1.49). There was no significant effect of *Lactobacillus rhamnosus* (OR=0.65, 95% CI 0.38–1.11) or *Bifidobacterium lactis* (OR=0.76, 95% CI 0.45–1.28) on atopy.

Conclusion: The protective effect we found for *Lactobacillus rhamnosus* against eczema but not atopy is consistent with findings from some other studies. The effect of probiotics on eczema may depend on the particular probiotic selected for use.

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Sublingual Immunotherapy with Honeybee venom reduces large local reactions: a randomized, double blind controlled study

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Introduction: Hymenoptera venom immunotherapy (VIT) is highly effective, since it confers a clinical protection against hymenoptera sting, including honeybee (HB) in about 90% individuals (1). The effectiveness of VIT can be easily demonstrated by the reduction of the intensity and size of large local reactions (LLR)(3, 4). VIT is currently given only by subcutaneous injections. Sublingual immunotherapy SLIT proved effective and safe in respiratory allergy, therefore its use in hymenoptera allergy can be hypothesized. We aimed at evaluating the clinical efficacy of SLIT with HB venom in beekeepers or their family members, by assessing the effect on LLR to HB stings. LLR was chosen as evaluation parameter for safety reasons, as this is a pilot study.

Methods: This was a randomised, double blind, placebo controlled study. Patients with LLR due to HB sting and skin tests/CAP-RAST assay solely positive to HB were enrolled. They were randomised to receive either HB SLIT (15 patients) or undistinguishable placebo (15 patients) for six months. The HB SLIT (Anallergo, Florence, Italy) involved a 6-week build-up, followed by a maintenance phase where 400 mcg were given monthly. LLR on HB sting were measured before SLIT and after no less than 3 months of

maintenance. Treated patients and placebo were compared by Pearson test. P values less than .05 were considered statistically significant.
Results: 30 adult patients were enrolled and 26 completed the study, with 1 dropout in the active and 3 dropouts in the placebo group. In SLIT patients the peak diameter of LLR was reduced > 50% in 8/14 (57%). No change in LLR was seen in 11 of 12 placebo patients, and a systemic reaction (generalized urticaria) occurred in a patient on re-sting. The difference was significant (P = .004) between the 2 groups. No adverse event was reported and no discontinuation of SLIT was required.
Conclusion: HB-SLIT significantly reduced the extent of LLR in HB allergic subjects, and its safety profile was satisfactory. This pilot study envisages the possible use of SLIT also in hymenoptera allergy, and trials in patients with systemic reactions should be the next step to confirm our observations.

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Mometasone furoate nasal spray effectively treats the ocular symptoms of seasonal allergic rhinitis in patients with all degrees of severity over 24 hours

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Background: Mometasone furoate nasal spray (MFNS) has been shown to be effective in controlling the nasal symptoms of seasonal allergic rhinitis (SAR). An analysis of subject-reported symptom data in MFNS trials was performed to determine the impact of MFNS on ocular symptoms. This analysis evaluated the effects of MFNS on SAR patients with varying degrees of symptom severity and differences in symptom reduction in the morning (AM) and evening (PM).
Methods: A retrospective analysis was conducted of data pooled from four randomized, double-blind, placebo-controlled clinical studies (three 2-week; one 4-week) with similar protocols that compared the effectiveness of MFNS 200 mcg QD and placebo in controlling ocular symptoms in subjects aged ≥12 years with a ≥2-year history of SAR. The analysis reviewed change from baseline over a 15-day period in symptom scores for eye tearing, itching, and redness on a 4-point scale (0=none; 3=severe), and calculated total ocular symptom score (TOSS). The effect size of the Days 1–15 average for subjects with mild, moderate, or severe symptoms at baseline was compared, and TOSS was evaluated approximately 12 hours postdose (PM) and on arising, approximately 24 hours postdose (prior to dosing on next day, AM).
Results: Subjects in the MFNS (n = 491) and placebo (n = 492) groups had similar baseline clinical and demographic characteristics. The mean baseline TOSS was 4.35 (PM 4.39; AM 4.32) for the MFNS group and 4.5 (PM 4.60; AM 4.43) for the placebo group. The decrease in TOSS with MFNS for the Day 1–15 interval was -19.8% vs -5.6% for placebo (P<0.001); the difference reached statistical significance at Day 3. For the same time period, scores for tearing decreased by -22.2% with MFNS vs -13.9% with placebo (P<0.001), for eye itching by -17.3% with MFNS vs -12.3% with placebo (P=0.002), and for redness by -15.7% with MFNS vs -8.3% with placebo (P=0.002). For the mild, moderate, and severe symptom groups, the improvements in TOSS with MFNS over placebo were 0.30, 0.42, and 0.50, respectively. Reductions in individual symptom scores were significantly greater with MFNS than with placebo in both the PM (P≤0.006) and AM (P≤0.001). Importantly, differences in TOSS between MFNS and placebo in the PM (0.36) and AM (0.43) were similar.
Conclusion: MFNS reduced ocular symptoms of SAR over 24 hours regardless of subjects' baseline symptom severity.

