Supporting Information

Antibody responses to endemic coronaviruses modulate COVID-19 convalescent plasma functionality

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This supporting information includes:

Figures S1-S6

Tables S1 and S2

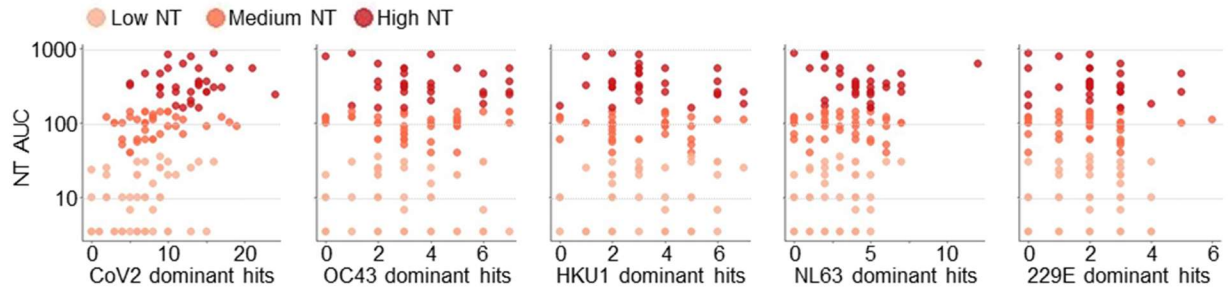

Figure S1. Polyclonality of antibody responses to CoV2 and NL63 immunodominant regions is associated with increased NT AUC

Number of reactive peptides from immunodominant regions of each coronavirus was compared to NT AUC. Polyclonal responses to CoV2 and NL63 correlate with increase NT AUC (Pearson's correlation, CoV2 $p < 10^{-8}$, R=.49; NL63 p=0.02, R=.21). Low NT: n=55, Medium NT: n=39, and High NT: n=32.

