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Supplementary information

TMPRSS2 is a functional receptor for human coronavirus HKU1

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TMPRSS2 is a functional receptor for human coronavirus HKU1

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Supplementary movie 2. Time lapse microscopy of TMPRSS2-scarlet-I (red) and HKU1A S-NeonGreen (green) expressing cells. 293T cells were transfected either with TMPRSS2-scarlet-I or HKU1A S-NeonGreen. At 24 h post transfection, the cells were mixed and imaged every 150 seconds for 2.5 h at 37°C, 5% CO_2. Two representative movies are shown.

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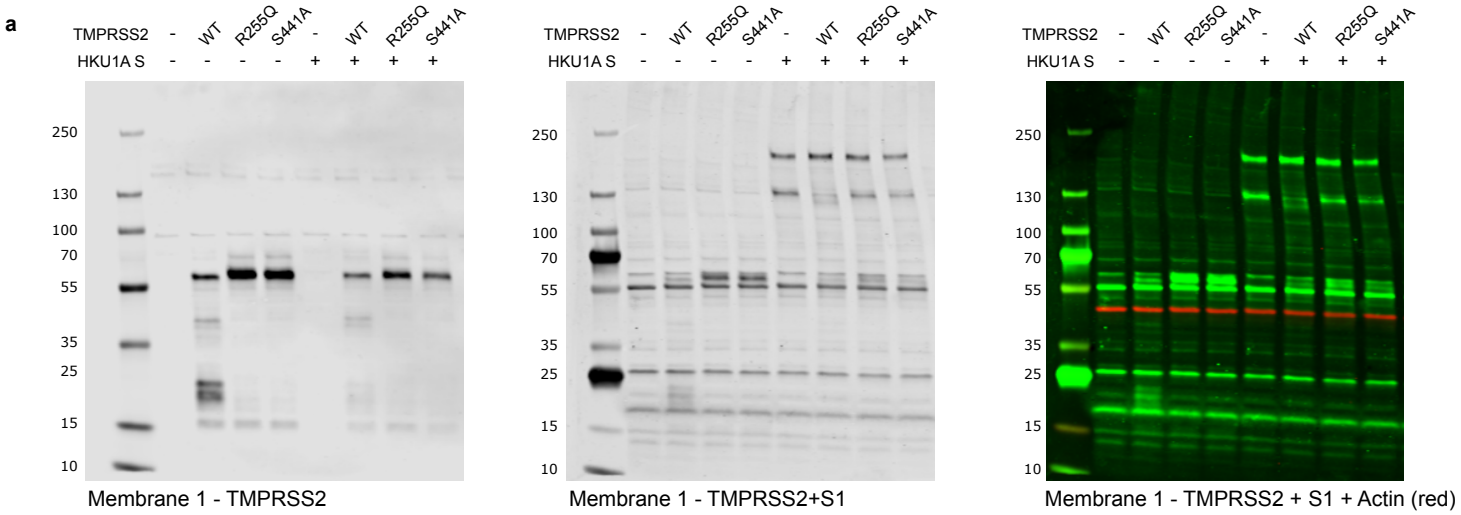
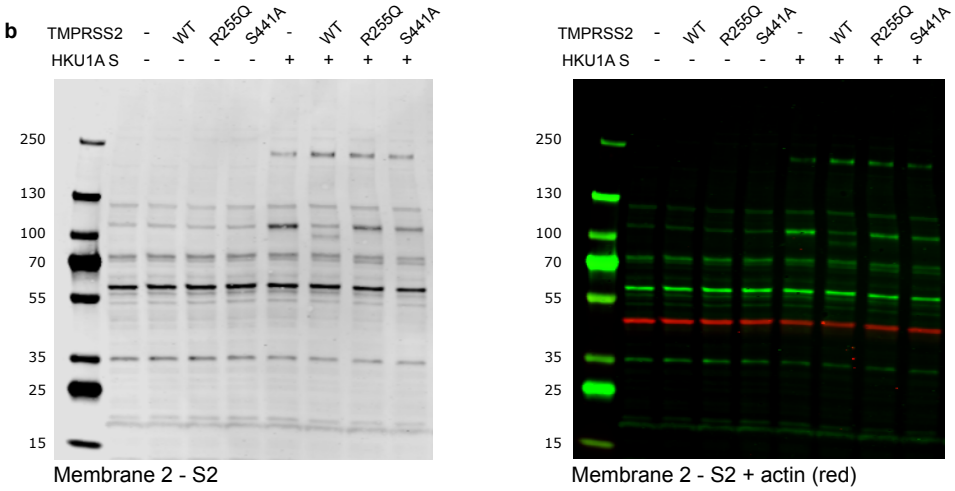
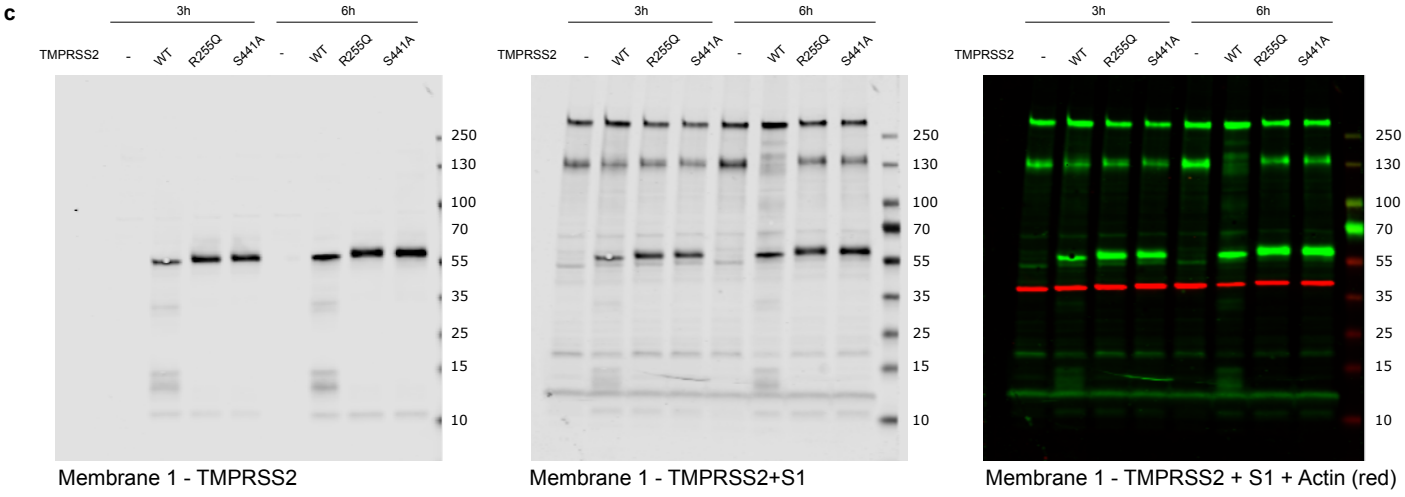
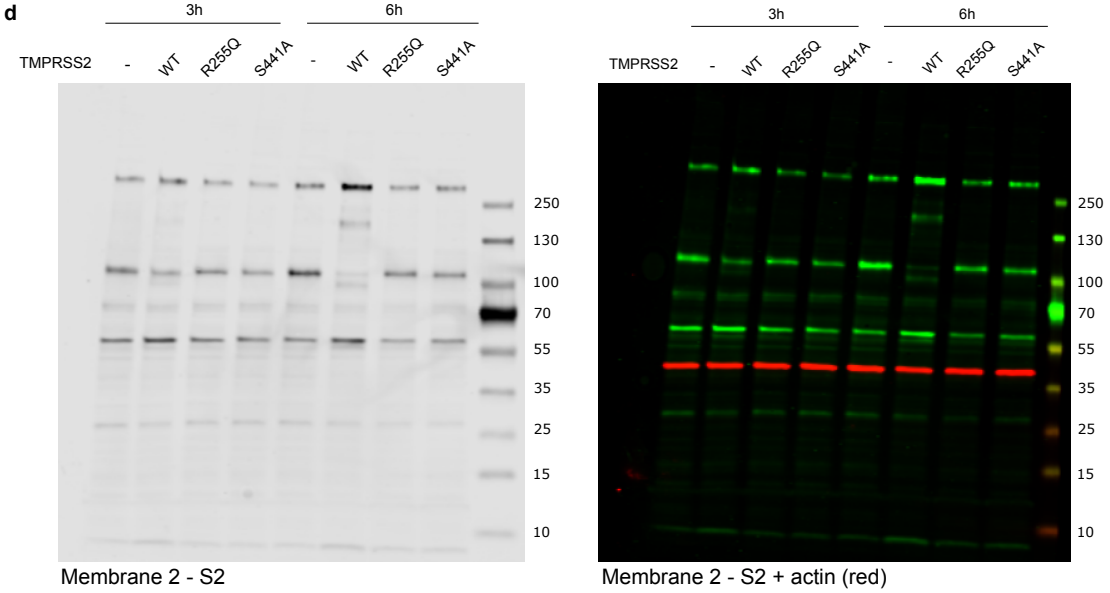
Supplementary information 1