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Shrimp allergy: effect of vinegar soaking on allergenicity

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Background: Previous studies demonstrated the muscle protein tropomyosin as the major shrimp allergen. This allergen, Sa-II, was shown to be a 34-kDa protein - rich in aspartic and glutamic acids. Heat-stability of this allergen has been documented. However, no studies have shown yet the effect of vinegar on this allergen. This study aimed to demonstrate if vinegar soaking prior to cooking would have an effect on shrimp allergenicity, based on skin test results.
Methods: Eighteen shrimp-allergic and eighteen non-shrimp-allergic pediatric patients were subjected to skin prick test using conventionally-prepared shrimp extract. Mean wheal diameters obtained were compared to mean wheal diameters obtained using 3 experimental shrimp extracts prepared with preliminary vinegar soaking. For the adult group, twenty-six shrimp-allergic and twenty-six non-shrimp-allergic parients were included.
Results: Mean wheal diameters obtained using shrimp extract prepared with preliminary vinegar soaking were significantly smaller than mean wheal diameters obtained using conventionally prepared extract. Wheal diameters obtained for shrimp-allergic patients were significantly bigger than wheal diameters obtained fo non-shrimp-allergic individuals. Results were similar for pediatric and adult patients.
Conclusion: Results indicate that vinegar soaking prior to cooking can reduce shrimp allergenicity. With resources, further validation of results using double-blind placebo-controlled food challenges woul be ideal.

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Mometasone furoate nasal spray improves symptoms of perennial allergic rhinitis in children and adults in Peru

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Background: Perennial allergic rhinitis (PAR) is a highly prevalent disease globally and is the most common chronic disease in children. Nasal obstruction and other bothersome symptoms of PAR need to be treated year round. Previous studies have shown the intranasal corticosteroid mometasone furoate nasal spray (MFNS) to be well tolerated and clinically effective in relieving the symptoms of PAR. A study was conducted to assess effectiveness of MFNS and patient satisfaction with treatment in pediatric and adult subjects.
Methods: In this prospective, open-label, multicenter study, 33 Peruvian subjects (age 8 to 69 years) with clinically diagnosed PAR recorded nasal symptom data in a diary. Total nasal symptom score (TNSS) was evaluated at 15 and 30 days. Subjects also rated satisfaction with treatment, and the investigators evaluated adverse events.
Results: Thirty-three subjects (61% female, 39% male) received MFNS 100 or 200 mcg QD (pediatric or adult dose). Their mean TNSS was reduced from 10.7 (± 2.9 SD) at baseline to 4.1 (± 2.9 SD) at 15 days and 1.6 (± 2.2 SD) at 30 days (P<0.001 for both reductions). At days 15 and 30, 25 subjects (76%) and 31 subjects (94%), respectively, rated themselves as satisfied or very satisfied with treatment. Mild adverse events (nasal irritation, sneezing, epistaxis, and cough) occurred in 7 (21%); one subject stopped treatment due to nasal irritation.
Conclusion: Mometasone furoate nasal spray 100 or 200 mcg QD safely and effectively improved symptoms of PAR in children and adults.

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Effect of postoperative mometasone furoate nasal spray on wound healing following endoscopic sinus surgery

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Background: Chronic sinusitis and nasal polyposis are diseases of the nasal and sinus mucosa that affect approximately 15% and 4% of the population, respectively. Functional endoscopic sinus surgery (FESS) is standard surgery

for both diseases after failure of medical treatment. Approximately 20% of surgery patients experience impaired wound healing following FESS, leading to recurrences of sinusitis or complications requiring further surgery. A double-blind multicenter study found that 6 months' treatment with the intranasal corticosteroid mometasone furoate nasal spray (MFNS) 200 mcg QD after surgery for bilateral nasal polyps significantly prolonged the time to recurrence compared with placebo. This study was undertaken to investigate whether use of MFNS following FESS improves wound healing over 6 months.

Methods: In this double-blind, randomized, placebo-controlled study, subjects with a diagnosis of chronic sinusitis or nasal polyps were randomized to receive 6 months' treatment with MFNS 200 mcg BID or placebo beginning approximately 2 weeks after FESS. Endoscopic scores were assessed at 1 and 2 weeks, and at 1, 2, 4, and 6 months. The primary efficacy endpoint was the investigator's assessment of treatment outcome at 6 months. A combination score was derived from scores for inflammation, edema, and polyps. Postoperative symptom scores were recorded at 1, 2, 4, and 6 months. Adverse events (AEs) were recorded throughout the study.

Results: Of the 99 randomized subjects, 91 (46 MFNS, 45 placebo) were included in the intent-to-treat analysis. Subjects in the MFNS group received oral steroids for the first 10 days postsurgery. For 24 subjects (11 MFNS, 13 placebo) who discontinued, results from the last study visit were used. Preoperative symptom scores in the two groups were similar. At 6 months, investigators' assessment of treatment outcome and reductions in total endoscopic scores in all subjects were superior, but did not reach statistical significance, with MFNS versus placebo. Combination endoscopic scores at 6 months showed significantly improved healing with MFNS versus placebo for all subjects (median scores: 0.0 vs 3.0, P=0.02) and those with nasal polyps (median scores: 2.0 vs 4.0, P<0.03). MFNS was well tolerated.

