

**Table S1:** Binding Kinetics of the interaction between SARS-CoV-2 RBD and sACE2 quantified by Biolayer Interferometry.

RBD	ACE2 binding (RBD/biolayer interferometry)					
	KD (nM)	Fold change	Ka (1/Ms)	Fold change	Kdis (1/s)	Fold change
WT	161	1	$4.27 \times 10^4$	1	$6.88 \times 10^{-3}$	1
N501Y	41.5	3.88	$3.59 \times 10^4$	0.84	$1.49 \times 10^{-3}$	0.22
K417N	215	0.75	$6.65 \times 10^4$	1.56	$1.43 \times 10^{-2}$	2.07
K417T	245	0.66	$4.76 \times 10^4$	1.11	$1.17 \times 10^{-2}$	1.70
E484K	132	1.22	$5.53 \times 10^4$	1.29	$7.31 \times 10^{-3}$	1.06
L452R	176	0.91	$2.74 \times 10^4$	0.64	$4.84 \times 10^{-3}$	0.70

Table S2. Summary of ACE2 Binding, Plasma binding, and Neutralization to Spike variants.

Variant / mutant	ACE2 binding <sup>a</sup>	Spike recognition by plasma <sup>b</sup>			Neutralization <sup>c</sup> ID50 (ratio to D614G)
		Previously infected (8 months PSO)	Previously infected Vaccinated (3 weeks post-vaccination)	Naïve vaccinated (3 weeks post-vaccination)	
	D614G	1.00	1.00	1.00	1.00
B.1.1.7	Full variant	5.43	1.15	1.00	0.78
	D614G/Δ69-70	1.51	1.33	1.14	0.92
	D614G/Δ144	0.95	1.14	0.93	0.90
	D614G/N501Y	2.52	1.65	1.17	1.09
	D614G/A570D	1.18	1.29	1.01	1.04
	D614G/P681H	0.81	1.26	0.88	0.90
	D614G/T716I	0.89	1.58	0.98	0.68
	D614G/S982A	0.77	1.06	0.50	0.89
	D614G/D1118H	1.23	1.96	1.09	0.93
	Full variant	3.56	0.96	1.28	1.17
B.1.351	D614G/L18F	0.93	1.66	1.30	0.79
	D614G/D80A	1.00	2.11	1.64	0.74
	D614G/D215G	1.19	2.29	1.79	0.77
	D614G/Δ242-244	0.74	1.97	1.43	0.73
	D614G/R246I	0.66	1.53	1.05	0.69
	D614G/K417N	0.13	2.16	1.76	0.57
	D614G/E484K	0.82	1.42	1.16	1.06
	D614G/N501Y	2.52	1.65	1.17	1.09
	D614G/A701V	0.99	0.90	0.84	0.91
	Full variant	4.24	0.77	0.88	0.79
P.1	D614G/L18F	0.93	1.66	1.30	0.79
	D614G/T20N	1.96	1.65	1.29	0.72
	D614G/P26S	1.58	0.93	0.81	0.73
	D614G/D138Y	1.32	0.64	0.73	0.64
	D614G/R190S	1.82	1.09	0.84	0.63
	D614G/K417T	0.78	1.10	1.04	0.91
	D614G/E484K	0.85	1.42	1.16	1.06
	D614G/N501Y	2.52	1.65	1.17	1.09
	D614G/H655Y	1.21	1.48	1.04	0.92
	D614G/T1027I	0.59	1.56	0.76	0.82
B.1.526	Full variant	1.78	1.02	1.34	1.19
	D614G/L5F	0.91	1.00	1.23	0.91
	D614G/T95I	0.75	1.00	1.16	0.98
	D614G/D253G	0.93	0.84	1.01	0.84
	D614G/E484K	0.85	1.42	1.16	1.06
	D614G/A701V	0.99	0.90	0.84	0.91
B.1.429	Full variant	2.82	0.91	0.90	0.89
	D614G/S13I	1.42	0.84	0.93	0.79
	D614G/W152C	1.33	0.87	0.97	0.76
	D614G/L452R	2.66	2.15	0.95	0.86
B.1.617	Full variant	1.85	0.82	0.77	0.85
B.1.617.1	Full variant	1.12	0.50	0.56	0.61
B.1.617.2	Full variant	2.34	0.52	0.61	0.53
					601 (0.16)

- a) ACE2-Fc binding was normalized to CV3-25 binding in each experiment. Values are presented as ratio of normalized ACE2-Fc binding obtain with the D614G Spike. Values represent the means of data obtained from at least three independent experiments.
- b) Plasma binding were normalized to CV3-25 in each experiment. Values are presented as ratio of normalized plasma binding obtained with the D614G Spike. Values represent the means of data obtained with 3-5 plasma from the same group.
- c) The ID50 represents the plasma dilution to inhibit 50% of the infection of 293T-ACE2 cells by recombinant viruses bearing the indicated Spike. Values are presented as the means of ID50 and as ratio of the ID50 obtained with virus bearing the D614G Spike. Values represent the means of data obtained with 3-5 different plasma.