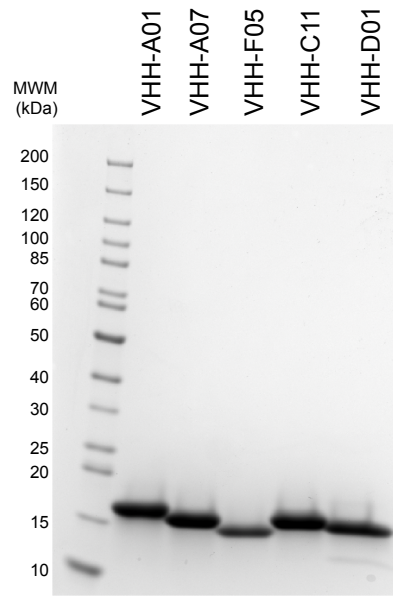






Supplementary information 2

a



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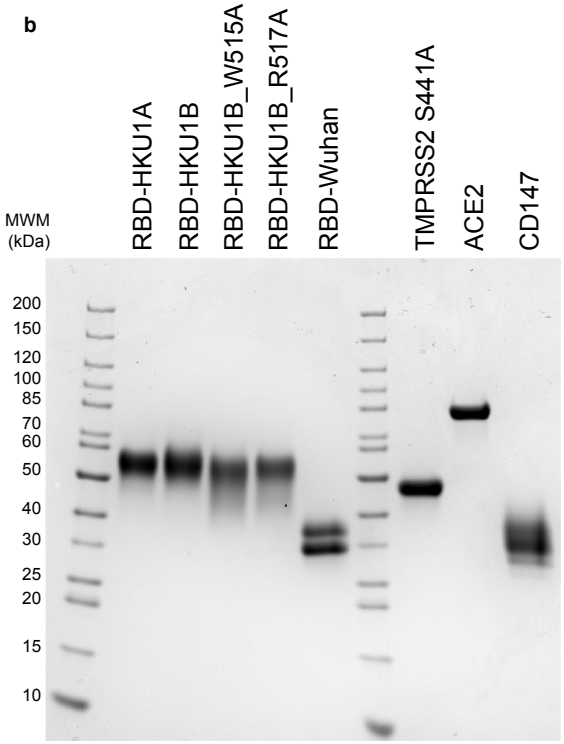