Conclusion: Treatment with MFNS following sinus surgery improved healing, particularly in subjects with an initial diagnosis of nasal polyps, and was well tolerated.

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Mometasone furoate nasal spray reduces symptoms and improves quality of life in chinese children with allergic rhinitis

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Background: Approximately 40% of children and adolescents in China suffer from symptoms of allergic rhinitis (AR). Effective treatment of AR in children is important in minimizing or preventing behavioral problems and poor academic performance and may help prevent the development of comorbid disorders such as asthma, adenoidal hypertrophy, and otitis media with effusion. This study was undertaken to evaluate the effects of mometasone furoate nasal spray (MFNS) on clinical symptoms and quality of life (QoL) in Chinese children with AR.

Methods: A total of 273 symptomatic children aged ≥6 years and ≤12 years with documented seasonal or perennial AR were treated in an open-label study of MFNS 100 mcg QD for 4 weeks. Eligible subjects had a nasal stuffiness/congestion score ≥2 (on a scale of 0=no symptoms and 3=worst symptoms) and a total nasal symptom score (TNSS) ≥8 (total of the individual symptom scores for rhinorrhea, congestion, nasal itching, and sneezing) at the start of treatment. The primary efficacy variable was a change in the TNSS, and the coprimary variable was a change in the individual symptom scores, while the secondary efficacy variables included overall efficacy and Pediatric Rhino-conjunctivitis QoL Questionnaire (PRQLQ) scores (assessing nasal and eye symptoms, behavior, and daily life).

Results: MFNS significantly (P<0.05) reduced TNSS from baseline (mean 9.16) after 1, 2, and 4 weeks of treatment (mean scores 5.41, 3.47, and 2.10, respectively). Significant improvements from baseline were observed at each week for all individual nasal symptoms. The percentages of patients achieving a 25% improvement (indicating at least moderate symptom reduction) from baseline in TNSS at weeks 1, 2, and 4 were 74%, 95%, and 96%, respectively. Moderate-to-complete symptom relief was reported by 71% of subjects at week 1 and 99% at week 4. Significant reductions (improvements) from baseline in total and individual PRQLQ scores were seen at weeks 2 and 4. Mean behavior PRQLQ scores decreased by a statistically significant amount from 13.99 at baseline to 5.70 at week 2 and 2.71 at week 4. Mean PRQLR scores decreased from 8.30 at baseline to 3.12 at week 2 and 1.72 at week 4. Less than 2% of subjects reported adverse events.

Conclusion: MFNS effectively relieved symptoms of AR, improved QoL, and was well tolerated in Chinese children with AR.

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Mometasone furoate nasal spray improves quality of sleep and reduces symptoms of allergic rhinitis in chinese patients

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Background: Between 20% and 40% of urban-dwelling adolescents and adults in China report symptoms of allergic rhinitis (AR). Unlike other regions where pollen allergy is common, in China dust mites and/or silk are the more prevalent allergens. Consequently, many Chinese people with AR suffer from symptoms throughout the year. One recent study found that once-daily mometasone furoate nasal spray (MFNS) was an effective treatment for AR in Chinese people in Hong Kong. An open-label study was undertaken to evaluate further the effect of MFNS on clinical symptoms and quality of life in Chinese patients with AR.

Methods: A total of 500 subjects aged ≥12 years to ≤70 years with a physician diagnosis and documented medical history of seasonal or perennial AR were treated with MFNS 100 mcg QD for 28 days at 10 centers in China. At the screening visit, eligible subjects had a total nasal symptom score (TNSS) ≥8 (maximum of 12) and a nasal stuffiness/congestion score ≥2 (maximum of 3). The primary efficacy variables were change from baseline in TNSS and individual symptom severity scores for congestion, rhinorrhea, sneezing, and itching. Secondary efficacy variables were change from baseline in scores for interference with sleep and daytime activities on the Nocturnal Rhinoconjunctivitis QoL Questionnaire (NRQLQ) and the Mini-Rhinoconjunctivitis QoL Questionnaire (Mini-RQLQ), respectively.

Results: MFNS significantly (P<0.05) reduced TNSS and individual nasal symptom scores from baseline at each evaluation. Mean TNSS at baseline and weeks 1, 2, and 4 were 9.24, 5.78, 3.99, and 2.98, respectively. Corresponding mean scores for nasal congestion were 2.37, 1.57, 1.14, and 0.84; similar improvement was seen with the other nasal symptoms. At week 4, 94.4% of subjects reported moderate-to-compete relief of AR symptoms. Total and individual NRQLQ scores and scores for sleep problems and for symptoms during sleep and on awakening were significantly reduced after 1 week of treatment. There were significant reductions from baseline in total Mini-RQLQ scores and scores for activity problems and symptoms during activity after 1 week. MFNS was well tolerated.

