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Published on: 19 Jul 2021 - [bioRxiv](#) (Cold Spring Harbor Laboratory)

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1 **Comparison of Neutralizing Antibody Titers Elicited by mRNA and**
2 **Adenoviral Vector Vaccine against SARS-CoV-2 Variants**

3

4

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22

23 **Abstract**

24 The increasing prevalence of SARS-CoV-2 variants has raised concerns regarding
25 possible decreases in vaccine efficacy. Here, neutralizing antibody titers elicited by
26 mRNA-based and an adenoviral vector-based vaccine against variant pseudotyped
27 viruses were compared. BNT162b2 and mRNA-1273-elicited antibodies showed modest
28 neutralization resistance against Beta, Delta, Delta plus and Lambda variants whereas
29 Ad26.COV2.S-elicited antibodies from a significant fraction of vaccinated individuals were
30 of low neutralizing titer ($IC_{50} < 50$). The data underscore the importance of surveillance for
31 breakthrough infections that result in severe COVID-19 and suggest the benefit of a
32 second immunization following Ad26.COV2.S to increase protection against the variants.

33 Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) vaccines from two
34 vaccine platforms have been granted U.S. Food and Drug Administration (FDA)
35 Emergency Use Authorization: mRNA-based (Pfizer and Moderna) and adenoviral
36 vector-based (Johnson & Johnson (J&J)), all of which have been shown to be highly
37 effective. The mRNA-based vaccines were 94-95% effective in preventing COVID-19¹
38 whereas the adenoviral vector-based J&J vaccine had 66.9% efficacy in preventing
39 moderate to severe disease². However, the ongoing emergence of highly transmissible
40 variants with mutations in the spike protein raises concerns regarding possible decreases
41 in vaccine effectiveness due to spike protein antigenic variability.

42
43 SARS-CoV-2 variants have been classified by the World Health Organization (WHO)
44 based on increased transmissibility and/or pathogenicity as variants of concern (VOC;
45 Alpha (B.1.1.7), Beta (B.1.351), Gamma (B.1.1.248) and Delta (B.1.617.2) and variants
46 of interest (VOI; Epsilon (B.1.427/B.1.429), Iota (B.1.526), and Delta plus (AY.1) and
47 Lambda (C.37)³. The increased transmissibility and/or pathogenicity of the variants is due,
48 at least in part, to mutations in the spike protein RBD that increase its affinity for ACE2
49 on target cells. Mutations in the Beta, Gamma and Delta variant spike RBDs have been
50 shown to cause partial resistance to neutralization by the serum antibodies of vaccinated
51 and convalescent individuals and therapeutic monoclonal antibodies⁴⁻¹¹.

52
53 This study compared the neutralization titers of serum antibodies from individuals
54 immunized with three U.S. FDA Emergency use authorization vaccines (BNT162b2,
55 mRNA-1273 and Ad26.COVS) against viruses with the VOC and Lambda spike proteins.

56 The study groups were controlled for age, clinical co-morbidity, history of pre-vaccination
57 infection and sera were collected on similar days post-vaccination. The results
58 demonstrate a high level of cross-neutralization by antibodies elicited by BNT162b2 and
59 mRNA-1273 on the variants but significantly decreased neutralization by those elicited by
60 the single dose Ad26.COVS.S.

61

62 **Variant pseudotyped lentiviruses.** The Delta plus spike contains K417N, L452R and
63 T478K in the RBD (**Figure S1A**). The Lambda spike protein contains novel L452Q and
64 F490S mutations in the RBD (**Figure S1A**). We previously described the production of
65 lentiviruses pseudotyped by the Alpha, Beta, Gamma and Delta spike proteins and here
66 report the generation of pseudotypes with the Delta plus and Lambda variant spike
67 proteins and the individual constituent mutations. The variant spike proteins were well
68 expressed, proteolytically processed and incorporated into lentiviral virions at a level
69 similar to that of the parental D614G spike protein in the producer cells and virions (**Figure**
70 **S1B**). The measurement of neutralizing antibody titers with such pseudotypes has been
71 shown to yield results consistent with those obtained with the live virus plaque reduction
72 neutralization test¹².