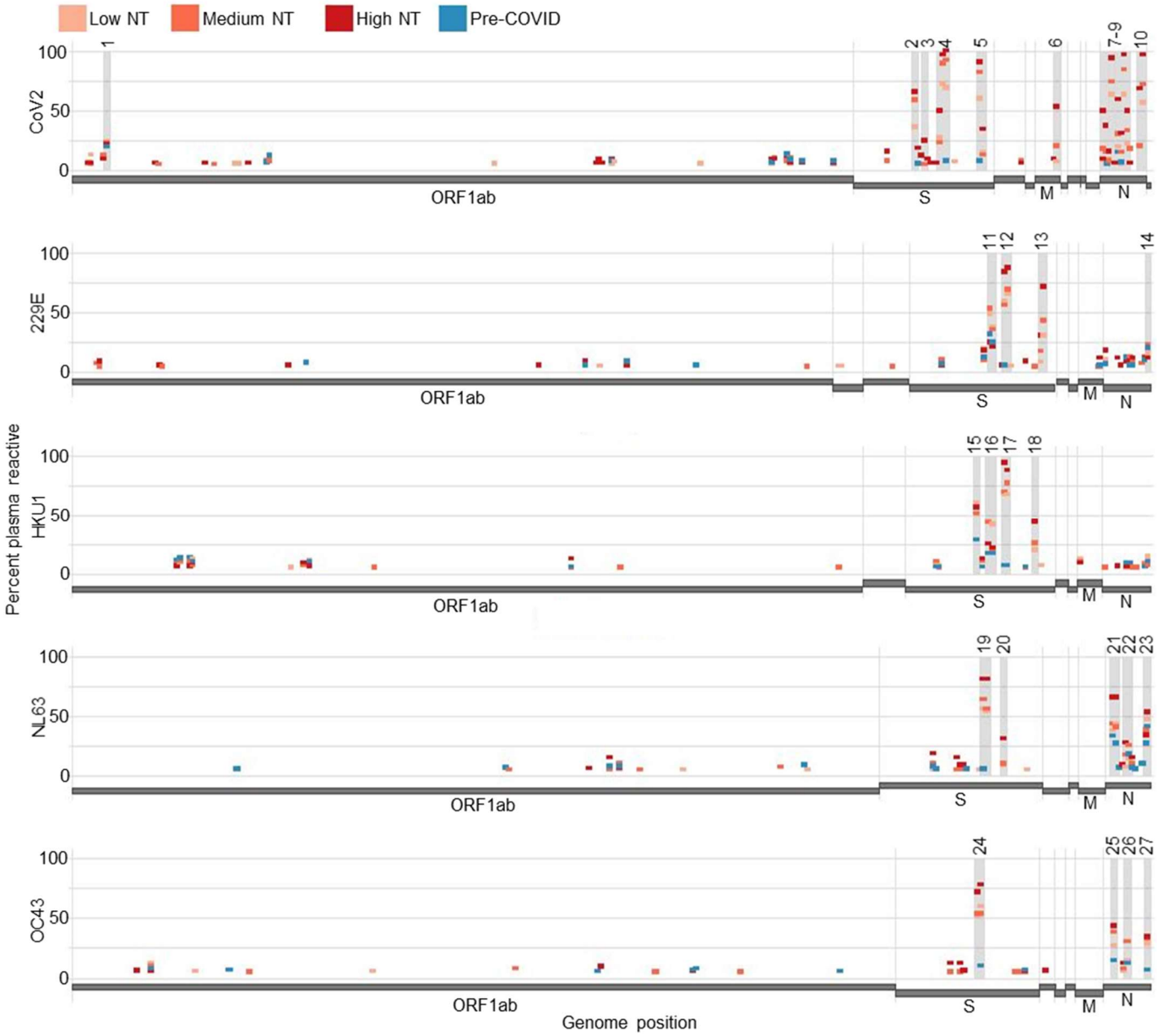

Figure S2. Whole genome reactivity plots
Antibody reactivity plots analogous to those in Figure 1 were created that include the poorly reactive ORF1. Pre-COVID: n=87, Low NT: n=55, Medium NT: n=39, and High NT: n=32.