**Table S3. List of single mutations of Spike Variants.**

<b>B.1.1.7</b>	<b>B.1.351</b>	<b>P.1</b>	<b>B.1.526</b>	<b>B.1.429</b>	<b>B.1.617</b>	<b>B.1.617.1</b>	<b>B.1.617.2</b>
Δ69-70	L18F	L18F	L5F	S13I	L452R	E154K	T19R
Δ144	D80A	T20N	T95I	W152C	E484Q	L452R	Δ156-158
N501Y	D215G	P26S	D253G	L452R	D614G	E484Q	L452R
A570D	Δ242-244	D138Y	E484K	D614G		D614G	T478K
D614G	R246I	R190S	D614G			P681R	D614G
P681H	K417N	K417T	A701V				P681R
T716I	E484K	E484K					D950N
S982A	N501Y	N501Y					
D1118H	D614G	D614G					
	A701V	H655Y					
		T1027I					

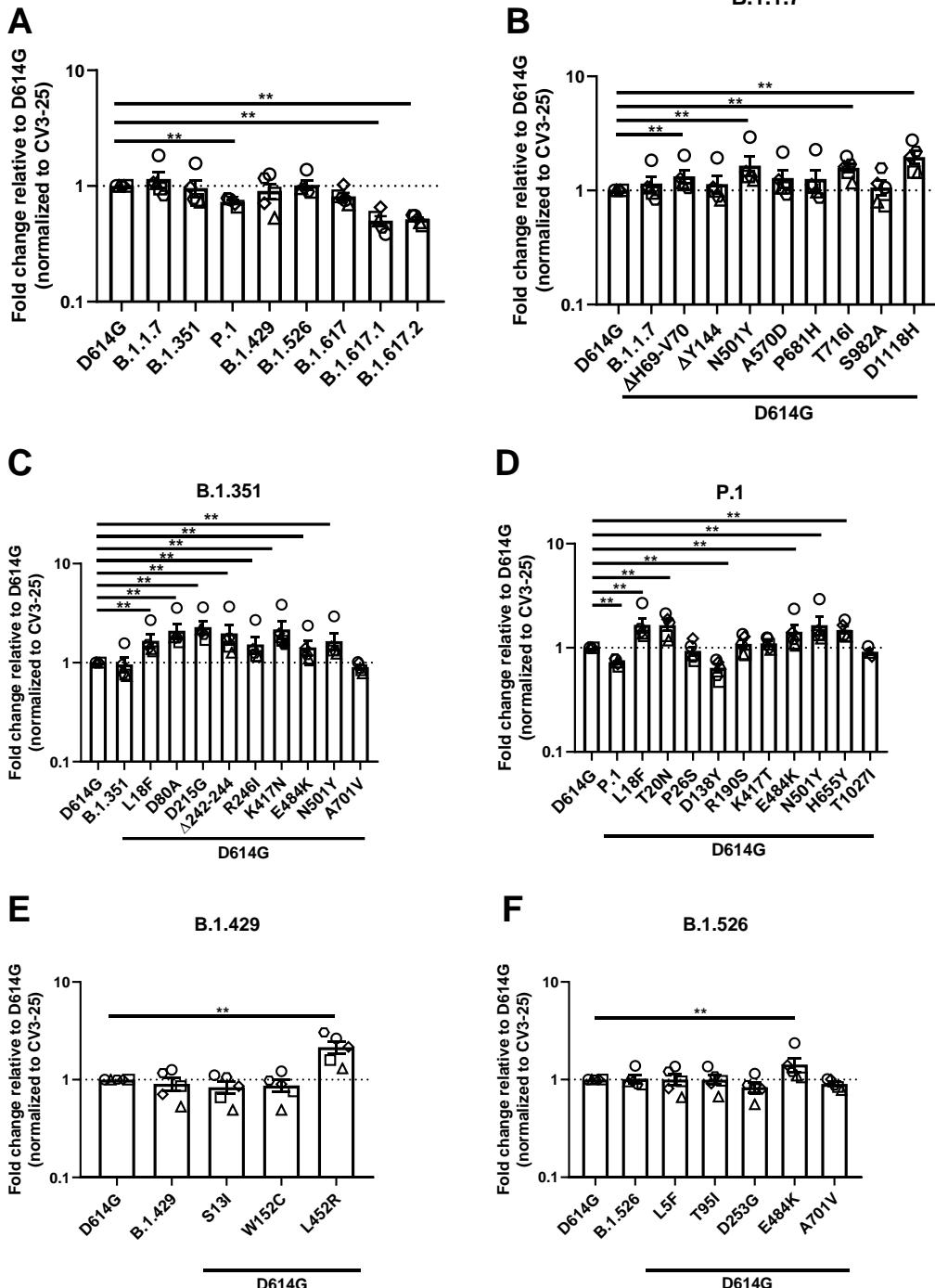


Figure S1

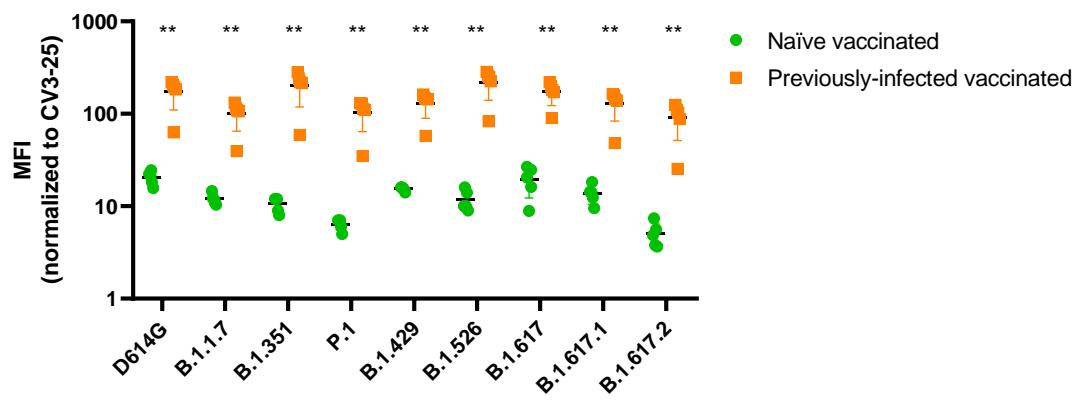


Figure S2