73
74 **Reduced sensitivity of virus with variant spikes to neutralization by convalescent**
75 **sera and mRNA vaccine-elicited antibodies.** Sera from individuals who had been
76 infected prior to the emergence of the variants (collected 32-57 days post symptom onset)
77 neutralized virus with the D614G spike protein with an average IC₅₀ titer of 346 and
78 neutralized the Alpha variant with a similar titer (IC₅₀ of 305). Neutralizing titers for Beta,
79 Delta, Delta plus and Lambda variants were decreased 3.2-4.9-fold relative to D614G,
80 indicative of a modest resistance to neutralization (**Figure 1A, Table S1**). The sera of
81 individuals vaccinated with BNT162b2 and mRNA-1273 that were collected 7-days post-
82 second injection – a peak antibody response timepoint - neutralized virus with the D614G
83 spike with significantly higher titer (1835 and 1594, respectively) relative to the
84 convalescent sera, and the antibodies cross-reacted on the variants with a modest 2.5-

85 4.0-fold decrease in titer (**Figure 1A**). The resistance of the Beta variant was attributed
86 to the E484K mutation whereas resistance of the Delta variant was attributed to the L452R
87 mutation (**Figure S2**). The resistance of the lambda variant was attributed to both the
88 L452Q and F490S mutations (**Figure S2**).

89

90 **Resistance of viruses with variant spike proteins to neutralization by Ad26.COVS-**

91 **elicited antibodies.** We next compared the neutralizing titers of antibodies elicited by the

92 BNT162b2 and mRNA-1273 mRNA vaccines with that of the Ad26.COVS adenoviral

93 vector-based vaccine. The sera analyzed were collected from individuals at similar time-

94 points post-final injection, on average (90 days for BNT162b2, 80 days for mRNA-1273

95 and 82 days for Ad26.COVS; **Table S2**) and from individuals of similar age and with

96 similar clinical co-morbidities (**Table S2**). None of the participants had a history of COVID-

97 19 pre- or post-vaccination and all were negative for antibodies against the SARS-CoV-

98 2 N protein (**Table S2**). The results showed that BNT162b2 sera neutralized virus with

99 the D614G and Alpha spikes with an average titer of 695 and 626. Compared to the

100 D614G, the neutralizing titer against Beta was decreased 6.1-fold and Delta plus was

101 decreased 2.7-fold. Results for the mRNA-1273 vaccine were similar with a 3.3-fold

102 decrease in neutralizing titer for Delta plus and 4.6-fold for Beta. Ad26.COVS sera

103 neutralized D614G and Alpha variants with average IC₅₀ titers of 221 and 232,

104 respectively, and neutralized the variants with titers that were decreased by 5.4-fold for

105 Delta plus to 6.7-fold for the Beta variant as compared to D614G (**Figure 1B**).

106 Presentation of the data grouped by variant shows the decreased neutralizing titers

107 against the variants by sera of the Ad26.COVS-vaccinated individuals (**Figure 1C**).

108
109 **The L452R/Q mutation of the Delta plus and Lambda spike proteins increases**
110 **infectivity and affinity for ACE2.** Measurement of the infectivity of the pseudotyped
111 viruses, normalized for particle number, showed that the Lambda variant spike protein
112 increased viral infectivity by 2-fold (**Figure 2A**), an increase equivalent to that of the Delta
113 and Delta plus variants. The increase was due to the L452Q mutation and was similar to
114 that of the L452R found in the Delta and Delta plus variants. The other mutations (Δ 246-
115 252, G75V-T76I, F490S and T859N) had no significant effect on infectivity (**Figure 2A**).
116 Measurement of the relative affinity of the variant spike proteins for ACE2 using sACE2
117 neutralization assay showed that variant spikes had a 3-fold increase in sACE2 binding
118 (**Figure 2B**). This increase was confirmed in a virion:ACE2 binding assay (**Figure 2C**).
119 The increase was caused by the L452R and L452Q mutation and were similar to the
120 increase caused by the N501Y mutation^{13,14}.

