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Supplemental information

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Supplementary Figures S1-S5
Supplementary Tables S1-S6

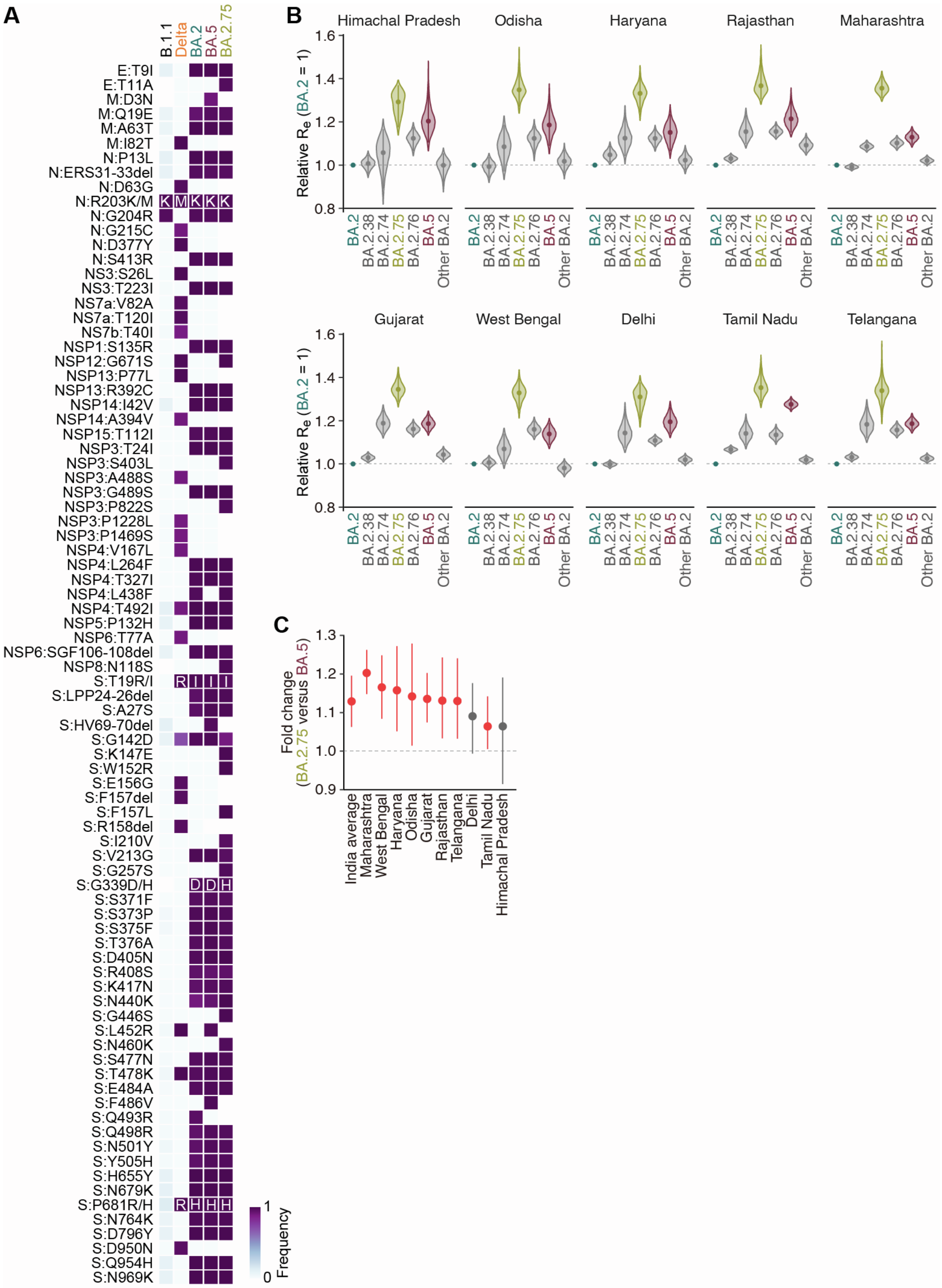

Figure S1. Epidemic dynamics of BA.2.75 in India, related to Figure 1

(**A**) Amino acid differences in B.1.1, Delta, BA.2, BA.5 and BA.2.75 compared to the SARS-CoV-2 A lineage. Heatmap color indicates the frequency of amino acid mutations.

(**B**) Estimated relative R_e of each viral lineage, assuming a fixed generation time of 2.1 days. The R_e value of BA.2 is set at 1. The posterior (violin), posterior mean (dot), and 95% CI (line) are shown. The R_e values for the respective Indian states are shown. The dynamics of the top seven predominant lineages in India were estimated. BA.5 sublineages are summarized as “BA.5”, and nonpredominant BA.2 sublineages are summarized as “other BA.2”. The raw data are summarized in **Table S1**.

(**C**) Fold change in R_e values between BA.2.75 and BA.5. The posterior mean (dot) and 95% CI (line) are shown. Red indicates that the 95% CI does not overlap with the value of 1.

