Supplementary information S3 (box) | Natural antisense transcripts in genomic imprinting

There are several studies suggesting a pivotal role for antisense transcripts in imprinted genes, such as IGF2R[1], KCNQ1[2], UBE3A[3], ATP10C, MKRN3, MAGEL2, NDN[4], Slc22a2, Slc22a3[5], GNAS[6] and Gnas[7].

***Kcnq1ot1* transcript:** The antisense RNA, *Kcnq1ot1*, is essential for the silencing activity of the *Kcnq1* imprinting control region, which in turn controls imprinting of a cluster of neighbouring genes on chromosome 11. Antisense transcript appears to recruit repressor complexes, turning chromatin into an inactive state[8]. Suppressive chromatin modifications are spreading in both direction to the neighbouring genes, similar to the X chromosome inactivation but with a limited penetrance[9]. *Kcnq1ot1* antisense RNA was shown to be involved in both the establishment of transcriptional silencing as well as in maintenance of silencing through subsequent cell divisions[10].

***Air* transcript:** A region that contains three imprinted, maternally expressed protein-coding genes (*Igf2r*/*Slc22a2*/*Slc22a3*) on chromosome 6 has been shown to be controlled by a paternally expressed noncoding *Air* RNA[5]. Expression of *Air* (named Air, for antisense Igf2r RNA) correlates with repression of all three genes[11]. *Air* is a 108-kb unspliced and repeat-rich transcript, overlaps with just one of these genes in an antisense orientation[12]. However, *Air* is required for silencing of all three genes, which is likely through a similar mechanism shown for the *Kcnq1* imprinted locus.

***UBE3A-ATS* transcript:** In the case of ubiquitin ligase E3A (*UBE3A*), natural antisense transcript *UBE3A-ATS* is a very long (460-kb), spliced noncoding RNA encoded on chromosome 15. Paternal expression of *UBE3A-ATS* is responsible for monoallelic (maternal) expression of the *UBE3A* gene in the brain[3,13,14]. *UBE3A-ATS* lies within a highly complex locus containing several other imprinted genes including *ATP10C* and *UBE3A,* both show genomic imprinting[15,16].

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