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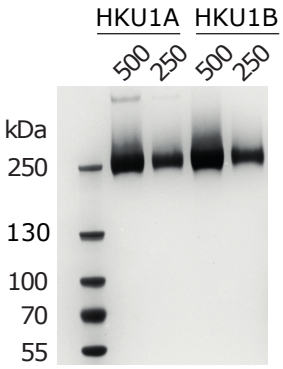
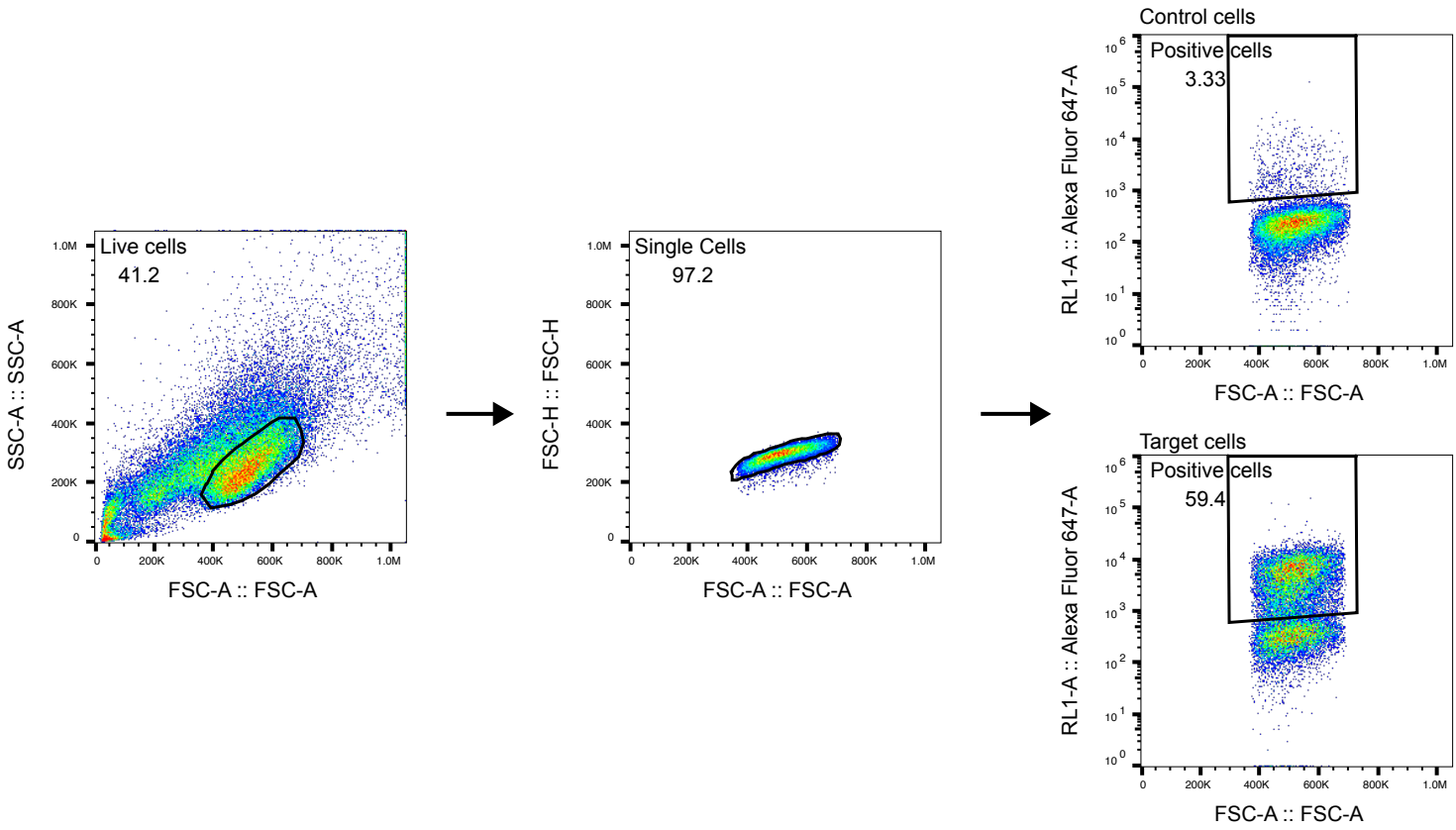