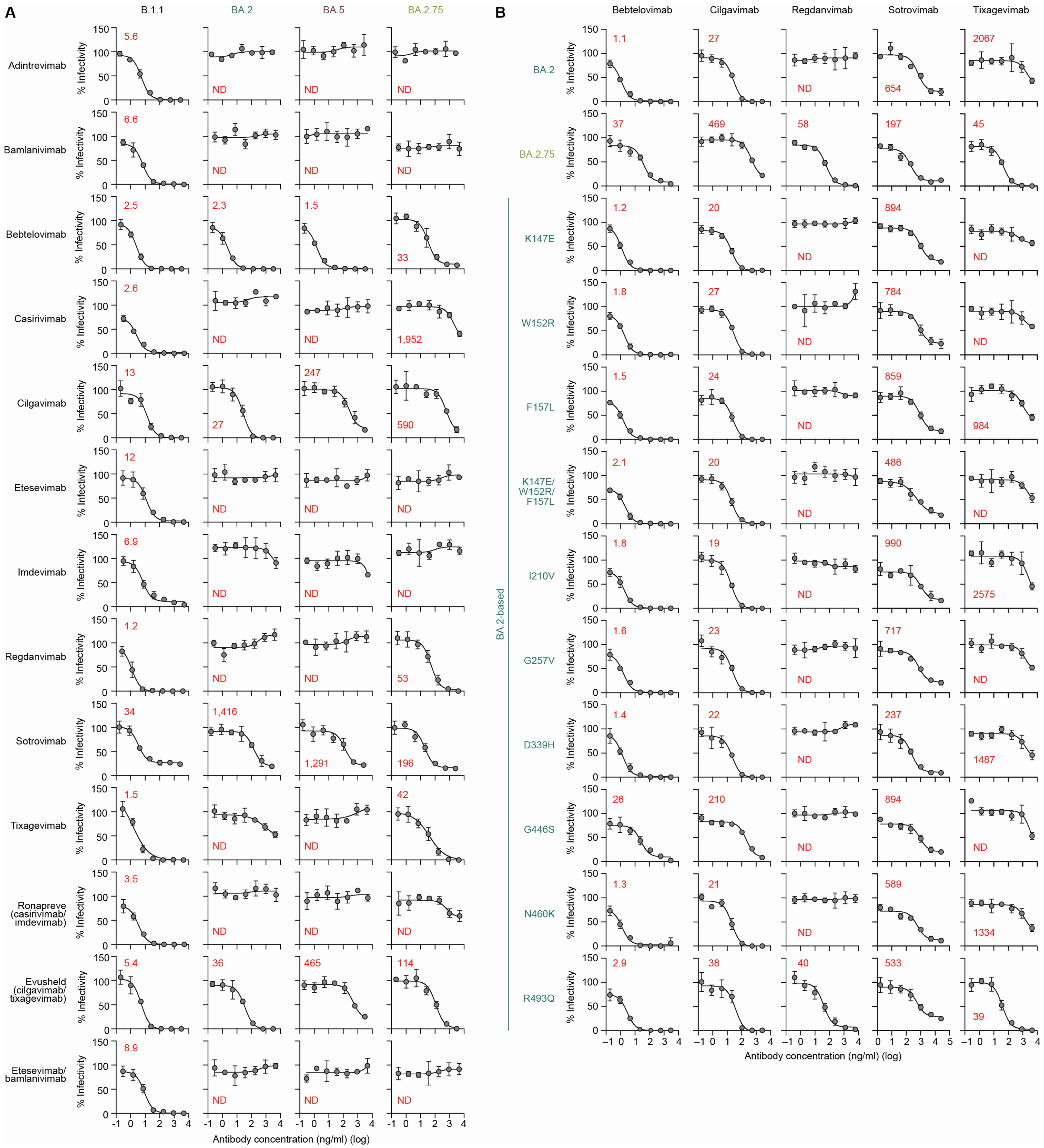

Figure S2. Representative neutralization curves, related to Table 2

A neutralization assay was performed using pseudoviruses harboring the SARS-CoV-2 S proteins of BA.2 and BA.5 and BA.2.75, the D614G-harboring B.1.1 lineage virus (B.1.1), and the BA.2 derivatives.

(**A**) Neutralization assay using pseudoviruses harboring the SARS-CoV-2 S proteins of BA.2, BA.5 and BA.2.75, the D614G-harboring B.1.1 lineage virus (B.1.1) with ten therapeutic monoclonal antibodies (adintrevimab, bamlanivimab, bebtelovimab, casirivimab, cilgavimab, etesevimab, imdevimab, regdanvimab, sotrovimab and tixagevimab) and three antibody cocktails [Ronapreve (casirivimab+imdevimab), Evusheld (cilgavimab+tixagevimab), and etesevimab+bamlanivimab]. A representative dataset is shown. The 50% inhibitory concentration (IC_{50}) value of each assay is summarized in **Table 1**.

(**B**) Neutralization assay using pseudoviruses harboring the SARS-CoV-2 S proteins of BA.2, BA.2.75, and the BA.2 derivatives with five therapeutic monoclonal antibodies (bebtelovimab, cilgavimab, regdanvimab, sotrovimab and tixagevimab). A representative dataset is shown. The IC_{50} value of each assay is summarized in **Table S3**.

The assay of each antibody was performed in triplicate at each concentration to determine the IC_{50} (ng/ml), and the assay was independently repeated four times. Representative neutralization curves of a neutralization assay are shown. The red numbers in the panels indicate the IC_{50} value. ND, not determined.

