VIROLOGICA SINICA

Electronic Supplementary Material

Serological Evidence of Bat SARS-Related Coronavirus Infection in Humans, China

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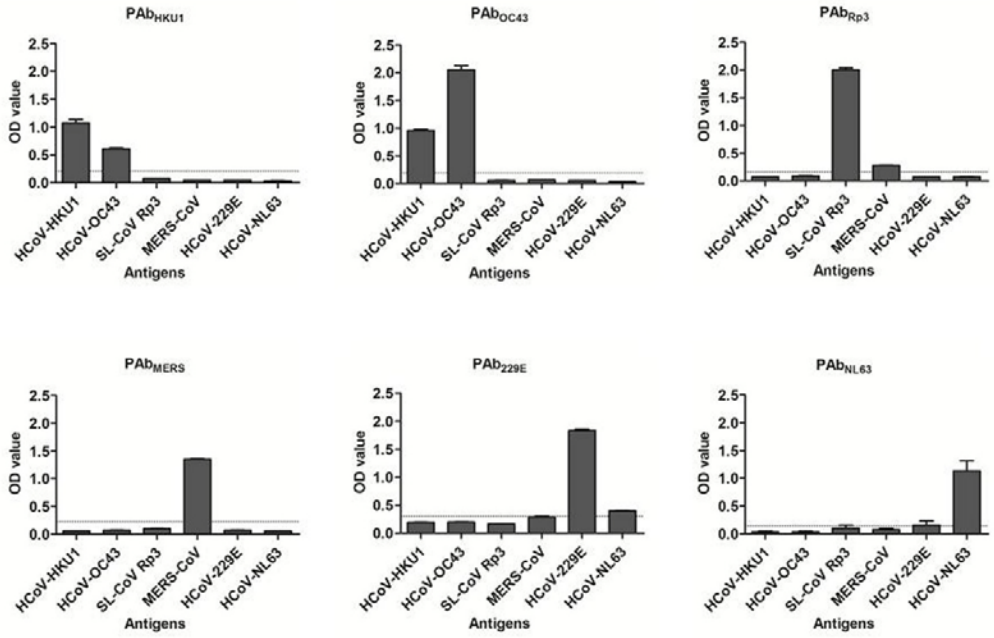

Figure S1. Two-way cross-reaction ELISA testing between 6 coronavirus NPs and their corresponding rabbit polyclonal antibodies. The NP proteins (100 ng/well) were coated in 96-well micro-plate and tested with polyclonal antibody against NPs of SARS-related CoV Rp3 (PAb_{Rp3}), HCoV HKU1 ($PAb_{HKU1)}$, HCoV OC43 (PAb_{OC43}), MERS-CoV (PAb_{MERS}), HCoV229E (PAb_{229E}) and HCoV NL63 (PAb_{NL63}), respectively. The serum was diluted at 1:16,000 or 1:64,000 (for PAb_{229E} and PAb_{NL63}). HRP labeled goat anti-rabbit IgG (1:20,000) was used as secondary antibody and detected with TMB substrate. The horizontal line in the diagram indicates cutoff value determined from negative rabbit sera collected before immunization.

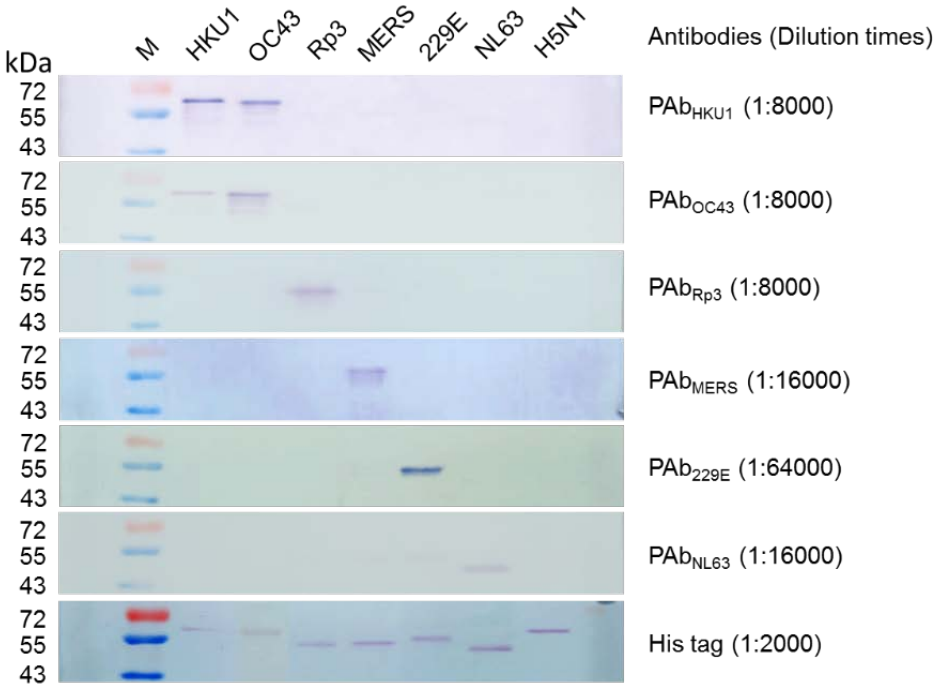

Figure S2. Two-way cross-reaction Western blotting between 6 coronavirus NPs and their corresponding rabbit polyclonal antibodies. The NP proteins (100 ng) were run on 12% SDS-PAGE and transferred to polyvinylidene difluoride membrane (Roche Diagnostics GmbH, Mannheim, Germany). The membrane was incubated with the different rabbit sera at different dilutions indicated on the right (in brackets). Goat anti-rabbit IgG conjugated with AP (Proteintech, Wuhan, China) were used for detection at a dilution of 1:2000. Influenza virus H5N1 NP was used as negative control. Numbers at the left are molecular masses (in kilodaltons).