121
122 **Neutralization by REGN10933 and REGN10987.** Analysis of REGN10933 and
123 REGN10987 monoclonal antibodies that constitute the REGN-COV2 therapy showed that
124 REGN10933 had decreased activity against the Beta variant spike which resulted in a
125 127-fold decrease in neutralizing titer. REGN10933 also had decreased activity against
126 the Delta plus variant which resulted in a 92.7-fold decrease in neutralizing titer. The
127 resistance to REGN10933 was attributed to K417N and E484K (**Figure S3**). REGN10933
128 neutralized virus with the Delta variant spike with a 12-fold decrease in titer which had
129 only a minor effect on the activity of the cocktail. REGN10987 showed a minor reduction
130 in neutralizing titer of virus with the Beta, Delta, Delta plus and Lambda variant spikes but

131 this had little effect on neutralization of the virus by the cocktail (**Figure 2D**). The
132 resistance of variants to REGN10987 was attributed to the L452R/Q (**Figure S3**).
133

134 Discussion

135 Several reports have shown partial resistance of SARS-CoV-2 VOCs to vaccine-elicited
136 antibodies⁴⁻¹¹. The data shown here extend those findings to the Delta plus and Lambda
137 variants. Delta plus and Lambda, VOIs, both displayed a degree of resistance to mRNA
138 vaccine-elicited antibodies similar to that of the Beta and Delta variants. In sera collected
139 ~3 months post-second immunization, BNT162b2 and mRNA-1273 mRNA vaccine-
140 elicited antibodies neutralized the variants with a modest 3-fold average decrease in titer
141 resulting in an average IC₅₀ of about 1:600, a titer that is greater than that of convalescent
142 sera and likely, in combination with post-vaccination T- and B-cell memory responses, to
143 provide durable protection. Ad26.COVS2 vaccination-elicited neutralizing antibodies
144 showed a more pronounced decrease in neutralizing titer against the variants, raising the
145 potential for decreased protection against the VOCs and the Lambda variant. Vaccination
146 with Ad26.COVS2 resulted in IC₅₀ titers against Beta, Delta, Delta plus and Lambda
147 variants that decreased 5-7-fold, resulting in mean neutralizing antibody titers of 33, 30,
148 41, and 36 against viruses with the Beta, Delta, Delta plus and Lambda variant spikes,
149 respectively, which according to mathematical modeling, could result in decreased
150 protection against infection¹⁵. Modeling predicts that 50% protection from infection is
151 provided by a titer that is 20% that of the mean convalescent titer. In this study, given a
152 mean convalescent titer of 346 (**Table S1**), 50% protection would correspond to an IC₅₀
153 of 69. The titer required to protect against severe disease was shown to be 3% that of the
154 mean titer of convalescent sera which in this study corresponds to a titer of 10. In a
155 published report of phase 3 trial data, a single dose of Ad26.COVS2, 28 days post
156 administration, provided 64.0% protection against moderate to severe disease and 81.7%

157 against severe-critical COVID-19 in a country where 95% of circulating SARS-CoV-2 was
158 the Beta variant². The authors considered possible roles for non-neutralizing antibody Fc-
159 mediated effector functions and the role of the T cell response in maintaining protection
160 against the partially neutralizing antibody-resistant Beta variant.

161
162 The data reported here differ somewhat from those reported by Barouch *et al.* and
163 Jongeneelen *et al.* who found that Ad26.COVS-elicited antibody titers were mostly
164 maintained against the variants^{16,17}. In addition, Alter *et al.* reported a 5-fold decrease in
165 neutralizing antibody titer against Beta and 3.3-fold decrease against the Gamma variant
166 by the sera from Ad26.COVS vaccination¹⁸ which were less pronounced than those
167 reported here. While the studies used similar assays to measure antibody neutralization
168 and analyzed sera collected at a similar time-point post-immunization, it is possible that
169 differences in the study populations accounted for the experimental differences.