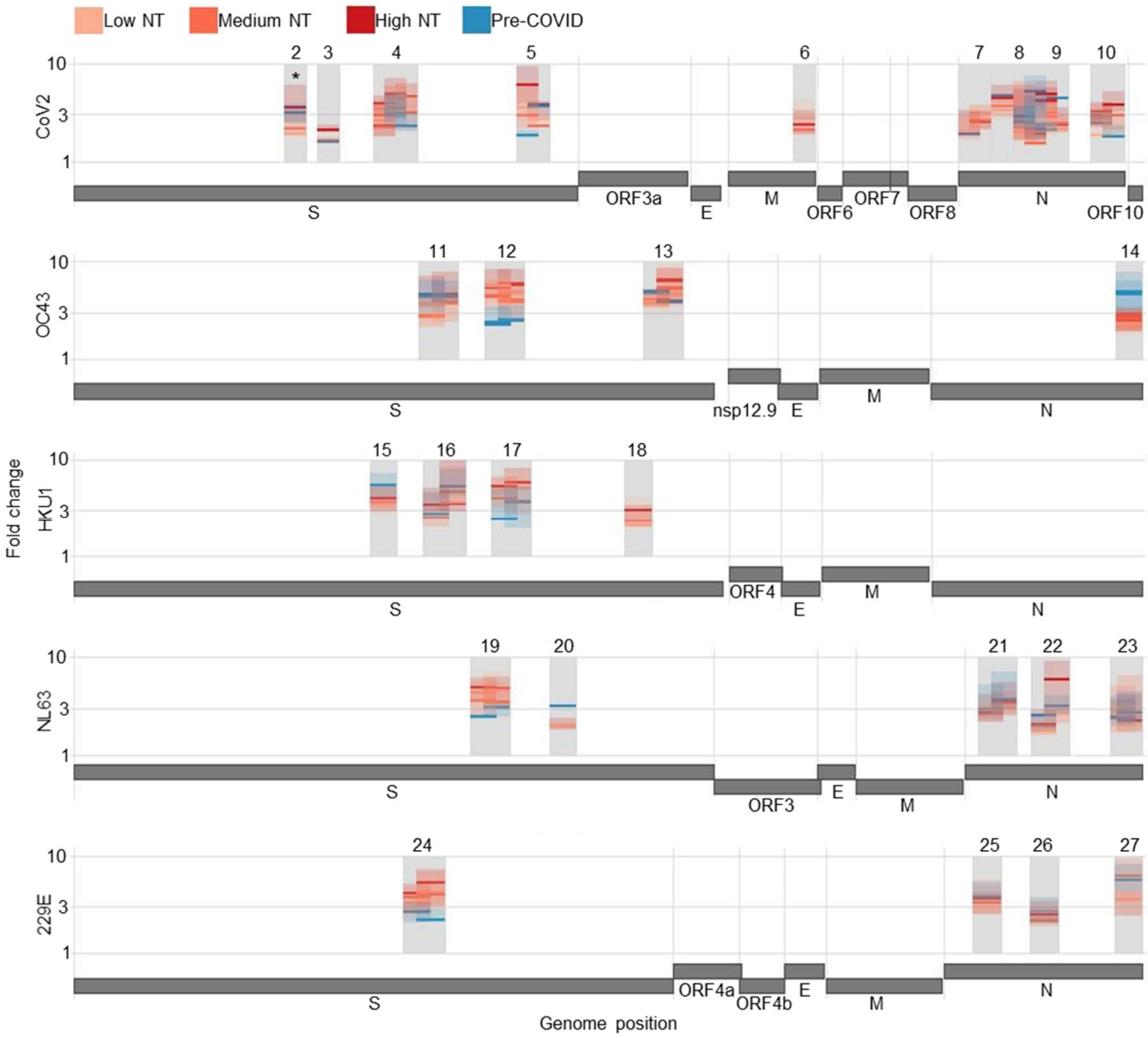

Figure S3. Magnitude of peptide reactivities does not distinguish plasma functionality
The median and interquartile range of antibody reactivity for each sample group is plotted for each immunodominant peptide. Pre-COVID: n=87, Low NT: n=55, Medium NT: n=39, and High NT: n=32. One CoV2 S immunodominant peptide (residues 533-588) indicated by asterisk show greater magnitude reactivities in High NT COVID-19 convalescent plasma compared to Medium NT or Low NT CCP (Wilcox test vs Medium NT $p=0.014$, vs Low NT $p<0.001$).