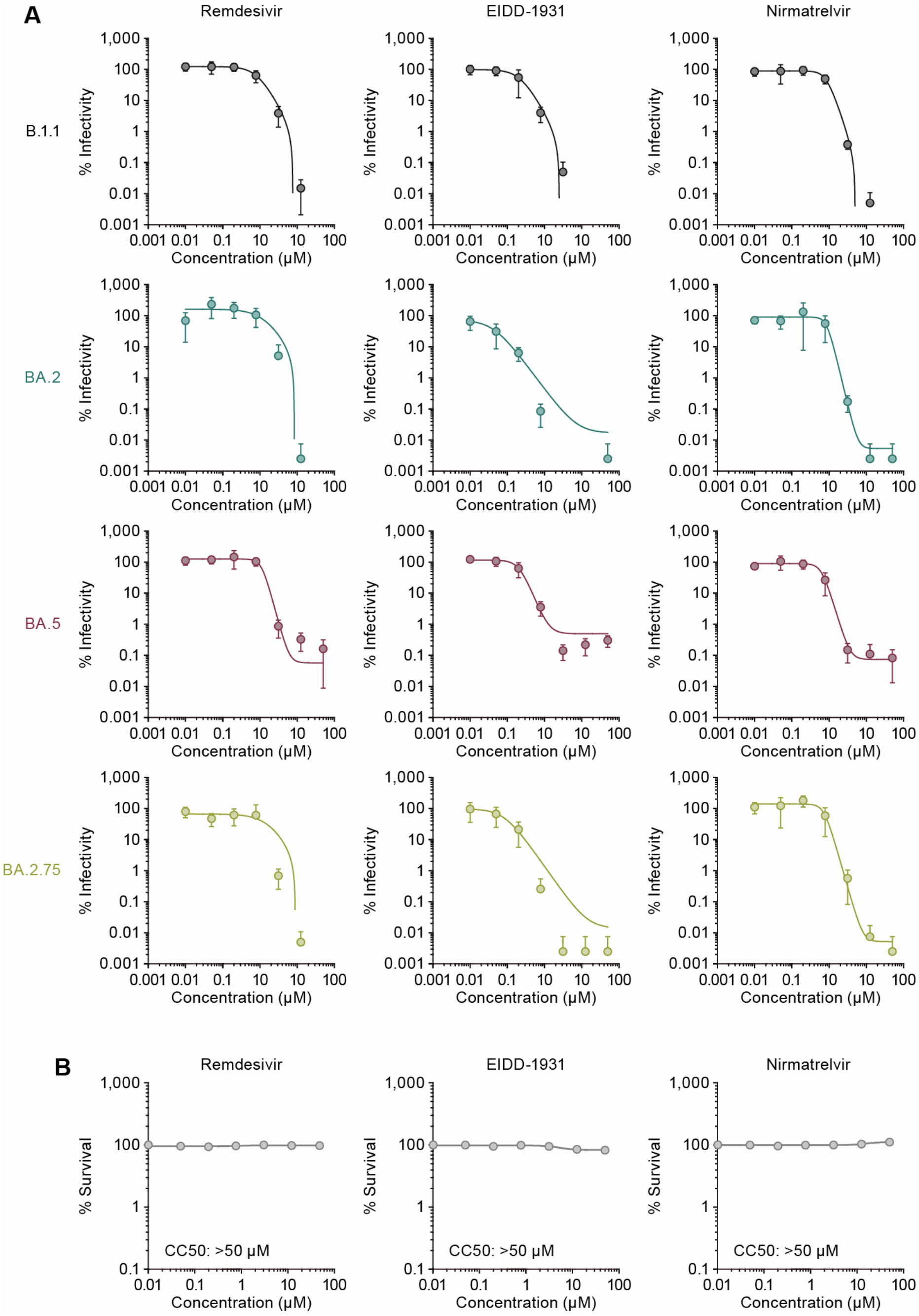

Figure S3. Effects of antiviral drugs in AOs, related to Table 2

(**A**) Antiviral effects of the three drugs in AO culture. The assay of each antiviral drug was performed in quadruplicate, and the 50% effective concentration (EC_{50}) was calculated. The data are summarized in **Table 2**.

(**B**) Cytotoxic effects of the three drugs in AO culture. The assay of each antiviral drug was performed in quadruplicate, and the 50% cytotoxic concentration (CC_{50}) was calculated. The CC_{50} values are indicated in the panels.

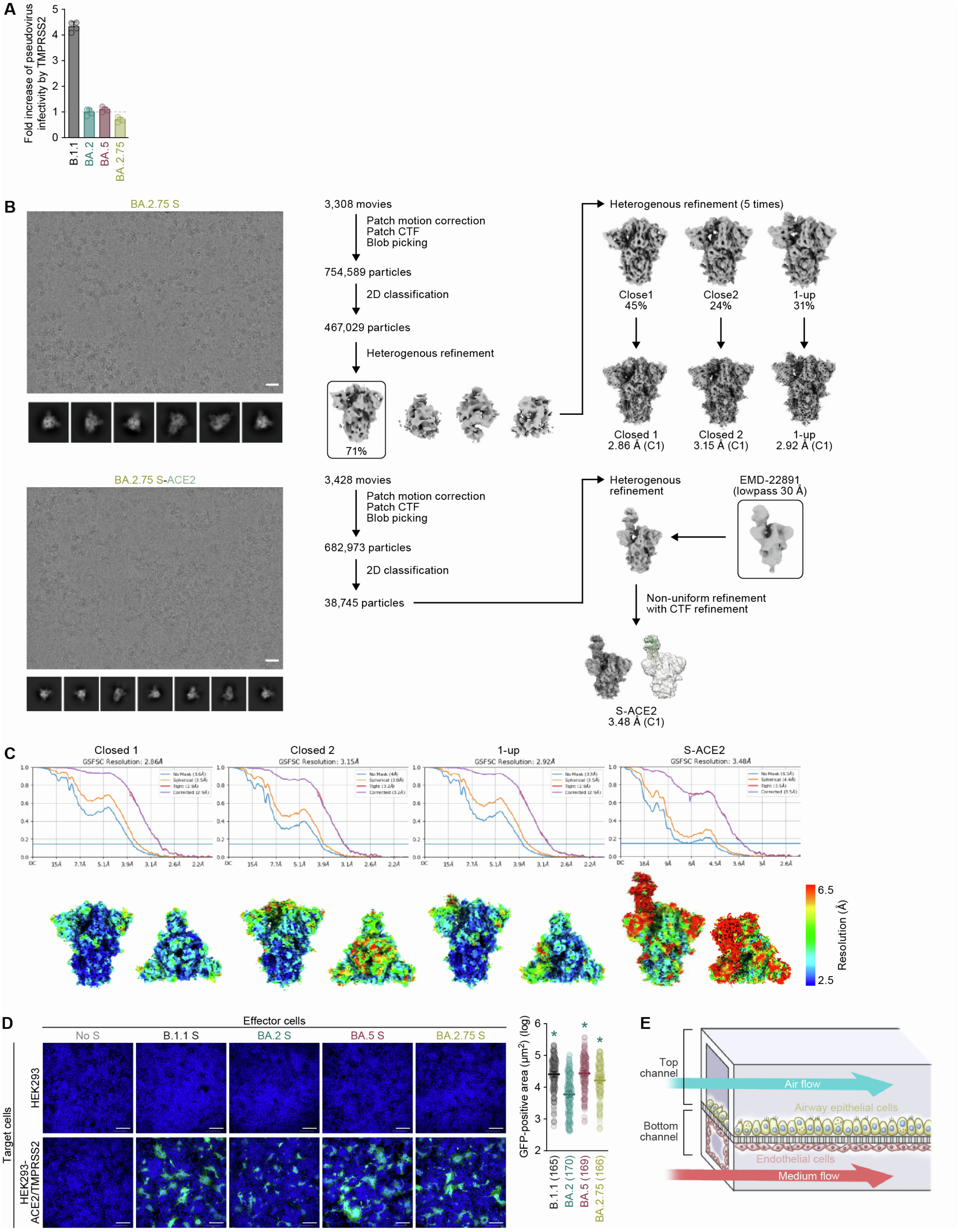
A
Fold increase of pseudovirus infectivity by TMPRSS2
0
1
2
3
4
5
B.1.1
BA.2
BA.5
BA.2.75
B
BA.2.75 S
3,308 movies
Patch motion correction
Patch CTF
Blob picking
754,589 particles
2D classification
467,029 particles
Heterogenous refinement
71%
Heterogenous refinement (5 times)
Close1
45%
Close2
24%
1-up
31%
Closed 1
2.86 Å (C1)
Closed 2
3.15 Å (C1)
1-up
2.92 Å (C1)
BA.2.75 S-ACE2
3,428 movies
Patch motion correction
Patch CTF
Blob picking
682,973 particles
2D classification
38,745 particles
Heterogenous refinement
EMD-22891
(lowpass 30 Å)
Non-uniform refinement
with CTF refinement
S-ACE2
3.48 Å (C1)
C
Closed 1
GSFSC Resolution: 2.86Å
Closed 2
GSFSC Resolution: 3.15Å
1-up
GSFSC Resolution: 2.92Å
S-ACE2
GSFSC Resolution: 3.48Å
6.5
2.5
Resolution (Å)
D
Effector cells
No S
B.1.1 S
BA.2 S
BA.5 S
BA.2.75 S
Target cells
HEK293
HEK293-ACE2/TMPRSS2
GFP-positive area (μm²) (log)
2
3
4
5
6
*
*
*
B.1.1 (165)
BA.2 (170)
BA.5 (169)
BA.2.75 (166)
E
Top channel
Bottom channel
Air flow
Airway epithelial cells
Endothelial cells
Medium flow