Conclusion: MFNS 100 mcg QD effectively reduced symptoms of AR, improved the quality of sleep and daytime activities, and was well tolerated in Chinese patients with AR.

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Frequency of Dermatophagoides mite allergens in the homes of allergic patients in Mashhad and Tabriz

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Introduction: Allergic diseases are common disorders with increasing rate during the last 30 years. almost 30% of people have a kind of allergic disorders during their life. Mites in house dust are the most important allergens in allergic disorders. The optimal growth conditions for mites are temperature and humidity and the most important sites are inside bedding, mattresses and carpets. The aim of this study is the survey of frequency of Dermatophagoides and their allergens in the homes of allergic patients.

Methods: This descriptive, cross-sectional study has been performed in Tabriz and Mashhad cities, two major industrial centers in north western and north eastern Iran, respectively, from 2005 to 2006. Twenty three allergic patients with positive skin prick test to mites from each city were selected and all of the methods for evaluation of mites and their allergens (specific antibodies of Derp and Derf, density of mites and concentrations of mite allergens) were measured. Because of high density of mites in carpets and bedding, sampling was obtained from these two sites.

Results: The frequency of mite density in the homes of allergic patients was 32.6%, 95%CI, 31.25 to 33.95; specific antibody to mite allergens was 23.9%, 95%CI: 23.78 to 24; and concentration of HDM allergens which is performed by ELISA, was not detectable.

Conclusion: Mites are common in worldwide, but the incidence of them is various in different parts of world. The frequency of mites and their allergens in two large provinces of Iran are lower than other studies, performed in other parts of world. The most important factor in frequency and growth of them is environmental humidity, and the other factors such as carpets and bedding have less importance.

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The differences of clinical characteristics between childhood and adult allergic rhinitis patients in Asians: according to ARIA classification

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Background: The prevalence of allergic rhinitis varied across centres from 0.8% to 14.9% in the 6–7-year-olds and from 1.4% to 39.7% in the 13–14-year-olds. The recent guidelines of Allergic Rhinitis and its Impact on Asthma (ARIA) group, classified it as the frequency and severity may not be appropriate for the children age group. The present study evaluates the childhood and adult patients with the ARIA classification and analyzed the clinical features of each age group.

Methods: 1,375 subjects with allergic rhinitis diagnosed by skin-prick test at our hospital from December 2005 to July 2007 were included. These patients were classifed according to age; babyhood(1–4 years), preschoolers (5–7 years), elementary schoolers (8–13 years), adolescents (14–19 years), young adults (20–35 years), adults (36–55 years), middle age (55–64 years), old age (65 years).Clinical features including nasal obstruction, rhinorrhea, nasal itching, sneezing, postnasal drip(PND), ocular symptoms and sleep disturbance and the allergen was analyzed in each age group according to the ARIA classification.

Results: A similar distribution of the ARIA classification groups were shown among all ages; Mild & intermittent group 27.5%, moderate to severe & intermittent group 17.8%, mild & persistent 18.5%, moderat to severe & persistent 34.9%. There was a higher proportion of nasal obstruction(p=0.04) and PND(p=0.042) in babyhood & preschoolers compared to adults, but ocular symptoms(p=o.01) and sneezing(p=0.01) were shown more frequently in adults. PND, sleep disturbance, nasal itching showed no distributional difference between children and adults. For allergens, housedust mite(p=0.07) showed a higher proportion in children whereas grass(p=0.01) and weed(p=0.021) was more frequent in adults. Clinical features, IgE and serum eosinophile count showed no significant relationship in each age ARIA classification group.

Conclusion: Classification of the ARIA group into different ages helded to identify the different clinical manifestations among children and adults. A new treatment strategy taking children's clinical features into account is deemed necessary.

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Chronic idiopathic urticaria and autoimmune thyroid disease

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Chronic idiopathic urticaria (CIU) persists for longer than 6 weeks, with a not determined cause. One of the interesting clinical associations is between chronic urticaria with positive autologous serum skin test (ASST) and thyroid disease. CIU and chronic tyroiditiis has not been completely investigated.

Methods: We studied 94 patients (65 female and 39 male; mean age 44.7 years; range 15–78 years) who had CIU and a history of thyroid dysfunction. Thyroid function was investigated by means of thyroid-stimulating hormone (TSH), fT3 and fT4 measurement and thyroid ultrasound. Thyroid antibodies (anti-thyroid peroxidase and anti-thyroglobulin), total serum IgE levels and ASST were performed in all patients.

Results: Antibodies to thyroglobulin and to peroxidase were positive in 26/94 patients (27.7%) with CIU and morphological or volumetric abnormalities of the thyroid gland by ultrasonography. All the CIU patients with autoimmune thyroiditis had a normal thyroid function, a negative ASST and a normal total serum IgE levels (74.3 +/− 14.9 IU/ml; normal value <100 IU/ml). Antibodies anti-thyroglobulin and anti-thyroid peroxidase were negative in 68/94 (72.3%) CIU patients, of which 44/68 (64.7%) had high total serum IgE levels (395.2 +/− 180.6 IU/ml) and a negative ASST. Four of the thirty four CIU patients (11.8%) with negative thyroid autoantibodies had a positive ASST and a normal serum IgE levels, while 16/68 (23.5%) patients had a multinodular goiter, ASST negative and normal value of total serum IgE levels.

