

Supplemental information

Delta-Omicron recombinant escapes

therapeutic antibody neutralization

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Supplemental Information

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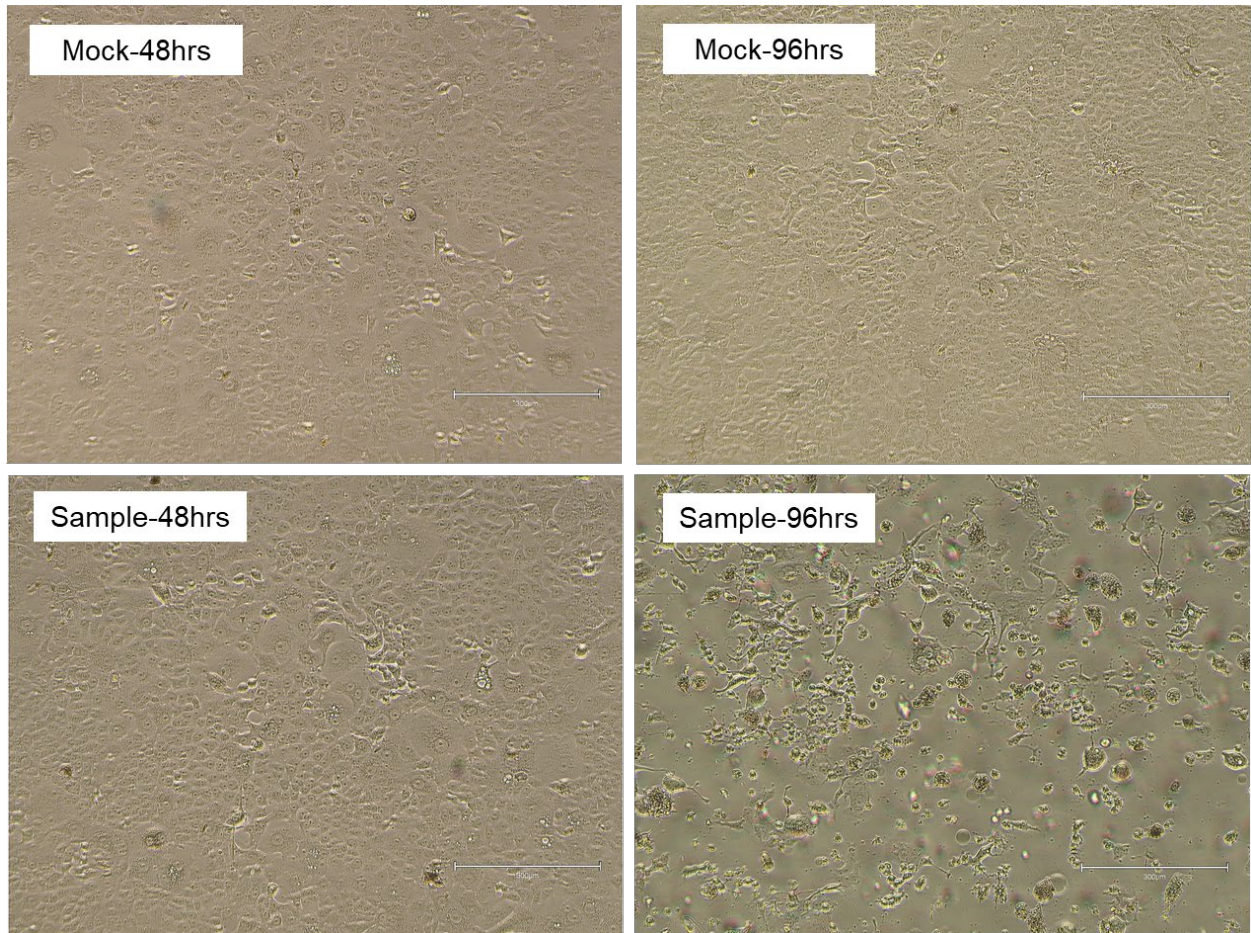


Figure S1. Cytopathic effect of the SARS-CoV-2 Delta-Omicron recombinant on VeroE6/TMPRSS2 cells, related to **Figure 1**.

A residual nasopharyngeal swab specimen of the patient infected with the recombinant variant was inoculated onto VeroE6/TMPRSS2 cells and observed daily for cytopathic effects (bottom row). The image on the lower right (Sample-96hrs) is the same as the left image in **Figure 1D** (Delta-Omicron recombinant). Images were taken with an EVOS M5000 inverted microscope (ThermoFisher Scientific, Waltham, MA); 10X magnification.