Figure S4. Virological features of BA.2.75 *in vitro* and the cryo-EM data of SARS-CoV-2 BA.2.75 S, related to Figures 3 and 4

(**A**) Fold increase in pseudovirus infectivity based on TMPRSS2 expression. Assays were performed in quadruplicate, and the presented data are expressed as the average ± SD. Each dot indicates the result of an individual replicate.

(**B and C**) Cryo-EM. (**B**) (Left) Representative micrograph and 2D classes showing spike-like images. Scale bars, 20 nm (Right) The cryo-EM data processing flowchart for BA.2.75 S and BA.2.75 S-ACE2. (**C**) Global resolution assessment of the S ectodomain by gold-standard Fourier shell correlation (FSC) curves at the 0.143 criteria. Local resolution is colored blue to red (2.5–6.5 Å).

(**D**) Coculture of S-expressing cells with HEK293-ACE2/TMPRSS2 cells. Left, representative images of S-expressing cells cocultured with HEK293 cells (top) or HEK293-ACE2/TMPRSS2 cells (bottom). Nuclei were stained with Hoechst 33342 (blue). Right, size distribution of syncytia (green). The numbers in parentheses indicate the numbers of GFP-positive syncytia counted. Each dot indicates the result of an individual replicate. Statistically significant differences between BA.2 and the other variants (*, $P < 0.05$) were determined by two-sided Mann–Whitney *U* tests. Scale bars, 200 μm.

(**E**) A scheme of the airway-on-a-chip system.


A
Delta
BA.2
BA.5
BA.2.75
B
Delta
8.2%
2.1%
1.8%
2.6%
BA.2
1.5%
1.1%
1.2%
0.9%
BA.5
1.8%
1.5%
1.2%
1.0%
BA.2.75
2.9%
1.7%
1.9%
1.7%
D
Delta
49.6%
38.8%
41.8%
56.7%
BA.2
25.0%
26.0%
20.4%
27.1%
BA.5
31.0%
40.5%
31.4%
48.8%
BA.2.75
42.9%
41.2%
48.9%
37.5%
C
Delta
4.4%
1.3%
2.6%
2.5%
BA.2
0.2%
0.3%
0.3%
0.1%
BA.5
0.8%
1.2%
0.7%
1.3%
BA.2.75
0.9%
0.5%
1.0%
0.4%

Figure S5. Histological observations in infected hamsters, related to Figure 5

(**A**) IHC of the viral N protein in the middle portion of the tracheas of all infected hamsters at 2 d.p.i (4 hamsters per infection group). Each panel shows a representative result from an individual infected hamster.

(**B and C**) IHC of the SARS-CoV-2 N protein in the lungs of infected hamsters at 2 d.p.i. (**B**) and 5 d.p.i (**C**) (4 hamsters per infection group). In each panel, IHC staining (top) and the digitalized N-positive area (bottom, indicated in red) are shown. The red numbers in the bottom panels indicate the percentage of the N-positive area. Summarized data are shown in **Figure 5E**.

(**D**) Type II pneumocytes in the lungs of infected hamsters (4 hamsters per infection group). H&E staining (top) and the digitalized inflammatory area with type II pneumocytes (bottom, indicated in red) are shown. The red numbers in the bottom panels indicate the percentage of inflammatory area with type II pneumocytes. Summarized data are shown in **Figure 5H**.

In **A–C**, N-positive cells are shown in brown.

Scale bars, 1 mm (**A**); 5 mm (**B–D**).

Table S1. Estimated relative Re values of viral lineages in India, related to Figure 1