d

Protein	Expected MW (kDa)	Number of glycosylation sites
VHH-A01	15.3	-
VHH-A07	15.5	-
VHH-F05	14.7	-
VHH-C11	15.5	-
VHH-D01	15.2	-
RBD-HKU1A	37.1	5
RBD-HKU1B	36.7	5
RBD-HKU1B-W515A	36.7	5
RBD-HKU1B-R517A	36.7	5
RBD-SARS-CoV-2	27.3	1
TMPRSS2 S441A	46.9	2
ACE2	70.0	6
CD147	22.5	3
S-HKU1A	148.2	28
S-HKU1B	145.2	30

Supplementary information 3



VHH Sequences

Monomeric VHH sequences :

>A01
QVQLVESGGGLVQPGGSLRLSCVVSGFSLDYYAIGWFRQAPGKEREGVSCIGSSGDKTNYADSVKGRFTISRDNAK
NTVYLQMNSLKPEDTAVYYCAAESALYSDCTEEQNPMLYDYWGQGTQVTVSS
>A07
QVQLVESGGGLVQPGGSLRLSCTSSGSPLEHYDIIWFRQAPGREREGVSSITTSGGHTNYADSVKGRFTISRDNAKN
VVYLQMNSLKPEDTAVYYCAGRVGGRRNWIVPLDGYDNAYWGQGTQVTVSS
>C11
QVQLVESGGGLVQPGGSLRLSCAASGFTLDYYDIYWFRQAPGKEREGVSSITTSGGRTNYADSVKGRFTISRDNAK
NTVYLQMNSLKPEDTAVYYCAAKVGGRRNWIAPLNGYENALWGKGTLVTVS
>D01
EVQLVESGGGLVQPGGPLRLSCASSGSTLEHYDIGWFRQVPGGLREGVSSITASGGRTNYADSVKGRFTISRDNGKN
AVYLQMNSLKPEDTAVYYCAGKIGGRRNWVAPLDGFENAYWGQGTQVTVSS
>F05
EVQLVESGGGLVQPGGSLRLSCAASGFTLDYYAIGWFRQAPGKEREGVSCISSSGDSIKYVDSVKGRFTISRDNAKN
TVYLQMNSLKPEDTAVYQCAADALGSGCLTGNYDYWGQGTRVTVSS
CDR1-CDR2-CDR3

VHH-Fc sequences :