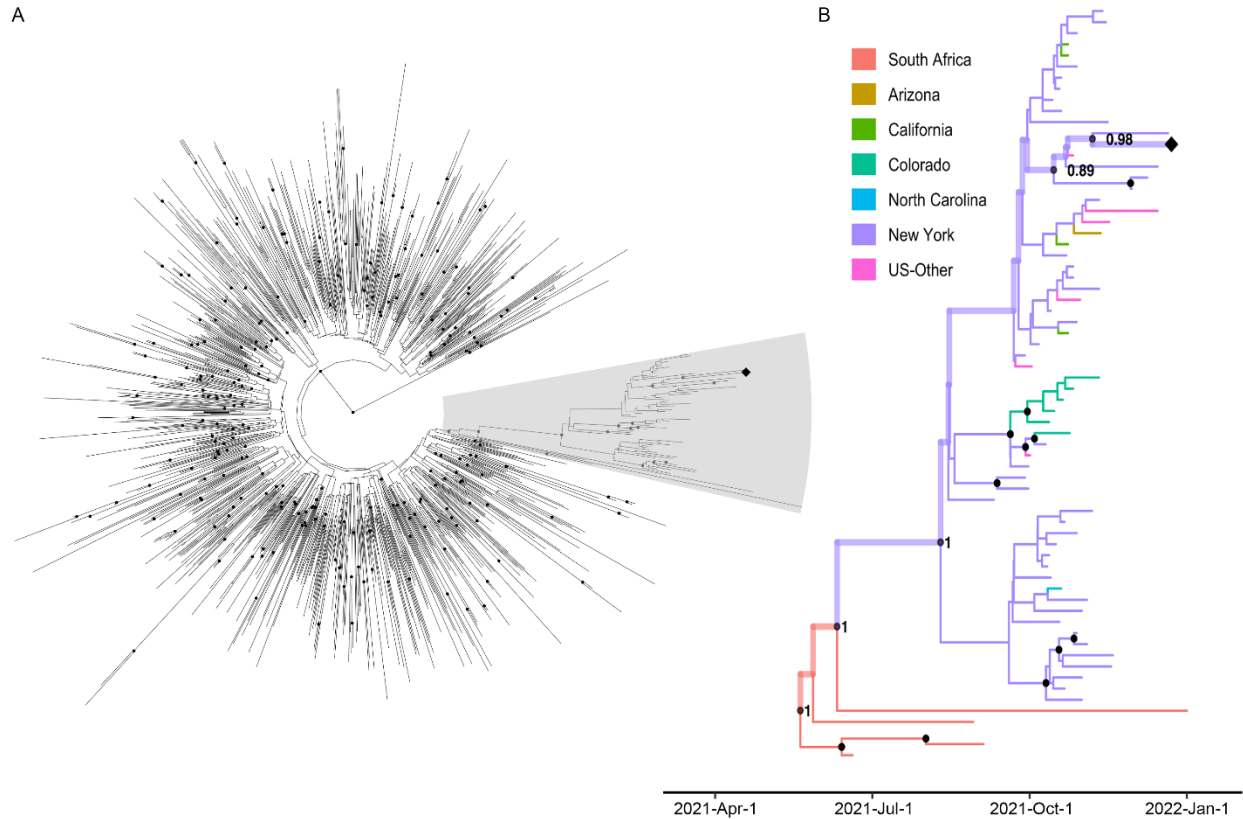


Figure S2. Phylogeographic analysis of the AY.45 cluster containing the recombinant, related to **Figure 1**.

(A) Maximum clade credibility tree of 1122 AY.45 genomes. The recombinant genome (AY.45 segment of it) is highlighted with a diamond symbol. Nodes with a posterior support above 70% are denoted with a black circle. A well supported phylogenetic cluster containing the recombinant genome is shaded in gray.

(B) Phylogeographic analysis of the phylogenetic cluster containing the AY.45 segment of the recombinant sequence. The recombinant is annotated with a diamond symbol, and branches corresponding to the Markov jump trajectory plot (**Figure 1C**) are highlighted (by thicker branches). Internal nodes with posterior support of 70% or higher are denoted with a black circle, whereas nodes with 100% support are annotated explicitly.