	Virus	Segment	Domain (Spike Only)	Start	End	Amino Acid Sequence
1	SARS-CoV-2	ORF1		281	336	IKTIQPRVEKKKLDGFMGRIRSVYPVASPNECNQMCLSTLMKCDHCGETSWQTGDF
2	SARS-CoV-2	S	RBD	533	588	LVKNKCVNFNFNGLTGTGVLTESNKKFLPFQQFGRDIADTTDAVRDPQTLEILDIT
3	SARS-CoV-2	S	CS	617	672	CTEVPVAIHADQLTPTWRVYSTGSNVFQTRAGCLIGAEHVNNSYECDIPIGAGICA
4	SARS-CoV-2	S	FP	757	812	GSFCTQLNRALTGIAVEQDKNTQEVFAQVKQIYKTPPIKDFGGFNFSQILPDPSKP
				757	812	GSFCTQLNRALTGIAVEQDKNTQEVFAQVKQIYKTPPIKDCGGFNFSQILPDPSKP
				785	840	VKQIYKTPPIKDCGGFNFSQILPDPSKPSKRSFIEDLLFNKVTLADAGFIKQYGDC
				785	840	VKQIYKTPPIKDFGGFNFSQILPDPSKPSKRSFIEDLLFNKVTLADAGFIKQYGDC
				813	868	SKRSFIEDLLFNKVTLADAGFIKQYGDCLGDIAARDLICAQKFNGLTVLPPLLTDE
5	SARS-CoV-2	S	HR2	1121	1176	FVSGNCDVVIGIVNNTVYDPLQPELDSFKEELDKYFKNHTSPDVDLGDISGINASV
				1149	1204	KEELDKYFKNHTSPDVDLGDISGINASVVNIQKEIDRLNEVAKNLNESLIDLQELG
6	SARS-CoV-2	M		169	224	ITVATSRTLSYYKLGASQRVAGDSGFAAYSRYRIGNYKLNTDHSSSSDNIALLVQ*
7	SARS-CoV-2	N		1	56	MSDNGPQNQRNAPRITFGGPSDSTGSNQNGERSGARSKQRRPQGLPNNTASWFTAL
				29	84	NGERSGARSKQRRPQGLPNNTASWFTALTQHGKEDLKFPRGQGVPINTNSSPDDQI
8	SARS-CoV-2	N		85	140	GYYRRATRRIRGGDGKMKDLSPRWYFYYLGTGPEAGLPYGANKDGIIWVATEGALN
9	SARS-CoV-2	N		141	196	TPKDHIGTRNPANNAAIVLQLPQGTTLPKGFYAEGSRGGSQASSRSSSRSRNSSRN
				141	196	TPKDHIGTRNPANNAAIVLQLPQGTTLPKGFYAEGSRGGSQASSRSSSRSRNSLRN
				169	224	KGFYAEGSRGGSQASSRSSSRSRNSSRNSTPGSSRGTSPARMAGNGGDAALALLLL
				169	224	KGFYAEGSRGGSQASSRSSSRSRNSLRNSTPGSSRGTSPARMAGNGGDAALALLLL
				197	252	STPGSNRGTSPARMAGNGGDAALALLLLDRLNQLESKMSGKGQQQQGQTVTKKSAA
				197	252	STPGSSRGTSPARMAGNGGDAALALLLLDRLNQLESKMSGKGQQQQGQTVTKKSAA
				225	280	DRLNQLESKMSGKGQQQQGQTVTKKSAAEASKKPRQKRTATKAYNVTQAFGRRGPE
10	SARS-CoV-2	N		337	392	IKLDDKDSNFKDQVILLNKHIDAYKTFPPTEPKKDKKKKADETQALPQRQKKQQTV
				337	392	IKLDDKDPNFKDQVILLNKHIDAYKTFPPTEPKKDKKKKADETQALPQRQKKQQTV
				365	420	PTEPKKDKKKKADETQALPQRQKKQQTVTLLPAADLDDFSKQLQQSMSSADSTQA*
11	OC43	S	CS	729	784	NSTAISVQTCDLTVGSGYCVDYSKNRRSRGAITTGYRFTNFEPFTVNSVNDSLEPV
				757	812	QRQRGHKNGQGENDNISVAVPKSRVQQNKSRELTAEDISLLKKMDEPYTEDTSEI*
12	OC43	S	FP	869	924	PQRQRGHKNGQGENDNISVAVPKSRVQQNKSRELTAEDISLLKKMDEPYTEDTSEI
				897	952	RGAITTGYRFTNFEPFTVNSVNDSLEPVGGLYEIQIPSEFTIGNMVEFIQTSSPKV
13	OC43	S	HR2	1205	1260	VVVMSTCAVNYTKAPYVMLNTSIPNLPDFKEELDQWFKNQTSVAPDLSLDYINVTF
				1233	1288	CSKASSRSAIEDLLFDKVKLSDVGFVEAYNNCTGGAEIRDLICVQSYKGIKVLPPL
14	OC43	N		393	448	LSTKLKDGVNFNVDDINFSPVLGCLGSECSKASSRSAIEDLLFDKVKLSDVGFVEA
				394	449	FKEELDQWFKNQTSVAPDLSLDYINVTFLDLQVEMNRLQEAIKVLNQSYINLKDIG
15	HKU1	S	RBD	617	672	GVCVNYDLYGITGQGIFKEVSAAYYNNWQNLLYDSNGNIIGFKDFLTNKTYTILPC
16	HKU1	S	CS	729	784	YSVSSCDLRMGSGFCIDYALPSSRRKRRGISSPYRFVTFEPFNVSFVNDSVETVGG
				757	812	GISSPYRFVTFEPFNVSFVNDSVETVGGLFEIQIPTNFTIAGHEEFIQTSSPKVTI
17	HKU1	S	FP	869	924	SNLNTNLHSDVDNIDFKSLLGCLGSQCGSSSRSLLEDLLFNKVKLSDVGFVEAYNN
				897	952	SSSRSLLEDLLFNKVKLSDVGFVEAYNNCTGGSEIRDLLCVQSFNGIKVLPPILSE
18	HKU1	S	HR2	1149	1204	KPTSFKTVLVSPGLCLSGDRGIAPKQGYFIKQNDSWMFTGSSYYYPEPISDKNVVF
19	NL63	S	FP	841	896	SLANVTSFGDYNLSSVLPQRNIHSSRIAGRSALEDLLFSKVVTSGLGTVDVDYKSC
				869	924	GRSALEDLLFSKVVTSGLGTVDVDYKSCTKGLSIADLACAQYYNGIMVLPGVADAE
20	NL63	S	HR1	1009	1064	IALNKIQDVVNQQGSALNHLTSQLRHNFQAISNSIQAIYDRLDSIQADQQVDRLIT
21	NL63	N		29	84	SDKAPYRVIPRNLVPIGKGNKDEQIGYWNVQERWRMRRGQRVDLPPKVHFYYLGTG
				57	112	NVQERWRMRRGQRVDLPPKVHFYYLGTGPHKDLKFRQRSDGVVWVAKEGAKTVNTS
22	NL63	N		141	196	EDRSNNSSRASSRSSTRNNSRDSSRSTSRQQSRTRSDSNQSSSDLVAAVTLALKNL
				169	224	RQQSRTRSDSNQSSSDLVAAVTLALKNLGFDNQSKSPSSSGTSTPKKPNKPLSQPR
23	NL63	N		309	364	KMLVAKDNKNLPKFIEQISAFTKPSSIKEMQSQSSHAVQNTVLNASIPESKPLADD
				323	378	IEQISAFTKPSSIKEMQSQSSHAVQNTVLNASIPESKPLADDDSAIIEIVNEVLH*
24	229E	S	FP	645	700	SADVSEMLTFDKKAFTLANVSSFGDYNLSSVIPSLPTSGSRVAGRSAIEDILFSKL
				673	728	SSVIPSLPTSGSRVAGRSAIEDILFSKLVTSGLGTVDADYKKCTKGLSIADLACAQ
25	229E	N		57	112	YWNVQKRFRTRKGKRVDLSPKLHFYYLGTGPHKDAKFRERVEGVVWVAVDGAKTEP
26	229E	N		169	224	NPSSDRNHNSQDDIMKAVAAALKSLGFDKPQEKDKKSAKTGTPKPSRNQSPASSQT
27	229E	N		337	392	GKFLEELNAFTREMQQHPLLNPSALEFNPSQTSPATAEPVRDEVSIETDIIDEVN*