>TMPR-A1-*Fc-humain* (VHH-A01-Fc)
QVQLVESGGGLVQPGGSLRLSCVVSGFSLDYYAIGWFRQAPGKEREGVSCIGSSGDKTNYADSVKGRFTISRDNAK
NTVYLQMNSLKPEDTAVYYCAAESALYSDCTEEQNPMLYDYWGQGTRVTVSS*EPKTPKPQPAAARSDKTHTCPPCP*
APELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQ
DWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYK
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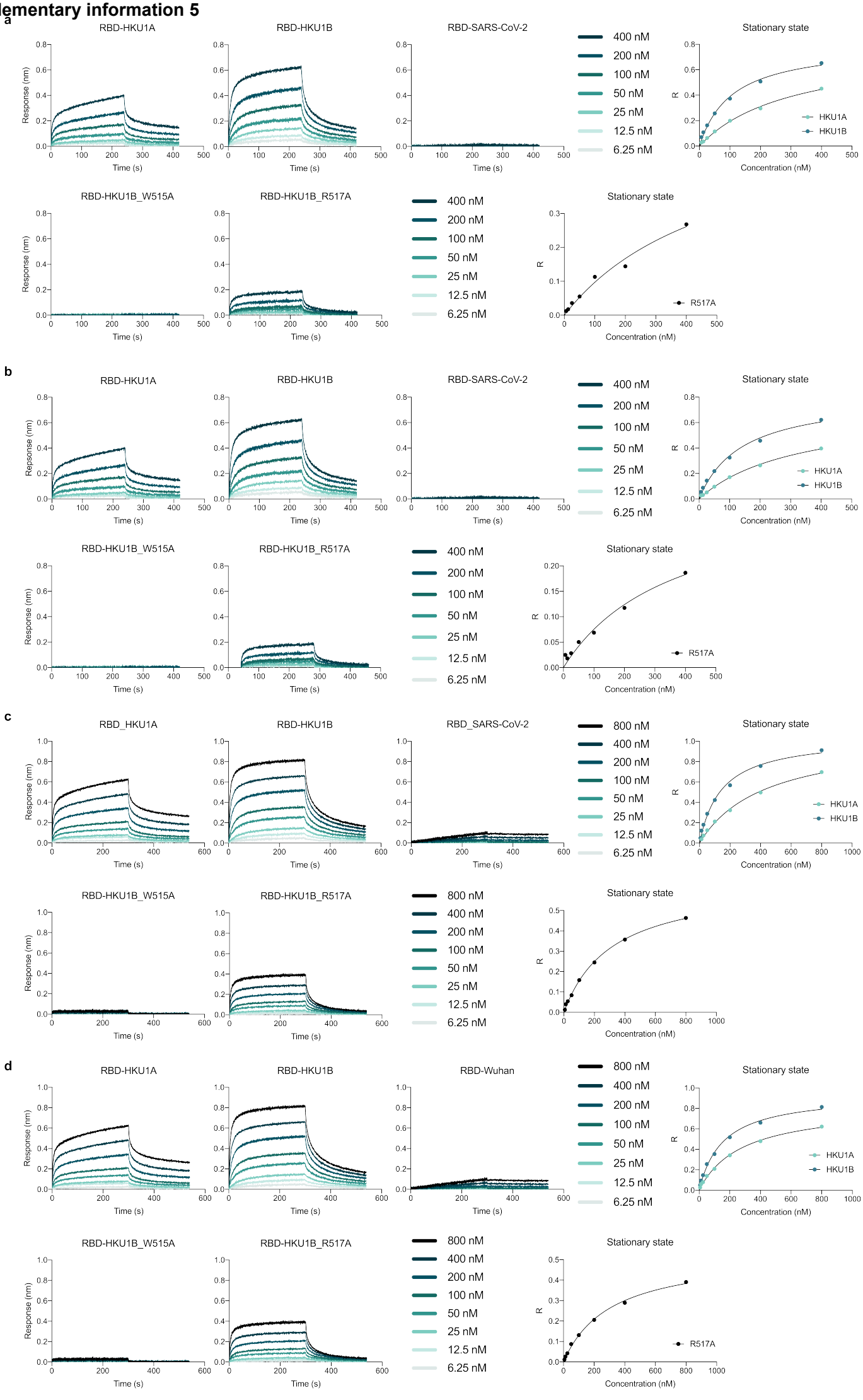
Supplementary information 5



Supplementary table 1

Replicate	RBD	Kd	95% CI	R^2	Range tested
1	HKU1A	304.5	226.6 to 433.3	0.9967	6.25-400 nM
2	HKU1A	232.1	245.1 to 440	0.9975	6.25-400 nM
3	HKU1A	430.1	328.6 to 574.4	0.9965	6.25-800 nM
4	HKU1A	279.6	221.1 to 356.9	0.9964	6.25-800 nM
1	HKU1B	106.9	75.9 to 152.6	0.9922	6.25-400 nM
2	HKU1B	140.6	94.9 to 213.5	0.9913	6.25-400 nM
3	HKU1B	144.2	107.6 to 194.2	0.9928	6.25-800 nM
4	HKU1B	155.1	119.8 to 201.5	0.9945	6.25-800 nM
1	HKU1B_R517A	515.1	211.9 to 3327	0.9812	6.25-400 nM
2	HKU1B_R517A	363.8	150.8 to 1523	0.9735	6.25-400 nM
3	HKU1B_R517A	316.9	267.6 to 377.3	0.9983	6.25-800 nM
4	HKU1B_R517A	308.0	246.7 to 388.0	0.9971	6.25-800 nM