Table S1. Summary of mutations detected in the Delta-Omicron recombinant by four sequencing methods on different sample aliquots, with or without in vitro culture, related to **Figure 1**.

gene	nucleotide	amino acid	donor	xGen	xGen (repeat)	shotgun	AmpliSeq	harvest	24h	48h	72h	96h
ORF1ab	C1122T	P106L	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	G4181T	A488S	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C5526T	T936I	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C6402T	P1228L	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C7124T	P1469S	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	G9053T	V167L	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	G9335A	V261I	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C10029T	T492I	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C11001T	T10I	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	A11201G	T77A	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	G13812T	M124I	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C14408T	P323L	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C15371T	T644M	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	G15451A	G671S	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C16466T	P77L	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	A17888G	E551G	Delta	X	X	X	X	X	X	X	X	X
ORF1ab	C19220T	A394V	Delta	X	X	X	X	X	X	X	X	X
Spike	C21618G	T19R	Delta	X	X	X	X	X	X	X	X	X
Spike	G21987A	G142D	Delta	X	X	X	X	X	X	X	X	X
Spike	GAGTTCA22028G	EFR156-158G deletion	Delta	X	X	X	X	X	X	X	X	X
Spike	AATT22193A	NL211-212I deletion	Omicron	X	X	X	X	X	X	X	X	X
Spike	T22204TGAGCCAGAA	214EPE insertion	Omicron	--	--	X	X	X	X	X	X	X
Spike	G22578A	G339D	Omicron	X	X	X	X	X	X	X	X	X
Spike	A22582C	E340D	Omicron	X	X	X	X	X	X	X	X	X
Spike	T22673C	S371P	Omicron	X	X	X	X	X	X	X	X	X
Spike	C22674T	S371F	Omicron	X	X	X	X	X	X	X	X	X
Spike	T22679C	S373P	Omicron	X	X	X	X	X	X	X	X	X
Spike	C22686T	S375F	Omicron	X	X	X	X	X	X	X	X	X
Spike	G22813T	K417N	Omicron	X	X	X	X	X ^a	N	N	X ^a	X ^a
Spike	T22882G	N440K	Omicron	X	X	X	X	X ^a	N	N	N	X ^a
Spike	G22898A	G446S	Omicron	X	X	X	X	X ^a	N	N	N	X ^a
Spike	G22992A	S477N	Omicron	X	X	X	X	X	X	X	X	X

Spike	C22995A	T478K	Omicron	X	X	X	X	X	X	X	X	X
Spike	A23013C	E484A	Omicron	X	X	X	X	X	X	X	X	X
Spike	A23040G	Q493R	Omicron	X	X	X	X	X	X	X	X	X
Spike	G23048A	G496S	Omicron	X	X	X	X	X	X	X	X	X
Spike	A23055G	Q498R	Omicron	X	X	X	N	X	X	X	X	X
Spike	A23063T	N501Y	Omicron	X	X	X	N	X	X	X	X	X
Spike	T23075C	Y505H	Omicron	X	X	X	X ^a	X	X	X	X	X
Spike	C23202A	T547K	Omicron	X	X	X	X	X	X	X	X	X
Spike	A23403G	D614G	Omicron	X	X	X	X	X	X	X	X	X
Spike	C23525T	H655Y	Omicron	X	X	X	X	X	X	X	X	X
Spike	T23599G	N679K	Omicron	X	X	X	X	X	X	X	X	X
Spike	C23604A	P681H	Omicron	X	X	X	X	X	X	X	X	X
Spike	C23854A	N764K	Omicron	X	X	X	X	X	X ^a	X	X	X
Spike	G23948T	D796Y	Omicron	X	X	X	X	X	X	X	X	X
Spike	C24130A	N856K	Omicron	X	X	X	X	X	X	X	X	X
Spike	A24424T	Q954H	Omicron	X	X	X	X	X	X	X	X	X
Spike	T24469A	N969K	Omicron	X	X	X	X	X	X	X	X	X
Spike	C24503T	L981F	Omicron	X	X	X	X	X	X	X	X	X
ORF3a	T25577C	I62T	Omicron	X	X	X	X	X	X	X	X	X
E	C26270T	T9I	Omicron	X	X	X	X	X	X	X	X	X
M	A26530G	D3G	Omicron	X	X	X	X	X	X	X	X	X
M	C26577G	Q19E	Omicron	X	X	X	X	X	X	X	X	X
M	G26709A	A63T	Omicron	X	X	X	X	X	X	X	X	X
N	C28311T	P13L	Omicron	X	X	X	X	X	X	X	X	X
N	GGAGAACGCA28361G	ERS31-33 deletion	Omicron	X	X	X	X	X	X	X	X	X
N	G28881A	R203K	Omicron	X	X	X	X	X	X	X	X	X
N	G28883C	G204R	Omicron	X	X	X	X	X	X	X	X	X