Region	PANGO lineage	Posterior mean	Posterior 2.5 percentile	Posterior 97.5 percentile	R-hat value	Effective sampling size (ESS_bulk)	Effective sampling size (ESS_tail)
Average in India	BA.2.38	1.014	0.980	1.047	1.001	7494	4609
Average in India	BA.2.74	1.114	1.046	1.181	1.001	6997	5146
Average in India	BA.2.75	1.325	1.260	1.386	1.001	4687	5344
Average in India	BA.2.76	1.126	1.088	1.163	1.001	7073	5603
Average in India	BA.5	1.175	1.123	1.229	1.000	6885	5935
Average in India	other_BA.2	1.012	0.988	1.034	1.000	5769	4540
Delhi	BA.2.38	0.996	0.981	1.011	1.000	6369	5656
Delhi	BA.2.74	1.143	1.067	1.242	1.001	7747	5090
Delhi	BA.2.75	1.289	1.190	1.373	1.001	3139	5654
Delhi	BA.2.76	1.104	1.083	1.126	1.000	6791	5237
Delhi	BA.5	1.192	1.131	1.262	1.000	7912	5379
Delhi	other_BA.2	1.016	0.997	1.036	1.000	6711	6376
Gujarat	BA.2.38	1.026	1.006	1.048	1.000	4749	6121
Gujarat	BA.2.74	1.189	1.134	1.252	1.000	7025	5472
Gujarat	BA.2.75	1.342	1.276	1.417	1.000	6187	5479
Gujarat	BA.2.76	1.160	1.129	1.195	1.001	4956	5585
Gujarat	BA.5	1.183	1.142	1.227	1.000	6536	5581
Gujarat	other_BA.2	1.037	1.011	1.067	1.001	3976	6060
Haryana	BA.2.38	1.047	1.007	1.090	1.001	6768	4498
Haryana	BA.2.74	1.119	1.026	1.228	1.001	6173	4544
Haryana	BA.2.75	1.317	1.222	1.410	1.000	5500	5097
Haryana	BA.2.76	1.116	1.068	1.163	1.000	6834	5939
Haryana	BA.5	1.141	1.052	1.227	1.000	6620	5776
Haryana	other_BA.2	1.015	0.974	1.059	1.002	6977	4660
Himachal Pradesh	BA.2.38	0.999	0.947	1.046	1.000	5939	5138
Himachal Pradesh	BA.2.74	1.032	0.869	1.165	1.001	4892	4578
Himachal Pradesh	BA.2.75	1.265	1.155	1.362	1.001	2204	5090
Himachal Pradesh	BA.2.76	1.112	1.049	1.174	1.000	6078	5973
Himachal Pradesh	BA.5	1.204	1.090	1.376	1.000	5117	3189
Himachal Pradesh	other_BA.2	0.996	0.920	1.042	1.001	4613	3718
Maharashtra	BA.2.38	0.980	0.964	0.996	1.001	4975	5164
Maharashtra	BA.2.74	1.070	1.045	1.095	1.002	5661	4211
Maharashtra	BA.2.75	1.374	1.315	1.438	1.001	2884	3583
Maharashtra	BA.2.76	1.074	1.045	1.104	1.001	5863	5574
Maharashtra	BA.5	1.124	1.084	1.165	1.000	6480	5592
Maharashtra	other_BA.2	1.008	0.988	1.028	1.000	5377	6103
Odisha	BA.2.38	0.967	0.897	1.024	1.000	6823	5270
Odisha	BA.2.74	1.072	0.961	1.183	1.001	6126	5321
Odisha	BA.2.75	1.330	1.229	1.450	1.002	5748	4262

Odisha	BA.2.76	1.111	1.026	1.192	1.000	6253	5024
Odisha	BA.5	1.164	1.060	1.275	1.001	6784	5692
Odisha	other_BA.2	1.007	0.954	1.055	1.000	6286	4461
Tamil Nadu	BA.2.38	1.070	1.055	1.085	1.000	7489	6863
Tamil Nadu	BA.2.74	1.145	1.084	1.214	1.000	7918	5399
Tamil Nadu	BA.2.75	1.356	1.274	1.468	1.001	4178	3730
Tamil Nadu	BA.2.76	1.135	1.100	1.173	1.000	7163	6076
Tamil Nadu	BA.5	1.278	1.253	1.304	1.001	7469	6402
Tamil Nadu	other_BA.2	1.018	1.004	1.034	1.001	6446	4780
Telangana	BA.2.38	1.031	1.014	1.049	1.000	5771	5813
Telangana	BA.2.74	1.187	1.117	1.273	1.000	6569	4791
Telangana	BA.2.75	1.332	1.213	1.476	1.002	5857	3985
Telangana	BA.2.76	1.158	1.126	1.192	1.001	6709	6026
Telangana	BA.5	1.185	1.151	1.220	1.001	6817	5522
Telangana	other_BA.2	1.022	1.005	1.039	1.001	5654	5842
West Bengal	BA.2.38	1.011	0.986	1.036	1.000	3768	5238
West Bengal	BA.2.74	1.066	0.999	1.139	1.000	5677	5609
West Bengal	BA.2.75	1.320	1.235	1.402	1.000	5705	4714
West Bengal	BA.2.76	1.165	1.125	1.208	1.000	4894	5535
West Bengal	BA.5	1.139	1.088	1.191	1.000	5542	6235
West Bengal	other_BA.2	0.982	0.950	1.014	1.000	2503	2162

Information on the averaged Re value in India and Re values in individual states are summarized.

Table S2. Human sera used in this study, related to Figure 2

Vaccine	Donor ID	Sex	Age	2nd vac	Sampling date 1 mo after 2-dose	3rd vac	Sampling date 1 mo after 3-dose	Sampling date 4 mo after 3-dose	Prior infection?
BNT162b2	5	Female	34	2021-04-30	2021-05-27	2022-01-22	2022-02-24	2022-05-13	No
BNT162b2	27	Male	41	2021-04-27	2021-05-25	2022-01-22	2022-02-24	2022-05-13	No
BNT162b2	28	Female	42	2021-04-28	2021-05-26	2022-01-22	2022-03-16	2022-05-13	No
BNT162b2	36	Male	34	2021-04-27	2021-05-27	2022-01-25	2022-02-25	2022-05-19	No
BNT162b2	38	Female	24	2021-04-27	2021-05-26	2022-01-26	2022-02-18	2022-05-19	No
BNT162b2	46	Female	42	2021-04-27	2021-05-25	2021-12-26	2022-02-14	2022-05-18	No
BNT162b2	55	Female	45	2021-04-30	2021-05-28	2022-01-28	2022-02-18	2022-05-19	No
BNT162b2	61	Female	48	2021-06-04	2021-07-02	2022-01-29	2022-02-14	2022-05-17	No
BNT162b2	69	Male	42	2021-07-17	2021-08-12	2022-01-28	2022-02-22	2022-05-19	No
BNT162b2	71	Male	34	2021-04-08	2021-05-06	2021-12-28	2022-01-21	2022-05-13	No
BNT162b2	87	Male	34	2021-06-02	2021-06-30	2022-01-26	2022-02-16	2022-05-19	No
BNT162b2	89	Male	45	2021-06-04	2021-06-29	2022-01-24	2022-02-15	2022-05-17	No
BNT162b2	97	Female	43	2021-06-04	2021-07-02	2022-01-26	2022-02-16	2022-05-18	No
BNT162b2	100	Male	32	2021-06-08	2021-07-07	2022-01-22	2022-02-17	2022-05-17	No
BNT162b2	102	Male	30	2021-05-31	2021-06-28	2022-01-22	2022-02-21	2022-05-17	No