Table S1. Amino acid sequences of immunodominant CoV2 and HCoV peptides

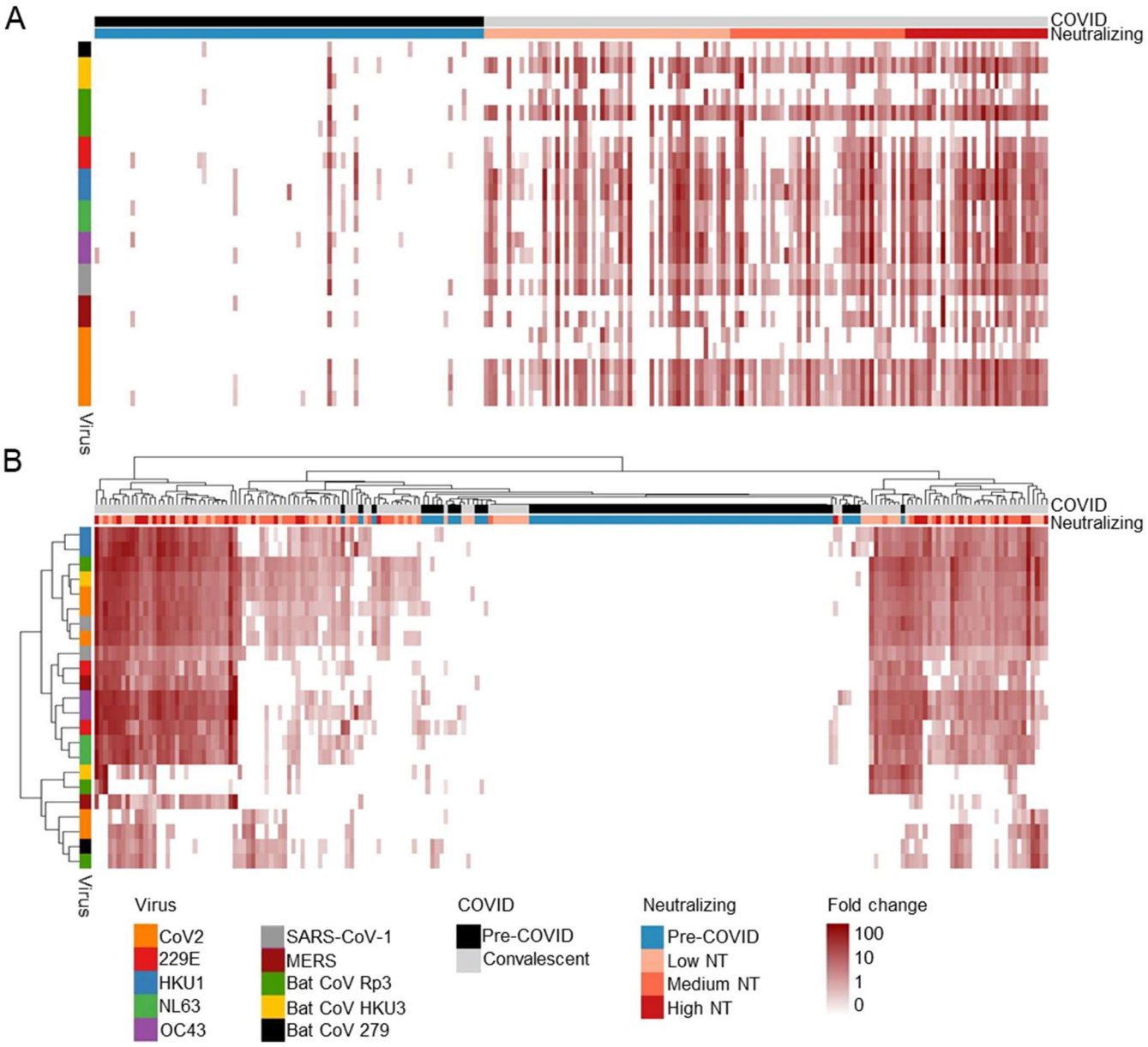

Figure S4. Pan-coronavirus fusion peptide antibody reactivity
Antibody binding of all study samples to all CoV FP peptides are shown in the form of a heatmap ordered by virus and sample group (**A**) and as a clustered heatmap (**B**). The FP of all CoVs represented in the VirScan library showed sequence homology to the dominant CoV2 FP peptides; reactivity was detected against every CoV FP. Pre-COVID: n=87, Low NT: n=55, Medium NT: n=39, and High NT: n=32.