170
171 Several recent studies have shown that boosting a single immunization of the
172 ChAdOx1nCoV-19 adenoviral vector vaccine with BNT162b2 resulted in high neutralizing
173 titer against the VOCs¹⁹⁻²¹. It is likely that neutralizing antibody titers against the VOCs
174 elicited by the single shot Ad26.COVS could similarly be improved by boosting with a
175 second immunization or by a heterologous boost with one of the mRNA vaccines. While
176 a single dose vaccination has advantages, the benefit provided by a second immunization
177 may be well worth the inconvenience.

178

179 The data presented here emphasize the importance of surveillance for breakthrough
180 infections with the increased prevalence of highly transmissible variants. If an increase in
181 breakthrough infections accompanied by severe COVID-19 is found following adenovirus
182 vector or mRNA vaccination, this would provide a rationale for public health policy-makers
183 and manufacturers to consider booster immunizations that would increase protection
184 against the VOCs and Lambda variant. As such a need is not currently evident, the public
185 health apparatus should focus on primary immunization in the U.S. and globally.

186

187

188 **Methods**

189 **Clinical Samples**

190 Convalescent sera were collected 32-57 days post-symptom onset. For the early time-
191 point, BNT162b2 and Moderna-vaccinated sera were collected on day 28 and 35,
192 respectively, 7 days post-second immunization. For the later time-point, BNT162b2-
193 vaccinated sera were on average collected 90 days post-second immunization and
194 mRNA-1273-vaccinated sera were collected on average 80 days post-second
195 immunization. Ad26.COV2.S-vaccinated sera were collected, on average, 82 days post-
196 immunization (**Table S2**). Blood was drawn at the NYU Vaccine Center with written
197 consent under IRB approved protocols (IRB 18-02035 and IRB 18-02037). REGN10933
198 and REGN10987 were generated as previously described²².

199

200 **SARS-CoV-2 spike lentiviral pseudotypes**

201 Lentiviruses pseudotyped by variant SARS-CoV-2 spikes were produced as previously
202 reported²³ and normalized for reverse transcriptase (RT) activity. Neutralization titers of
203 sera, monoclonal antibody and soluble ACE2 (sACE2)²⁴ were determined as previously
204 described²³.

205

206 **sACE2 pull-down assay**

207 sACE2-bound-beads were mixed with pseudotyped virions as previously described²⁴.
208 The amount of virus bound was quantified by immunoblot analysis of bound p24.

209

210 **Statistical Analysis**

211 All experiments were in technical duplicates or triplicates. Statistical significance was
212 determined by two-tailed, unpaired t-test with confidence intervals shown as the mean \pm
213 SD or SEM. (* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$). Spike protein structure
214 (7BNM)²⁵ was downloaded from the Protein Data Bank.

215

216 **Acknowledgements**

217 The work was funded in part by grants from the NIH to N.R.L. (DA046100, AI122390 and
218 AI120898) and to M.J.M. (UM1AI148574). T.T. was supported by the Vilcek/Goldfarb
219 Fellowship Endowment Fund. M.J.M. and M.I.S. were partially supported by NYU
220 Grossman SOM institutional support.

221

222 **Author contributions**

223 T.T. and N.R.L. designed the experiments. H.Z., T.T. and B.M.D. carried out the
224 experiments and analyzed data. T.T., H.Z. and N.R.L. wrote the manuscript. M.I.S. and
225 M.J.M. designed and supervised the specimen selection, clinical information collection
226 and the N ELISAs, and provided key reagents and useful insights. All authors provided
227 critical comments on manuscript.