Conclusion: We found a correlation between autoimmune thyroiditis and normal total serum IgE levels in CIU patients with ASST negative. High total serum IgE levels were found in CIU patients without thyroid disease. Thus, in CIU patients the tests to detect thyroid autoantibodies are relevant in particular in patients with normal value of total serum IgE levels. Further investigation of mast cell activation by thyroid antigens is incoming.

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Perioperative allergic reactions: a case of intraoperative anaphylactic shock

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The incidence published on perioperative allergic reactions is highly variable, fluctuating between 1:3.500 and 1:20.000 anaesthesias, with a mortality rate of 3 to 6%. During surgical procedures it is difficult to recognise an allergic reaction.Under anaesthesia , bronchospasm and hypotension are common and lacking in a set of symptoms typical of an allergic reaction;especially when they occur separately;if the response to treatment is effective, the symptoms are often not investigated and they are attributed to another cause. Furthermore, 10–14% of the reactions,primarily the most severe, affect just one system;and that,too, leads to many cases not being diagnosed. Therefore, epidemiological monitoring of these reactions is required. This is a case of intraoperative anaphylactic shock: A 59-years old female scheduled for an abdominal hysterectomy due to uterine sarcoma; her

personal records showed no known drug allergies. General anaesthesia was induced without incidents. After 15 minutes ,when beginning the surgical incision and coinciding with the start of intravenous infusion of the following drugs: cefazolin, ketorolac, ranitidine and ondasentron, a sudden severe bronchospasm appeared, followed by cardiovascular collapse with severe hemodynamic deterioration, which was treated immediately with i.v. corticoids and ephedrine without an effective response. The surgical procedure was interrupted and ,now with suspicions of a possible allergic reaction, all of the drugs that were being administered were withdrawn, i.v. adrenaline was administered up to a total of 1 mgr., forcing fluid resuscitation with crystalloids and colloids.The shock was reversed in approximately 5 minutes. The surgical procedure resumed 20 minutes after the symptoms, with clinical stability subsequently maintained. The immediate study revealed a high serum tryptase level 30 minutes and 6 hours following the reaction,the allergy study showed a decidedly positive prick test for cephalosporins, and the intradermal reaction was positive for cefazolin. The diagnosis was anaphylactic shock due to the administration of cefazolin and hypersensitivity to cephalosporins.

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Swimming in asthma increases the risk or improved respiratory functions in atopic patients

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Background: This study designed to evaluate the role of swimming in mechanics of lung in healthy individual and patients with asthma.

Methods: A total 76 girls who take part in course of regular swimming session three day per week for eight weeks enrolled in this study. All of them completed International Study of Asthma and Allergy in Childhood written questionnaire and person who suspicious to asthma or other atopic diseases referred to allergist for more evaluation. Peak expiratory flow rate was recorded for participants at beginning, one hour after swimming and two months later.

Results: According to International Study of Asthma and Allergy in Childhood questionnaire 35.4% had asthma or other atopic diseases Increase in Peak expiratory Flow rate more than 20% of personal best was seen in 21.9% after one hour swimming and in 27.6% after two months Increased in Peak expiratory Flow rate was significant in healthy individual and asthmatic patients and obese but not significant in patients with allergic rhinitis or eczema.

Conclusion: Swimming in indoor chlorinated pool not only is less asthmogenic but also can improve lung mechanics in normal and asthmatic patients. Because breathing large volume of warm and moist air are good for lung. The serious health problem with chlorinated pools is too much chlorine uses to sanitize the water and don't have adequate ventilation in indoor pool.

1005
Are fast foods and additives main culprit of increasing atopy?

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Background: Frequency of atopic disease increased in last 3 decades and hypothesis explained for this, among several factors that could be related to increasing frequency of atopic disease. Using additives and fast food have the more important role, so this study designed for checking this purpose.

Methods: 110 patients with atopic disease according to history and physical exam and Para clinic study such as total IgE, skin prick test and spirometry, compare with 110 normal healthy match group for using fast food, additives and habitual diet.

Results: In case group 46.4% had allergic rhinitis and 34.5% had asthma. Pizza was the most culprit in flare up allergic symptoms (9.1%), and the most symptoms that exaggerated with fast foods was cough (29.5%) and the next one was flare up of skin manifestation such as itching or Urticaria. The diet with out curry and additive causes fewer symptoms in atopic patients and healthy individuals.

Conclusion: Fast foods and additives can increase and exaggerate symptoms of atopic patient, they have important role in triggering allergic symptoms in patients with atopic back ground.

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Biphasic anaphylaxis after multiple Hymenoptera stings in patient with mastocytosis

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Of note the mastocytosis is an important factor of risk for severe anaphylaxis, in particular from Hymenoptera stings. There is very few documentation about anaphylaxis with bifasic course during stings. Equally multiple Hymenoptera stings do not seem to affect additional risk for anaphylactic reactions.