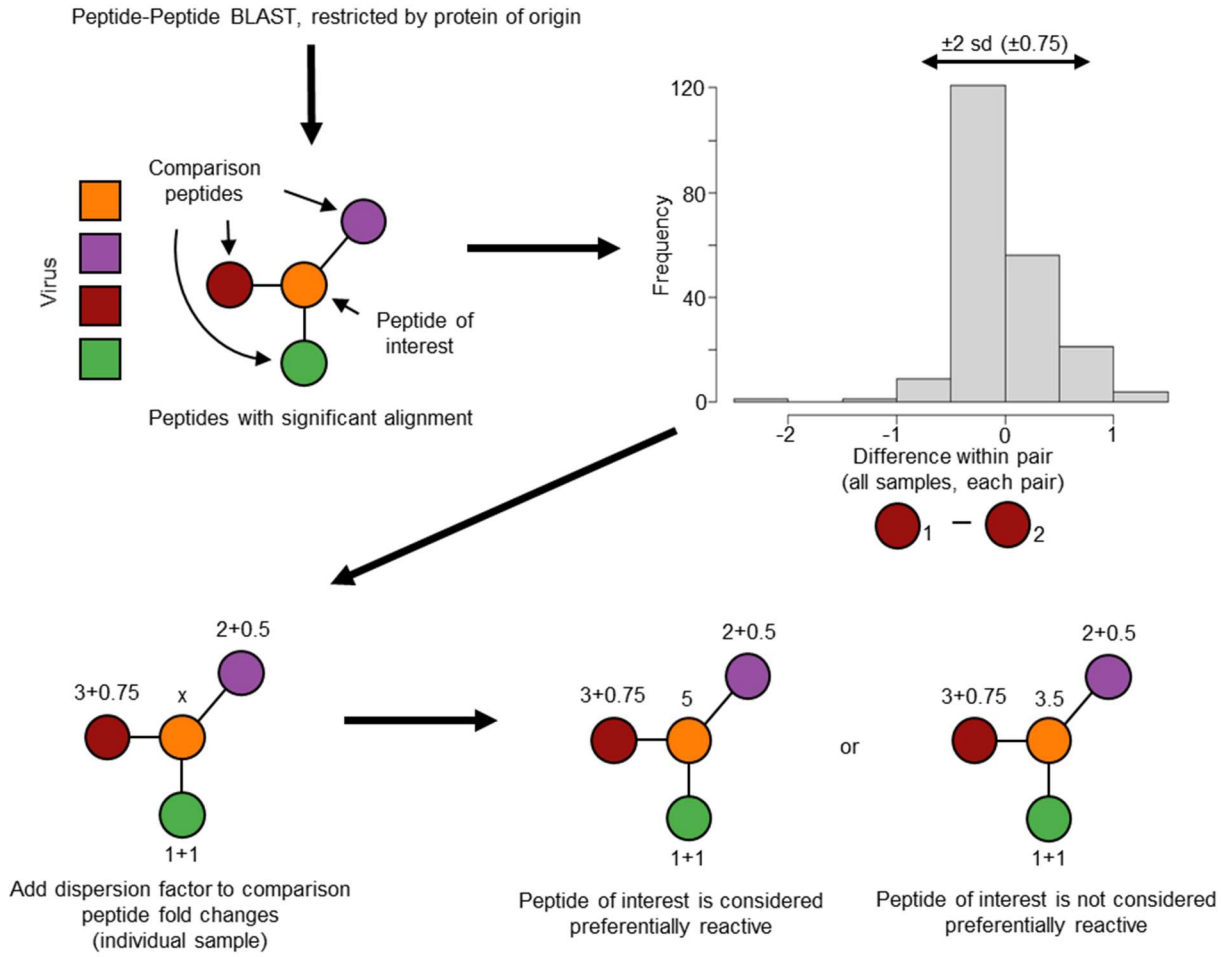

Figure S5. Deconvolution algorithm schematic
Peptides from different viruses of origin and the same protein of origin underwent peptide-peptide blastp. If peptides showed significant alignment (evalue<100), they were considered potentially cross-reactive. By taking advantage of the duplicate representation of each peptide in the library, a measure of expected technical dispersion was calculated. If a target peptide displayed enrichment greater than any comparison peptide plus the factor for technical dispersion (2 standard deviations), it was considered a target-preferred reactivity.