228

229 **Declaration of Interests.**

230 The authors declare no competing interests except M.J.M. who received research
231 grants from Lilly, Pfizer, and Sanofi, and serves on advisory boards for Pfizer and
232 Meissa Vaccines

233

234 **Figure legends**

235

236 **Figure 1. Comparison of neutralization titers of variant spike protein pseudotyped**
237 **viruses by convalescent sera, antibodies elicited by BNT162b2, mRNA-1273,**
238 **Ad26.COVS2.S.**

239 (A) Neutralization of variant spike protein pseudotyped viruses by convalescent serum
240 (n=8) (left). Neutralizing titers of serum samples from BNT162b2 vaccinated individuals
241 (n=15) (middle). Neutralizing titers of serum samples from mRNA-1273 vaccinated
242 donors (n=6) (right). The serum was collected at early time point (7 days after second
243 immunization). The neutralization IC₅₀ from individual donors is shown. Significance is
244 based on two-sided t-test.

245 (B) Comparison of neutralization of variants by convalescent serum (n=8, the same
246 donors in A), BNT162b2 vaccinated individuals (n=9), mRNA-1273 vaccinated donors
247 (n=8), Ad26.COVS2.S vaccinated donors (n=10), sera from vaccinated individuals were
248 collected at later time points (90, 80, 82 days on average after last immunization of each
249 vaccine, see the table S2) . Each line shows individual donors.

250 (C) Comparison of neutralization potency of each vaccine by different SARS-CoV-2
251 variants. The neutralization IC₅₀ from individual donors vaccinated by BNT162b2 (yellow),
252 mRNA-1273 (pink), Ad26.COVS2.S (black) is shown. Significance is based on two-sided
253 t-test.

254

255 **Figure 2. Neutralization of variant spike protein pseudotyped viruses by**
256 **monoclonal antibodies and sACE2.**

257 (A) Infectivity of virus pseudotyped by variant and D614G spike proteins. Viruses were
258 normalized for RT activity and applied to target cells. Infectivity of viruses pseudotyped
259 with the variant proteins or the individual Lambda mutations were tested on ACE2.293T.
260 Luciferase activity was measured two days post-infection. Significance was based on two-
261 sided t-test.

262 (B) Neutralization of variant spike protein variants by sACE2. Viruses pseudotyped with
263 variant spike proteins were incubated with a serially diluted recombinant sACE2 and then
264 applied to ACE2.293T cells. Each plot represents the percent infectivity of D614G and
265 other mutated spike pseudotyped virus. The diagram shows the IC₅₀ for each curve.

266 (C) Nickel beads were coated for 1 hour with 1, 0.5 and 0.1 μ g of sACE2 proteins.
267 Unbound protein was removed and SARS-CoV-2 variant pseudotyped virions (D614G,
268 Delta, Lambda) were incubated with the beads. After 1 hour, the bound virions were
269 analyzed on an immunoblot with antibody p24 antibody. Beads-bound p24 (ng) was
270 calculated and indicated in the bottom (left). Input virions were analyzed on an
271 immunoblot with anti-p24 antibody (middle). Input sACE2 proteins were analyzed on an
272 immunoblot with anti-His-tag antibody (right).

273 (D) Neutralization of Beta, Delta, Delta plus and Lambda variant spike protein variants by
274 REGN10933 and REGN10987 monoclonal antibodies. Neutralization of D614G and
275 variant pseudotyped viruses by REGN10933 (left), REGN10987 (middle), and 1:1 ratio of
276 REGN10933 and REGN10987 (right). The IC₅₀ values of REGN10933, REGN10987 and
277 the cocktail is shown in the table.

278

279

280 **Supplemental Figure S1.**

281 **The structure of variant spikes and immunoblot analysis of spike proteins.**

282 (A) The domain structure of the SARS-CoV-2 spike is diagrammed with Delta (B.1.617.2),
283 Delta plus (AY.1), Lambda (C.37) variant amino acid residues indicated. NTD, N-terminal
284 domain; RBD, receptor-binding domain; RBM, receptor-binding motif; SD1 subdomain 1;
285 SD2, subdomain 2; CS, cleavage site; FP, fusion peptide; HR1, heptad repeat 1; HR2,
286 heptad repeat 2; TM, transmembrane region; IC, intracellular domain. Key mutations are
287 shown in 3D structure (top view).