Case report: Male, 65 years old with anamnesis of urticaria pigmentosa. In the past we observed urticaria angioedema after assumption of FANS and assumption of amoxicillin/clavulanic acid. During the last years of observation the patient present recurrent abdominal pain, headache and flashes. He came in hospital after 5 vespides stings, with immediate symptoms of diffused erythema, dyspnoea and loss of conscience, for a long time. He was discharged after 6 hours of observation. After 12 hours from the multiple stings he showed relapse of diffuse erythema and dyspnoea, that due to a new recovery.

We found Positive Skin tests for polistes Dominulus to 0.1 mcg; Positive RAST to 1,23 KU. Tryptase 39,5 ng/ml. He now still presents clinician symptoms of systemic mastocytosis with presence of 1 minor criterion. The patient is now attending of bone marrow diagnostyic biopsy to confirmation. The venom immunotherapy with Polistes dominulus venom has been assigned to the dose of 200 mcg and well tolerated from the patient. It would be possible that multiple hymenoptera stings and systemic mastocytosis had caused a reaction with severe symptoms and biphasic course.

1007
Does prolonged breastfeeding prevent asthma and allergic diseases?

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Background: Medical literature regarding the relationship between infant feeding practices and the development of childhood asthma and allergic diseases is conflicting with hardly any studies from developing countries.

Aim: The aim of this study is to assess the relationship between breast-feeding (BF) and the development of childhood asthma, recurrent wheezing, and allergic diseases after adjustment for maternal asthma.

Design: This is a cross sectional survey.

Setting: Immunization Centre and Pediatric Department at the 11 Primary Health Care Centers and Hamad General Hospital, Hamad Medical Corporation, State of Qatar.

Subjects: A multistage sampling design was used and a representative sample of 1500 infants and pre-school children with age range of 0–5 years and mothers aged between 18 to 47 years were surveyed during the period from October 2006 to September 2007 in Qatar. A total of 1278 mothers (85.2%) made consent to participate in this study.

Methods: A confidential, anonymous questionnaire was completed by the selected subjects assessing BF and allergic diseases. Questionnaires were administered to women who were visiting Primary Health Centers for immunization of their children. Questionnaire included asthma, recurrent wheezing allergic rhinitis. Additional questions included breast feeding and duration, passive tobacco exposure, number of siblings, family income, level of maternal education, family history of allergies. Univariate and multivariate statistical methods were performed for statistical analysis.

Results: Out of the 1500 child mothers, 1278 mothers agreed to participate in this study with the response rate of 85.2%. More than half of infants 758(59.3%) initially were exclusively breastfed and followed by partial breastfed (29.9%) and formula fed (10.8%). Length of breastfeeding was associated with maternal age. Of these infants, prevalence of allergic diseases in general and asthma (15.6%) allergic rhinitis (22.6%), and wheezing (12.7%) were significantly less frequent in those with prolonged and exclusive BF. The association between breastfeeding and asthma tended to be similar in children with positive family history of asthma ($p<0.001$) and allergic rhinitis ($p<0.001$) compared children without.

Conclusion: The current study indicated that exclusive BF has protective effect against asthma and allergic diseases. Educated women were aware of the importance of the BF. The study findings suggested possible avenue for intervention to increase awareness and increase of BF. Stressing the practice of BF could be one of effective measures directed against development childhood asthma and allergic diseases in third world countries.

1008
The association of prolonged breastfeeding and allergic disease: does breast-feeding reduce the risk for childhood eczema?

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Background: Prolonged breast-feeding (BF) was shown to reduce the risk of diseases. In 2001 the World Health Organization (WHO) issued a revised global recommendation that mother should breast feed exclusively for six

months. This recommendation is likely to influence national policies on the recommended age for the first introduction of solids, so it is important that pediatricians are aware of the issues and the evidence (or lack of it) on which the WHO recommendation is based. The fact that BF may protect against allergic disease remains controversial, with hardly any reports from developing countries.

Aim: The aim of this study to determine whether the protective effect of longer breast-feeding on the risk of allergic diseases and eczema.

Design: This is a cross sectional survey.

Setting: Immunization Centre and Pediatric Department at the 11 Primary Health Care Centers and Hamad General Hospital, Hamad Medical Corporation, State of Qatar.

Subjects: A multistage sampling design was used and a representative sample of 1500 infants and pre-school children with age range of 0–5 years and mothers aged between 18 to 47 years were surveyed during the period from October 2006 to September 2007 in Qatar. 1278 mothers (85.2%) gave consent to participate in this study.

Methods: A confidential, anonymous questionnaire was completed by the selected subjects assessing BF and allergic diseases. Questionnaires were administered to women who were visiting Primary Health Centers for immunization of their child. Questionnaire included allergic rhinitis, wheezing, eczema, and additional questions included breast feeding and duration, tobacco smoke exposure, number of siblings, family income, level of maternal education, and family history atopy. Univariate and multivariate statistical methods were performed for statistical analysis.