Vaccine	Donor ID	Sex	Age	2nd vac	Sampling date 1 mo after 2-dose	3rd vac	Sampling date 1 mo after 3-dose	Sampling date 4 mo after 3-dose	4th vac	Sampling date 1 mo after 4-dose	Prior infection?
BNT162b2/mRNA-1273	4	Female	43	2021-06-04	2021-07-01	2022-01-28	2022-02-24	2022-05-25	2022-08-01	2022-08-22	No
BNT162b2/mRNA-1273	8	Female	56	2021-06-04	2021-07-01	2022-01-28	2022-02-24	2022-05-20	2022-08-05	2022-08-26	No
BNT162b2/mRNA-1273	14	Female	43	2021-06-04	2021-07-01	2022-01-28	2022-02-18	2022-05-20	2022-08-05	2022-08-16	No
BNT162b2/mRNA-1273	28	Female	42	2021-04-28	2021-05-26	2022-01-22	2022-03-16	2022-05-13	2022-07-30	2022-08-23	No
BNT162b2/mRNA-1273	62	Male	46	2021-06-10	2021-07-08	2022-01-28	2022-02-22	2022-05-20	2022-08-05	2022-08-26	No
BNT162b2/mRNA-1273	63	Female	49	2021-05-31	2021-06-28	2022-02-05	2022-03-01	2022-05-26	2022-08-02	2022-08-25	No
BNT162b2/mRNA-1273	64	Female	42	2021-05-31	2021-06-28	2022-02-03	2022-02-24	2022-05-25	2022-08-01	2022-08-25	No
BNT162b2/mRNA-1273	67	Male	38	2021-06-01	2021-06-29	2022-02-01	2022-02-25	2022-05-25	2022-08-03	2022-08-24	No
BNT162b2/mRNA-1273	90	Male	45	2021-06-02	2021-06-30	2022-01-25	2022-02-15	2022-05-18	2022-08-02	2022-08-24	No
BNT162b2/mRNA-1273	92	Female	30	2021-06-10	2021-07-08	2022-01-26	2022-02-15	2022-05-18	2022-08-04	2022-08-26	No
BNT162b2/mRNA-1273	94	Male	36	2021-06-01	2021-06-29	2022-02-02	2022-02-25	2022-05-24	2022-08-05	2022-08-26	No
BNT162b2/mRNA-1273	97	Female	43	2021-06-04	2021-07-02	2022-01-26	2022-02-16	2022-05-18	2022-07-21	2022-08-19	No
BNT162b2/mRNA-1273	103	Male	35	2021-04-07	2021-05-07	2022-01-25	2022-02-15	2022-05-18	2022-08-02	2022-08-26	No
BNT162b2/mRNA-1273	113	Female	32	2021-06-04	2021-07-02	2022-01-28	2022-02-18	2022-05-20	2022-08-02	2022-08-23	No
BNT162b2/mRNA-1273	122	Female	45	2021-06-11	2021-07-08	2022-02-16	2022-03-16	2022-06-07	2022-08-01	2022-08-22	No

Table S2. Human sera used in this study, related to Figure 2 (continued)

SARS-CoV-2 infected	Donor ID	Sex	Age	Date of test	Sampling date	Prior infection?	Prior vaccination?	Vaccine	Date of 1st vaccination	Date of 2nd vaccination	Date of 3rd vaccination	Date of 4rd vaccination
BA.1	P264	Male	60	2022/01/03	2022/01/29	No	Yes	BNT162b2	2021/08/27	2021/09/18		
BA.1	P265	Female	57	2022/01/04	2022/01/28	No	Yes	BNT162b2	2021/10/04	2021/10/25		
BA.1	P274	Female	20	2022/01/15	2022/01/29	No	Yes	mRNA-1273	2021/09/11	2021/10/09		
BA.1	P276	Male	44	2022/01/20	2022/01/30	No	Yes	BNT162b2	2021/08/06	2021/08/22		
BA.1	P279	Male	61	2022/01/18	2022/02/05	No	Yes	BNT162b2	2021/07/03	2021/07/24		
BA.1	P282	Male	56	2022/01/22	2022/02/06	No	Yes	BNT162b2	2021/09/18	2021/10/12		
BA.1	P285	Female	65	2022/01/23	2022/02/06	No	Yes	BNT162b2	2022/01/07	2022/01/28		
BA.1	P295	Female	41	2022/01/25	2022/02/06	No	Yes	BNT162b2	2021/08/07	2021/08/28		
BA.1	P288	Female	76	2022/01/24	2022/02/07	No	Yes	BNT162b2	2021/06/29	2021/07/20		
BA.1	P289	Female	51	2022/01/24	2022/02/07	No	Yes	BNT162b2	2021/08/02	2021/08/23		
BA.1	P290	Male	53	2022/01/25	2022/02/07	No	Yes	BNT162b2	2021/09/02	2021/09/30		
BA.1	2651	Male	71	NA	2022/02/14	No	Yes	BNT162b2	2021/06/14	2021/07/12		
BA.1	1880	Female	25	2022/01/07	2022/02/03	No	Yes	BNT162b2	2021/09/09	2021/09/30		
BA.1	2187	Male	32	2022/01/20	2022/02/05	No	Yes	mRNA-1273	2021/07/09	2021/08/06		
BA.1	2137	Female	32	2022/01/19	2022/02/05	No	Yes	mRNA-1273	2021/07/09	2021/08/06		
BA.1	2550	Female	24	2022/01/25	2022/02/05	No	Yes	BNT162b2	2021/10/12	2021/11/02		
BA.2	P378	Male	43	2022/03/28	2022/04/10	No	Yes	BNT162b2	2021/10/10	2021/10/31		
BA.2	P398	Male	48	2022/04/13	2022/04/30	No	Yes	BNT162b2	2021/09/18	2021/10/09	2022/04/09	
BA.2	P407	Male	29	2022/05/01	2022/05/12	No	Yes	mRNA-1273	2021/09/13	2021/10/11		
BA.2	P401	Male	35	2022/04/22	2022/05/05	No	Yes	BNT162b2	2021/09/09	2021/09/30		
BA.2	P412	Female	82	2022/05/04	2022/05/26	No	Yes	BNT162b2	2021/06/11	2021/07/09		
BA.2	6449	Male	43	2022/04/03	2022/04/23	No	Yes	BNT162b2	2021/08/13	2021/09/11		
BA.2	6355	Male	50	2022/04/02	2022/04/20	No	Yes	NA	2021/04/28	2021/05/19	2022/01/19	
BA.2	6547	Male	54	2022/04/06	2022/04/22	No	Yes	BNT162b2	2021/08/25	2021/09/15		
BA.2	7951	Female	71	2022/04/25	2022/05/12	No	Yes	Mix	2021/06/20 (BNT162b2)	2021/07/16 (BNT162b2)	2022/02/16 (mRNA-1273）	
BA.2	8645	Female	41	2022/05/07	2022/05/20	No	Yes	BNT162b2	2021/05/23	2021/06/13	2022/01/20	
BA.2	8682	Female	25	2022/05/08	2022/05/24	No	Yes	BNT162b2	2021/09/03	2021/09/27		
BA.2	5949	Male	24	2022/03/22	2022/05/22	No	Yes	mRNA-1273	2021/08/05	2021/09/02		
BA.2	8463	Male	84	2022/05/05	2022/05/23	No	Yes	Mix	2021/05/27 (BNT162b2)	2021/06/21 (BNT162b2)	2022/02/22 (mRNA-1273）	
BA.2	8796	Female	34	2022/05/10	2022/06/05	No	Yes	BNT162b2	2021/09/26	2021/10/17		
BA.5	P427	Female	49	2022/07/06	2022/07/25	No	Yes	NA	2021/07/30	2021/08/25	2022/03/18	
BA.5	P440	Male	25	2022/07/24	2022/08/07	No	Yes	BNT162b2	2021/11/24	2021/12/15		
BA.5	P439	Female	73	2022/07/23	2022/08/08	No	Yes	BNT162b2	2021/06/19	2021/07/20	2022/02/04 (mRNA-1273)	
BA.5	P451	Female	55	2022/07/29	2022/8/12	No	Yes	BNT162b2	2021/04/26	2021/05/20	2022/01/18	
BA.5	P456	Male	44	2022/08/04	2022/08/14	No	Yes	BNT162b2	2021/08/11	2021/09/01	2022/03/13 (mRNA-1273)	
BA.5	P455	Male	29	2022/08/03	2022/08/17	No	Yes	mRNA-1273	2021/07/26	2021/08/27	2022/04/18	
BA.5	P464	Male	63	2022/08/08	2022/08/19	No	Yes	BNT162b2	2021/08/08	2021/08/29	2022/04/07 (mRNA-1273)	
BA.5	9341	Male	56	2022/06/12	2022/06/30	No	Yes	BNT162b2	2021/08/10	2021/08/31	2022/03/18	