288 (B) Immunoblot analysis of the Delta (B.1.617.2), Delta plus (AY.1), single point mutated
289 of Lambda (C.37) variant, Lambda (C.37) variant spike proteins in transfected 293T cells.
290 Pseudotyped viruses were produced by transfection of 293T cells. Two days post-
291 transfection, virions were analyzed on an immunoblot probed with anti-spike antibody and
292 anti-HIV-1 p24. The cell lysates were probed with anti-spike antibody and anti-GAPDH
293 antibodies as a loading control.

294

295 **Supplemental Figure S2.**

296 **Neutralization titers of spike protein pseudotyped viruses (single point mutations)**
297 **by convalescent sera, antibodies elicited by BNT162b2, mRNA-1273.**

298 (A) Neutralization of variant spike protein (single point mutations) pseudotyped viruses by
299 convalescent serum (n=8). Dots represent the IC₅₀ of single donors.

300 (B) Neutralizing titers of serum samples from BNT162b2 vaccinated individuals (n=15).
301 The serum was collected at early time point (7 days after second immunization). Each dot
302 represents the IC₅₀ for a single donor.

303 (C) Neutralizing titers of serum samples from mRNA-1273 vaccinated donors (n=6). The
304 serum was collected at early time point (7 days after second immunization). The
305 neutralization IC₅₀ from individual donors is shown. Significance is based on the two-sided
306 t-test.

307

308 **Supplemental Figure S3.**

309 **Neutralization titers of spike protein pseudotyped viruses (single point mutations)** 310 **by monoclonal antibodies.**

311 Neutralization of variant spike protein variants (single point mutations) by REGN10933
312 and REGN10987 monoclonal antibodies. The IC₅₀ of REGN10933, REGN10987 and the
313 cocktail is shown in the table.

314

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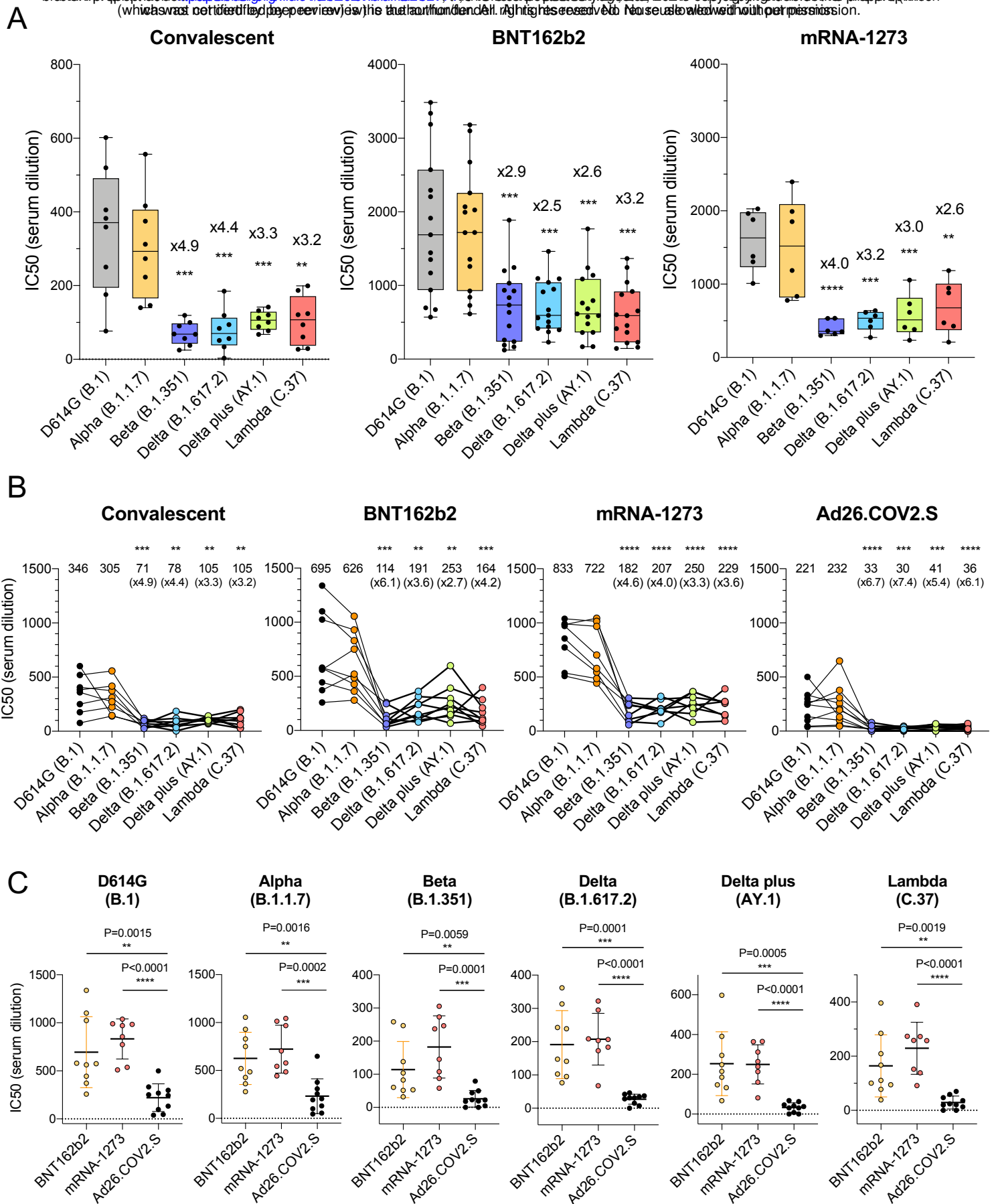