Results: Out of the 1278 infants included in the study, more than half of infant 758(59.3) initially were exclusively breastfed. Length of BF was associated with maternal age [29.1±5.6, mean±SD]. Of these children, prevalence of allergic diseases in general and allergic rhinitis (22.6%), wheezing (12.7%) and eczema (19.4%) were significantly less frequent in those with prolonged and exclusive BF. The association between BF and eczema tended to be similar in children positive family history of atopy ($p<0.001$) and eczema ($p<0.001$) compared to those without.

Conclusion: This study confirms that prolonged duration of BF has protective effect against development of allergic diseases and eczema in children. This opens a possible avenue for intervention to increase awareness and enforce BF practices as one possible way to reduce the risk of onset of childhood allergic diseases and eczema.

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Coincidence of gastroesophagealreflux and laryngopharengeal reflux in our asthmatics patients

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Introduction and aim: We don't know certainly, whether or not GER is a cause of the asthma or a result of it. Esophagitis has been shown in 30%–50% of patients with typical symptoms of heartburn. Nonesophageal manifestations of the regurtation of gastric contents promotes the use of the term extraesophageal reflux. EER should be considered in patient. We aimed to investigate the prevelance of GER and laryngopharengeal reflux (LFR) symptoms in asthmatic patients and to determine the contribution of the laboratory findings (endoscopic evaluation and pulmonary function test) to the symptom based diagnosis.

Materials and Methods: We investigated 37 asthmatics patients having primary reflux symptoms; heartburn and regurtation. At the same time we asked other gastroesophageal and laryngopharengeal reflux symptoms. We performed pulmonary function test and pulmonary carbon monoxide diffusion test to all of the patients. All of asthmatic patients with GER were performed esophagogastrodoudenoscopy by gastroenterologist and laryngoscopy by ENR phycisian. All statistics were performed by using UNISTAT 5.1 statistics pack program.

Results: Heartburn frequency was daily in 28.1%, a few days in a week in 18.8% and once a week in 26.6% of the patients. Regurtgitation was occuring daily in 34.4% and once a week in 23.4% of the cases. 34.4% of the cases were complaining of daily globus. FVC percentage was lower in severe cases when compared to mild regurgitation ($p=0.024$). CO monoxide diffusion capacity did not have a statistical difference according to severity of the symptoms. The pulmonary diffusion capacity of obese cases were lower than the cases with BMI < 29 ($p=0.038$). Reflux esophagitis in 24 of cases were also confirmed with pathological diagnosis. Avarage refflux finding score (RFS) was calculated as 5.2±4.15(min 0–max 15). Posterior comissure hypertrophy was the most common finding 72%.

Conclusion: Laryngoscopic findings and RFS did not help in diagnosis. We determined that reflux symptoms did not have any effect on reflux esophagitis. Diffusion capacity changes result from high body mass index. Conducting controlled studies with different diagnostic methods and a larger number of case series which will contribute to the symptom based reflux diagnosis are needed.

1010

Doxycycline reduces ECP, MMP9 and nasal polyp size, in a double-blind, randomized, placebo controlled, multicenter trial

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Background: Recent evidence suggests that a chronic microbial trigger might play an important role in the pathogenesis of chronic rhinosinusitis with nasal polyposis (NP). For treatment, doxycycline was selected based on its antimicrobial effect and anti-inflammatory effect. The aim of the study is to evaluate the effect of oral doxycycline on nasal polyp size, nasal peak inspiratory flow (nPIF), signs and symptoms of nasal polyps.

Methods: In a randomized, double-blind, placebo controlled, parallel group, multicenter study, 32 subjects with bilateral nasal polyps were randomized to receive placebo or antibiotics at a dose of 200 mg doxycycline at day one, followed by 100 mg doxycycline from day two till day 20. The effect was assessed by endoscopic evaluation of polyp size, symptoms, peripheral eosinophil counts, local IgE, MPO, MMP-9 and ECP levels.

Results: Doxycline treatment resulted in a significant decrease of the endoscopic nasal polyp score after 4 to 12 weeks after the start of the doxycline treatment compared to placebo. The concentrations of IgE, MPO, MMP-9 and ECP decreased significantly in nasal secretions of doxycycline treated patients.

Conclusion: This is first study demonstrating the positive clinical effect of doxycyline on nasal polyposis, paralleled by a sign impact on markers of local inflammation and remodelling. Further, in a clinical setting the use of doxycycline offers an additional advantage of providing both antimicrobial and anti-inflammatory effects resulting in a decrease in nasal polyp size.

1011

Diagnosis of pollen sensitization using natural and recombinant allergen in children with bronchial asthma

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Birch is one of the main causes of pollinosis in children with bronchial asthma in Europe and in Moscow. The aim of this study was to evaluate the potential of natural and recombinant allergens from tree and grass pollen (Bet v1, Bet v2, Phl p1, Phl p2, Phl p5, Phl p6, Phl p7) as diagnostic reagents in comparison with standardized extract.

Methods: Prick-test with commercial pollen extracts were conducted in 31 children with bronchial asthma aged 8–15 years. Recombinant forms of both allergens were determined with ImmunoCAP.