	Virus	Segment	Domain (Spike Only)	Start	End	Percent Samples Enriched				Association with Phenotype (-log(p))			
						Pre-COVID n=87	Low NT n=55	Medium NT n=39	High NT n=32	NT AUC	ADCP	ADCC	ADCD
1	SARS-CoV-2	ORF1		281	336	8	7.3	10.3	9.4	0.3	0.2	0.3	0.2
2	SARS-CoV-2	S	RBD	533	588	1.1	1.8	10.3	18.8	2.6	4.4	4.7	3.7
3	SARS-CoV-2	S	CS	617	672	1.1	0	5.1	25	3.5	4.5	3.6	1
4	SARS-CoV-2	S	FP	757	812	0	9.1	7.7	9.4	0	0.3	0.4	0.2
				757	812	0	20	17.9	34.4	1	2.4	2.3	3.1
				785	840	1.1	0	2.6	6.3	1	0.7	1	1.2
				785	840	0	1.8	10.3	9.4	1.4	1.4	0.7	1.6
				813	868	0	0	5.1	3.1	0.6	1.1	0.5	0.1
5	SARS-CoV-2	S	HR2	1121	1176	0	20	23.1	40.6	1.4	2.7	1	1.8
				1149	1204	1.1	5.5	0	0	1.1	0.5	0.2	0.2
6	SARS-CoV-2	M		169	224	0	7.3	20.5	50	4.7	5.8	6.7	1.5
7	SARS-CoV-2	N		1	56	1.1	9.1	17.9	50	4.8	4.7	6.4	3.5
				29	84	0	5.5	15.4	37.5	4.4	3.5	4.4	2.1
8	SARS-CoV-2	N		85	140	0	34.5	48.7	46.9	1.5	4.7	1.6	0.6
9	SARS-CoV-2	N		141	196	11.5	18.2	25.6	43.8	1.5	1	3.2	3.4
				141	196	14.9	20	30.8	59.4	2.5	1.9	3.2	3.3
				169	224	2.3	5.5	2.6	12.5	0.7	0.1	1.4	1
				169	224	5.7	3.6	5.1	18.8	1.5	1	1.3	2
				197	252	2.3	49.1	76.9	96.9	4.9	3.9	3.4	2.7
				197	252	3.4	47.3	64.1	93.8	7.1	6.8	3.7	2.4
				225	280	1.1	20	33.3	40.6	1.7	3	4.2	1.8
10	SARS-CoV-2	N		337	392	2.3	20	17.9	65.6	4.3	4.1	5.4	3.2
				337	392	2.3	16.4	17.9	56.3	4.6	5	5.9	3.1
				365	420	3.4	56.4	71.8	96.9	4.3	4.3	4.1	0.7
11	OC43	S	CS	729	784	14.9	7.3	7.7	9.4	0	0.1	0.3	0.2
				757	812	12.6	5.5	5.1	6.3	0.1	0.3	0	0.6
12	OC43	S	FP	869	924	0	0	0	6.3	1.3	0.5	0.3	0.5
				897	952	0	0	2.6	6.3	1.4	1.1	0.9	1
13	OC43	S	HR2	1205	1260	1.1	0	0	0	0	0	0	0
				1233	1288	1.1	9.1	10.3	6.3	0	0	0.3	0.5
14	OC43	N		393	448	20.7	9.1	15.4	9.4	0.2	0	0	0.4
				394	449	19.5	10.9	17.9	9.4	0.2	0	0.2	0.1
15	HKU1	S	RBD	617	672	28.7	50.9	35.9	40.6	1	1	0.6	0.1
16	HKU1	S	CS	729	784	1.1	7.3	2.6	6.3	0.1	0	0	0
				757	812	8	10.9	2.6	6.3	0.7	0.2	0.3	0.1
17	HKU1	S	FP	869	924	1.1	3.6	0	6.3	0.2	0.3	0.3	0.2
				897	952	0	12.7	2.6	12.5	0	0.5	1.3	0.6
18	HKU1	S	HR2	1149	1204	0	20	23.1	43.8	1.6	2.4	3.1	4.5
19	NL63	S	FP	841	896	0	0	0	3.1	0.7	0.9	0.8	0.6
				869	924	0	0	0	0	0	0	0	0
20	NL63	S	HR1	1009	1064	0	5.5	5.1	18.8	1.3	0.3	0.3	0.3
21	NL63	N		29	84	13.8	5.5	2.6	0	1.3	1.2	0	0.6
				57	112	16.1	7.3	12.8	9.4	0.1	0.6	0.3	0.8
22	NL63	N		141	196	1.1	3.6	2.6	12.5	0.8	0.3	1.1	0.8
				169	224	17.2	9.1	15.4	15.6	0.7	0.1	0.1	0.2
23	NL63	N		309	364	8	3.6	7.7	3.1	0.2	0.2	0.1	0
				323	378	14.9	9.1	15.4	12.5	0	0.2	0.1	0.4
24	229E	S	FP	645	700	0	0	0	0	0	0	0	0
				673	728	0	1.8	0	0	0.2	0.2	1	0.5
25	229E	N		57	112	5.7	10.9	5.1	0	1.8	1.1	1.2	0.5
26	229E	N		169	224	5.7	5.5	5.1	0	0.5	0.3	0.5	0.3
27	229E	N		337	392	4.6	10.9	15.4	9.4	0.4	0.1	0.2	0.4