Figure 1

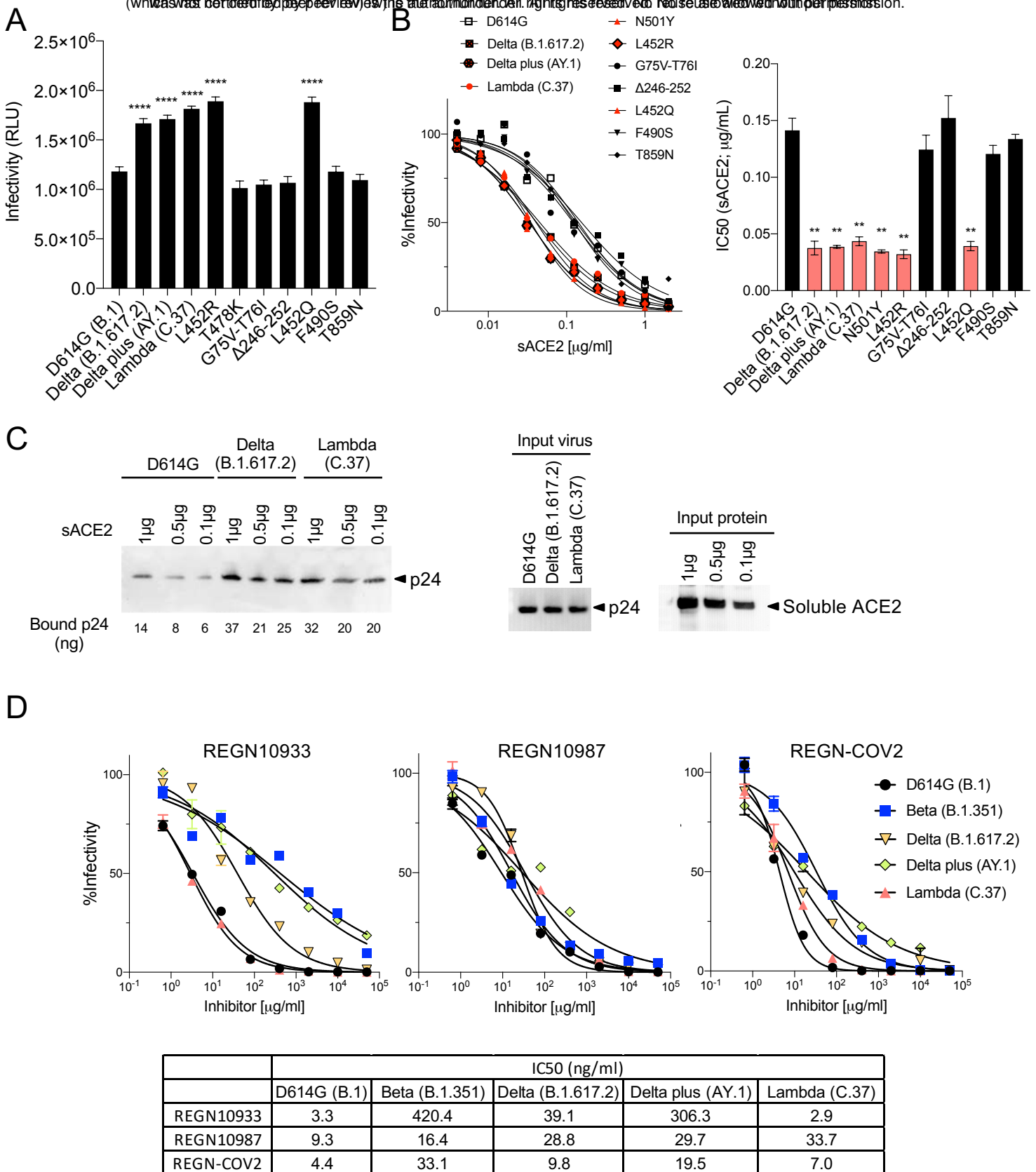