BA.5	9584	Male	55	2022/07/08	2022/07/25	No	Yes	BNT162b2	2021/07/14	2021/08/05	2022/03/22	
BA.5	11318	Female	51	2022/07/24	2022/08/05	No	Yes	BNT162b2	2021/09/01	2021/09/22	2022/05/19 (mRNA-1273)	
BA.5	23S-08	Male	25	2022/07/23	2022/08/08	No	Yes	BNT162b2	2021/04/27	2021/05/18	2022/01/11	
BA.5	11597	Female	41	2022/07/26	2022/08/08	No	Yes	BNT162b2	2021/04/30	2021/05/21	2022/01/11	
BA.5	10978	Female	46	2022/07/22	2022/08/11	No	Yes	BNT162b2	2021/08/27	2021/09/17	2022/05/15	
BA.5	10826	Male	63	2022/07/21	2022/08/11	No	Yes	BNT162b2	2021/07/27	2021/08/17	2022/03/04	2022/8/9 (mRNA-1273)
BA.5	11079	Female	65	2022/07/23	2022/08/11	No	Yes	mRNA-1273	2021/07/08	2021/08/05	2022/03/17	
BA.5	14847	Female	70	2022/08/13	2022/08/25	No	Yes	BNT162b2	2021/07/13	2021/08/20	2022/03/08 (mRNA-1273)	
BA.5	13180	Female	63	2022/08/04	2022/08/25	No	Yes	BNT162b2	2021/07/16	2021/08/06	2022/03/08 (mRNA-1273)	
BA.5	12912	Male	64	2022/08/02	2022/08/25	No	Yes	mRNA-1273	2021/09/02	2021/09/30	2021/04/01	
BA.5	14956	Female	33	2022/08/13	2022/08/28	No	Yes	BNT162b2	2021/09/06	2021/10/07		
BA.5	15707	Female	52	2022/08/16	2022/08/29	No	Yes	BNT162b2	2021/08/07	2021/08/28	2022/04/03	

NA, not applicable.

Table S3. IC50 values of five therapeutic monoclonal antibodies against BA.2.75 and BA.2 derivatives, related to Table 1

	BA.2	BA.2.75	BA.2 K147E	BA.2 W152R	BA.2 F157L	152R/F157L)	BA.2 I210V	BA.2 G257S	BA.2 D339H	BA.2 G446S	BA.2 N460K	BA.2 R493Q
Bebtelovimab	1.1	37.4	1.2	1.8	1.5	2.1	1.8	1.6	1.4	25.6	1.3	2.9
Cilgavimab	26.7	468.5	19.7	27.1	24.4	20.1	18.6	22.5	22.3	210.2	21.1	38.0
Regdanvimab	>6175	57.9	>6175	>6175	>6175	>6175	>6175	>6175	>6175	>6175	>6175	39.7
Sotrovimab	653.8	196.6	893.6	783.9	858.8	485.9	990.3	716.5	236.5	894.0	589.4	533.3
Tixgevimab	2067.0	44.9	>4375	>4375	983.9	>4375	2575.0	>4375	1487.0	>4375	1334.0	38.6

The average 50% inhibitory concentration (IC50; ng/mL) ± 95% confidential interval is shown. Representative neutralization curves are shown in **Figure S2B**.