^a: low sequencing depth (<40%);

AmpliSeq: AmpliSeq Insight sequencing (performed at NY State DOH);

harvest: ARTIC NGS amplicon sequencing using cells and supernatant after 96h virus culture;

shotgun: metagenomics shotgun sequencing (performed at NYU);

N: not covered;

X: mutation is present;

xGen: IDT xGen NGS amplicon sequencing (performed at NYU);

24h-96h: ARTIC NGS amplicon sequencing using supernatants after the indicated culture time points (performed at NY State DOH);

--: no coverage

Table S2. Real-time RT-PCR results and infectious virus titers during culture of the SARS-CoV-2 Delta-Omicron recombinant in VeroE6/TMPRSS2 cells, related to **Figure 1**.

Sample	Ct value (N1)	Virus titer (log ₁₀ PFU/ml)
NPS specimen	30.76	ND
24 hpi	30.55	2.67
48 hpi	23.10	4.66
72 hpi	16.77	6.97
96 hpi	13.50	6.67

Abbreviations: Ct: cycle threshold value, hpi: hours post infection, ND=not done, NPS: naso-pharyngeal swab, PFU: plaque-forming units.

Table S3. cDNA gene synthesis for the generation of the SARS-CoV-2 Delta-Omicron spike plasmid, and primers for the insertion of point mutations into plasmids for pseudotyped virus generation, related to **Figures 3-5**.

synthesized cDNA gene for Delta-Omicron spike (Delta segment to be fused to BA.1)	
KpnI site	GGTACC
Kozak	gccacc
gene	ATGTTTCGTGTTTCTGGTGCTGCTGCCTCTGGT GTCCAGCCAGTGTGTGAACCTGAGGACCAGA ACACAGCTGCCTCCAGCCTACACCAACAGCTT TACCAGAGGCGTGTACTACCCCGACAAGGTG TTCAGATCCAGCGTGCTGCACTCTACCCAGGA CCTGTTCTGCCTTTCTTCAGCAACGTGACCT GGTTCCACGCCATCCACGTGTCCGGCACCAAT GGCACCAAGAGATTCGACAACCCCGTGCTGC CCTTCAACGACGGGGTGTACTTTGCCAGCACC GAGAAGTCCAACATCATCAGAGGCTGGATCTT CGGCACCACACTGGACAGCAAGACCCAGAGC CTGCTGATCGTGAACAACGCCACCAACGTGGT CATCAAAGTGTGCGAGTTCCAGTTCTGCAACG ACCCCTTCTGGACGTCTACTACCACAAGAAC AACAAGAGCTGGATGGAAAGCGGGGTGTACA GCAGCGCCAACAACACTGCACCTTCGAGTACGT GTCCCAGCCTTTCCTGATGGACCTGGAAGGCA AGCAGGGCAACTTCAAGAACCTGCGCGAGTT CGTGTTCAAGAACATCGACGGCTACTTCAAGA TCTACAGCAAGCACACCCCTATCATCGTGCGG GATCTGCCTCAGGGCTTCTCTGCTCTGGAACC