Table S2. Deconvoluted immunodominant coronavirus peptides and their functional correlates
The frequency of enrichment of 52 immunodominant peptides among each sample group is shown post deconvolution. The percentage of samples with a specific reactivity is shown (red shading). Associations with COVID-19 convalescent plasma functionality were defined by dichotomizing all convalescent plasma by presence or absence of each particular reactivity followed by a two sided Wilcox test. The negative log transformed p values are shown (green shading).

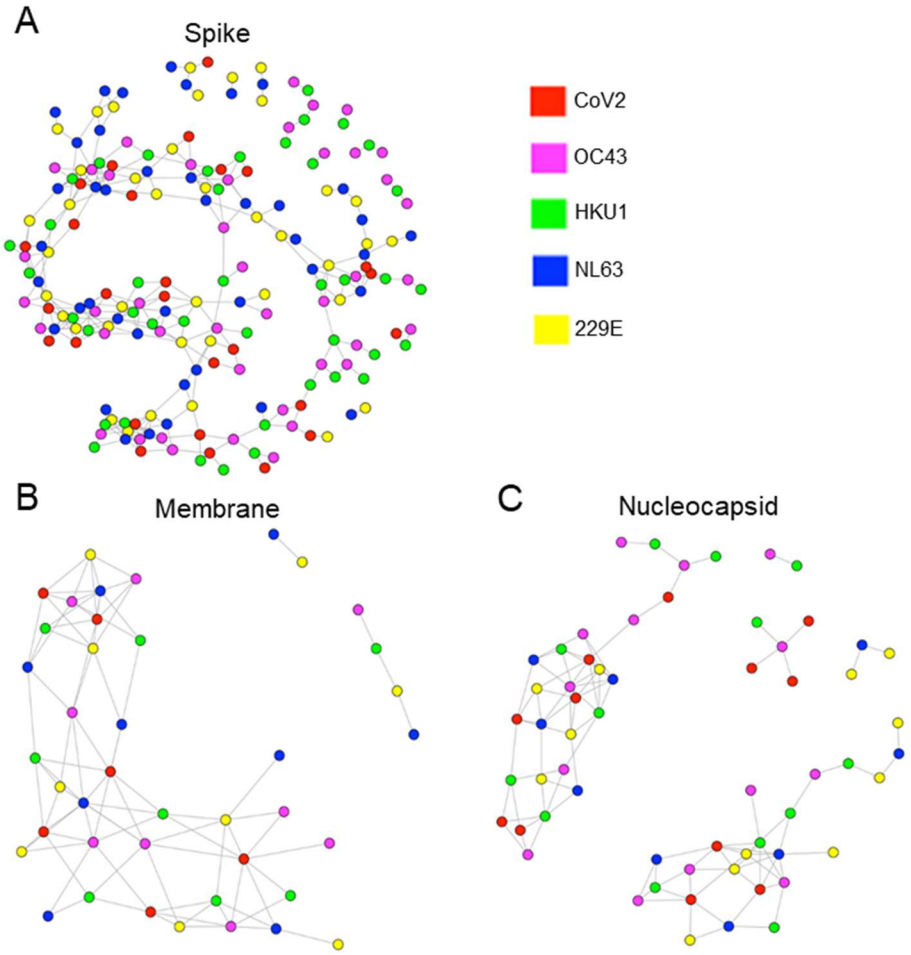

Figure S6. Visualization of CoV2 and HCoV peptide sequence homologies
Network graphs show sequence homologies among the spike (**A**)**,** membrane (**B**) and nucleocapsid (**C**) peptides between CoV2 and each HCoV. Nodes (peptides) are colored by their corresponding virus. Peptides are linked by an edge if they share blastp sequence similarity. Only homologies among peptides from different viruses are shown for simplicity.