Table S4. Statistics of the cryo-EM dataset, data processing and structure refinement, related to Figure 3

	SARS-CoV-2 BA.2.75 spike			SARS-CoV-2 BA.2.75 spike S RBD-ACE2
Structures	closed 1	closed 2	1-up	
EMDB ID	EMD-34221	EMD-34222	EMD-34223	EMD-34224
PDB ID	8GS6	-	-	-
Data collection				
Microscope	Krios G4			Krios G4
Detector	Gatan Biocontinuum energy filter with Gatan K3 direct electron detector			Gatan Biocontinuum energy filter with Gatan K3 direct electron detector
Voltage (kV)	300			300
Nominal magnification	130,000			130,000
Defocus range (μm)	–0.8 to –1.8			–0.8 to –1.8
Physical pixel (Å)	0.67			0.67
Electron dose (e/Å)	50			50
Number of raw frames	50			50
Data processing				
Extracted particles (n)	754589			682973
Particles for final map (n)	139418	76153	96181	38745
Symmetry imposed	C1	C1	C1	C1
Resolution	2.86	3.15	2.92	3.48
FSC threshold	0.143	0.143	0.143	0.143
Model composition				
Initial model used	7UB0			
Non-hydrogen atoms	24538			
Protein residues	3058			
Ligands	NAG:41			
R.m.s. deviations				
Bond length (Å)	0.004			
Bond angles (°)	0.59			
Validation				
MolProbity score	1.54			
Clash score	6.1			
Rotamer outliers (%)	0			
Ramachandran plot				
Favored (%)	95.64			
Allowed	4.36			
Outliers	0			
Model vs. Data				
CC (mask)	0.84			

Table S5. Primers used for the construction of SARS-CoV-2 S expression plasmids, related to Figures 2 and 3

Primer name	Sequence (5'-to-3')
Omicron universal Fw	cactatagggcgaattgggtaccatgtttgtgttcctggt
BA2 Rv	agctccaccgcggtggcggccgctcaggtgtagtgcagtttca
pC_S_BA2_147_152_157_R	cagactccattctggacttgttgttctcgtggtagtagac
pC_S_BA2_147_152_157_F	caacaagtccagaatggagtctgagctgagggtctactcc
pC_S_BA2_K147E_R	ggacttgttgttctcgtggtagtagac
pC_S_BA2_K147E_F	gtctactaccacgagaacaacaagtcc
pC_S_BA2_W152R_R	ctcagactccattctggacttgttgtt
pC_S_BA2_W152R_F	aacaacaagtccagaatggagtctgag
pC_S_BA2_F157L_R	ggagtagaccctcagctcagactccat
pC_S_BA2_F157L_F	atggagtctgagctgagggtctactcc
pC_S_BA2_I210V_R	cctgcccaggttcactggtgtgtgttt
pC_S_BA2_I210V_F	aaacacacaccagtgaacctgggcagg
pC_S_BA2_G257S_R	tcctgctgtccagctagaggaggagtc
pC_S_BA2_G257S_F	gactcctcctctagctggacagcagga
pC_S_BA2_D339H_R	attgaacacctcgtgaaatggacacag
pC_S_BA2_D339H_F	ctgtgtccatttcacgaggtgttcaat
pC_S_BA2_G446S_R	gttgtagttgccgctcaccttgctgtc
pC_S_BA2_G446S_F	gacagcaaggtgagcggcaactacaac
pC_S_BA2_N460K_R	aaatggtttcagcttgctcttcctgaa
pC_S_BA2_N460K_F	ttcaggaagagcaagctgaaaccattt
pC-S_BA2_R493Q-F	ttactttccactccaatcctatggcttca
pC-S_BA2_R493Q-R	tgaagccataggattggagtggaaagtaa
BA2_G446S_R	CCTGAACAGTCTGTAGAGGTAGTTGTAGTTGCCACTCACCTTGCTGTCCAGTTTGTTG
BA2_D339H_F	GTAGCGGAGGCGGAGGGTCGGCTAGCCATCCAAACATCACCAACCTGTGCCCTTTTCATGAAGTTTTTAACGCCACCAGG
BA2_N460K_R	CTCTGTGCTGATGTCCCTCTCAAATGGTTTCAGTTTGCTCTTCCTGAACAGTCTGTAGAGG
BA2_R493Q_R	GCCCACTCCATAGGTTGGACGGAAGCCATAGGACTGGAGTGGAAAGTAACAGTTGAAGCCG
BA2_CON3_F	GGCGGTAGCGGAGGCGGAGGGTCGGCTAGCCATCCAAACATCACCAACCTGTGCCCTTTTGATGAAGTTTTTAACGCCACCAGG
RBDO_CON3_F	GGAGGCGGTAGCGGAGGCGGAGGGTCGGCTAGCCATCCTAATATCACCAATCTGTGCCCTTTTGGTGAAGTTTTTAACG
BA275_N460N_F	CTACAACTACCTCTACAGACTGTTCAGGAAGAGCAACCTGAAACCATTTGAGAGGGACATCAG
ACE2_N90Q_F	CACTTGCCCAAATGTATCCACTACAAGAAATTCAGGAACTCACAGTCAAGCTTCAGCTGC

Table S6. Summary of unexpected amino acid mutations detected in the working virus stocks, related to Figures 4 and 5 and Table 2

Original clinical isolate	Reference	Position in Reference (nt)	REF	ALT	Quality score	Depth in REF	Depth in ALT	Mutation type	ORF	Amino acid substitution
B.1.1 (EPI_ISL_479681)	NC_045512.2	14408	C	T	225	0	190	missense_variant	NSP12	P303L
BA.1 (EPI_ISL_7418017)	NC_045512.2	10448	CC	TA	225	0	218	missense_variant	NSP5	P132Y
BA.1 (EPI_ISL_7418017)	NC_045512.2	11750	C	T	159	177	56	missense_variant	NSP6	L260F
BA.1 (EPI_ISL_7418017)	NC_045512.2	20486	A	C	222	76	86	missense_variant	NSP15	K289T
BA.2 (EPI_ISL_9595859)	NC_045512.2	no additional mutation								
BA.5 (EPI_ISL_12812500)	NC_045512.2	2388	C	T	228	1	195	missense_variant	NSP2	T528I
BA.5 (EPI_ISL_12812500)	NC_045512.2	11522	T	G	79	180	45	missense_variant	NSP6	F184V
BA.5 (EPI_ISL_12812500)	NC_045512.2	11750	C	T	222	108	71	missense_variant	NSP6	L260F
BA.2.75 (EPI_ISL_13969765)	NC_045512.2	no additional mutation								