Results: Of 15 children with prick-test of birch extracts (++), nine patients displayed low levels of Bet v1, Bet v2; four children had moderate and two had high allergen-specific IgE concentration. Four of six children with skin prick-tests (++++) had low levels of allergen-specific IgE. Two of three children with skin prick-tests (+++++) has low levels of allergen-specific IgE against birch while 1 child had medium level. Only seven children were sensitized to timothy-grass pollen. The concentration of allergen-specific IgE against timothy-grass correlated with skin prick-tests results as three children with ++ tests and four children with +++ and ++++ tests revealed very high levels of Phl p5 and Phl p7.

Conclusion: Prick-tests with natural extracts of birch pollen did not show 100% sensitivity with recombinant allergens while timothy-grass allergens sensitivity was significantly higher.

1012

Correlation between fungus-specific IgE and skin test for fungus allergens in patients with chronic cough or bronchial asthma

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Skin tests can provide useful confirmatory evidence for a diagnosis of specific allergy in patients with chronic cough and bronchial asthma. Skin prick tests are relatively good correlation with specific IgE, but skin tests with fungi are not. We aimed to investigate the correlation between fungus-specific IgE and skin tests in patients with chronic cough and asthma.

Methods: We measured fungus-specific IgE(Penicillium notatum, Cladosporium herbarum, Aspergillus fumigatus, Alternaria alternata, Fumigarium moniliforme) testing with immuoCAP in 537 patients with chronic cough and bronchial asthma., who were 1) all negative or 2) positive for one or more of fungi or 3) positive for one mixed threshings in skin tests with common 55 allergens.

Results: 1. In 537 patients with chronic cough and asthma, 340 patients were all negative, 34 patients were positive only for one or more fungi, 53 patients were positive for one or more fungi with other one or more common allergens and 62 patients were positive only for mixed threshings. 2. Total agreement rate of 5 fungous allergens between skin test and fungus-specific IgE is 95.2%, but 8 patients(2.4%) were positive for one or more fungus-specific IgEs among 340 patients with all negative skin tests. 9 patients(8.2%) were positive for one or more fungus-specific IgEs among 110 patients with positive for mixed threshings skin tests and negative fungus skin test. 3. The positive agreement rate of Alternaria alternata-specific IgE was 41.7%(5/12), that of Fumigarium moniliforme-specific IgE was 42.9%(3/7), that of Penicillium notatum-specific IgE in positive skin tests was 70%(7/10), that of Cladosporium herbarum-specific IgE was 100%(1/1), and that of Aspergillus fumigatus-specific IgE was 100%(1/1). 4. The positive agreement rate of skin test for Alternaria alternata in positive Alternaria alternata -specific IgEs was 41.7%(5/12), that for Fumigarium moniliforme was 50%(3/6), that for Penicillium notatum in positive Penicillium notatum-specific IgEs was 63.6%(7/11), that for Cladosporium herbarum was 11%(1/9), and that for Aspergillus fumigatus was 3.6%(1/28).

Conclusion: Fungus skin tests did not provide useful evidence for a diagnosis of fungus-specific allergy in patients with chronic cough and bronchial asthma. The clinical significance of serum fungus-specific IgEs will be evaluated. Fungus-specific IgE may be considered to measure in mixed threshings-positive patients with chronic cough and bronchial asthma.

1013

Tobacco smoking is a risk factor for asthma symptoms in teenagers independent of weight and antioxidant diet

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Background: Multiple risk factors have been associated to the presence of asthma symptoms, including overweight and environmental tobacco smoke (ETS); however active smoking and the possible attenuation of its effects by antioxidant diet have not been described in adolescents.

Methods: Cross sectional survey of 3000 13–14 year old school attendants, in north Argentina as part of the ISAAC study. Self reported questionnaires were used to determine the prevalence of wheezing in last 12 months, and the presence of parents smoking at home and active smoking. Food intake habits allow grouping those having fruit and vegetables more than 3 times per week, and body mass index (BMI) was calculated from weight and height described. Contingency table was applied in order to determine the odds ratio and 95% confidence interval (OR, 95% CI) for each variable; and logistic regression analysis was used with all the variables related to wheezing.

Results: 55% were female, and 12.5% reported wheezing in last 12 months; 12.9% of adolescents are active smokers and 49.9% have at least one parent smoking. 67.8% of them eat fruit and vegetables regularly, and obese were considered together with overweight since the prevalence of BMI above 25 is only 5.1%. The odds of wheezing for active smokers were 1.85 (95% CI 1.40–2.44), and for passive smoking 1.48 (95% CI 1.18–1.84). Neither gender, nor BMI or antioxidant diet were significantly associated to the presence of wheezing; when applying logistic regression, none of them could modify the effect of active ($p = 0.001$) and passive smoking ($p = 0.006$).

Conclusion: Smoking is a significant risk factor for asthma symptoms, not affected by gender, BMI and antioxidant diet. Both active and passive smoking confirm their detrimental effects on respiratory symptoms in adolescent population.

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