	CCTGGTGGATCTGCCATCGGCATCAACATCA CCCGGTTTCAGACACTGCTGGCCCTGCACAG AAGCTACCTGACACCTGGCGATAGCAGCAGC GGATGGACAGCTGGTGCCGCCGCTTACTATG TGGGCTACCTGCAGCCTAGAACCTTCCTGCTG AAGTACAACGAGAACGGCACCATCACCGACG CCGTGGATTGTGCTCTGGATCCTCTGAGCGAG ACAAAGTGCACCCTGAAGTCCTTCACCGTGGA AAAGGGCATCTACCAGACCAGCAACTTCCGG GTGCAGCCCACCGAATCCATCGTGCCGGTTCC CCAATATACCAATCTGTGCCCTTCGACGAC GTCTTTAACGCAACACGGTTCGCCTCAGTG
XhoI site	CTCGAG
Primer	
DO-Fusion-F	CACCAATCTGTGCCCTTCGACGACGTCTTTA ACGCAACACGGTTCGCCTCAGTG
DO-Fusion-R	CACTGAGGCGAACCGTGTTGCGTTAAAGACGT CGTCGAAGGGGCACAGATTGGTG
E340D-F	CACCAATCTGTGCCCTTCGGCGACGTGTTCA ATGCCACCAGATTC
E340D-R	GAATCTGGTGGCATTGAACACGTCGCCGAAG GGGCACAGATTGGTG
E340K-F	CACCAATCTGTGCCCTTCGGCAAGGTGTTCA ATGCCACCAGATTC
E340K-R	GAATCTGGTGGCATTGAACACCTTGCCGAAGG GGCACAGATTGGTG
E340A-F	CACCAATCTGTGCCCTTCGGCGCGGTGTTCA ATGCCACCAGATTC
E340A-R	GAATCTGGTGGCATTGAACACCGCGCCGAAG GGGCACAGATTGGTG
E340V-F	CACCAATCTGTGCCCTTCGGCGTGGTGTTC ATGCCACCAGATTC
E340V-R	GAATCTGGTGGCATTGAACACCACGCCGAAG GGGCACAGATTGGTG
P337L-F	GGTCCCAATATACCAATCTGTGCCTCTTC GGCGAGGTGTTCAATGCCACCAG
P337L-R	CTGGTGGCATTGAACACCTCGCCGAAGAGGC ACAGATTGGTGATATTGGGGAACC
BA.1-E340D-F	CATCACTAATTTGTGCCCTTCGACGA _c GTCTT TAACGCAACACGGTTCGCC
BA.1-E340D-R	GGCGAACCGTGTTGCGTTAAAGAC _g TCGTCTGA AGGGGCACAAATTAGTGATG
DO-D340E-F	TCACCAATCTGTGCCCTTCGACGA _a GTCTTTA ACGCAACACGGTTCGCC
DO-D340E-R	GGCGAACCGTGTTGCGTTAAAGAC _i TCGTCTGA AGGGGCACAGATTGGTGA

Table S4. IC50 of BNT162b2-elicited antibodies against viruses with variant spike proteins in sera collected from COVID-19-unexperienced and experienced donors, related to **Figure 5.**

BNT162b2, COVID-19-Unexperienced													
				D614G		Delta		Omicron BA.1		Omicron BA.2		Delta-Omicron	
Donor	Age	Sex	Comorbidities	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster
1	34	M	None	1250	4106	105	371	37.84	1027	15.27	326.5	24.54	621.9
2	29	F	None	572	3206	156.2	1851	14.11	824.8	4.605	238.9	21.51	363.7
3	37	F	Allergy	946	6359	386	3234	58.34	677.5	70.71	319.1	76.41	372.2
4	62	M	Hypertension, Hyperlipidemia	671.2	3789	1032	4024	85	1154	ND	428.8	39.5	681
5	52	F	Hypertesion	581.1	3275	265.4	7040	66.11	2221	149.1	141	58.47	33.25
6	34	M	Hypothyroidism	966.2	6301	433.1	6319	ND	527	100.6	54.87	105.7	74.84
7	38	F	Asthma, Anemia, Tinea versicolor	1060	5541	444.8	4045	2.097	1315	ND	724.6	ND	715.2
8	38	M	None	423.3	1882	373.2	8163	38.33	678.7	27.7	231.6	38.27	324.4
9	52	F	None	484.8	6286	514.4	12903	92.72	709.6	46.95	266	62.37	538
Mean (SD)	41 (9)			772	4527	412	5327	44	1014	46	303	47	413

BNT162b2, COVID-19-Experienced													
				D614G		Delta		Omicron BA.1		Omicron BA.2		Delta-Omicron	
Donor	Age	Sex	Comorbidities	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster	1 month post vax-2	1 month post booster
1	45	M	Asthma	5958	10443	5883	11917	169	1695	134.7	1456	337.7	1138
2	37	F	None	8016	8991	5506	8828	208	1389	309.9	516.5	512.6	883.6

3	54	F	Hypertension, Obesity	11024	10453	2128	7968	174.3	1012	349.5	729	208.9	512.9
4	54	M	Cardiovascular disease	3989	7811	1406	8946	223.5	1566	79.65	1455	130.5	530.1
5	25	F	None	7768	10831	2073	8626	109.2	329.1	234.3	690.9	110	438.6
6	42	F	Diabetes, Herpes simplex	10747	9331	3240	17502	345.1	1364	180.5	712.2	256	235.1
7	43	M	None	5974	13891	2064	15888	171.7	334.3	211.7	346.1	275.8	353.1
Mean (SD)	41 (9)			7639	10250	3185	11382	200	1098	214	843	261	584