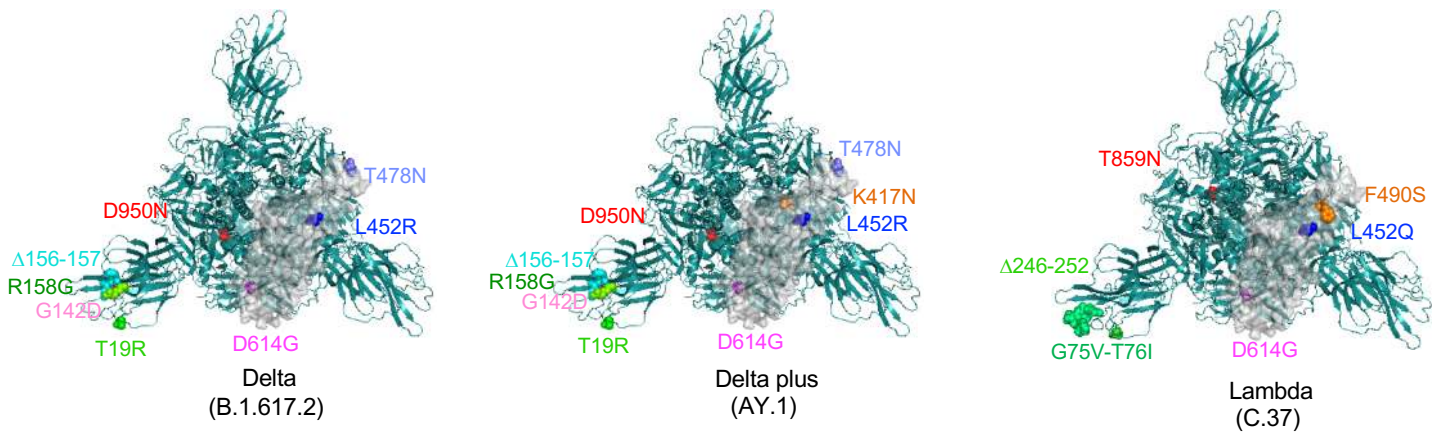
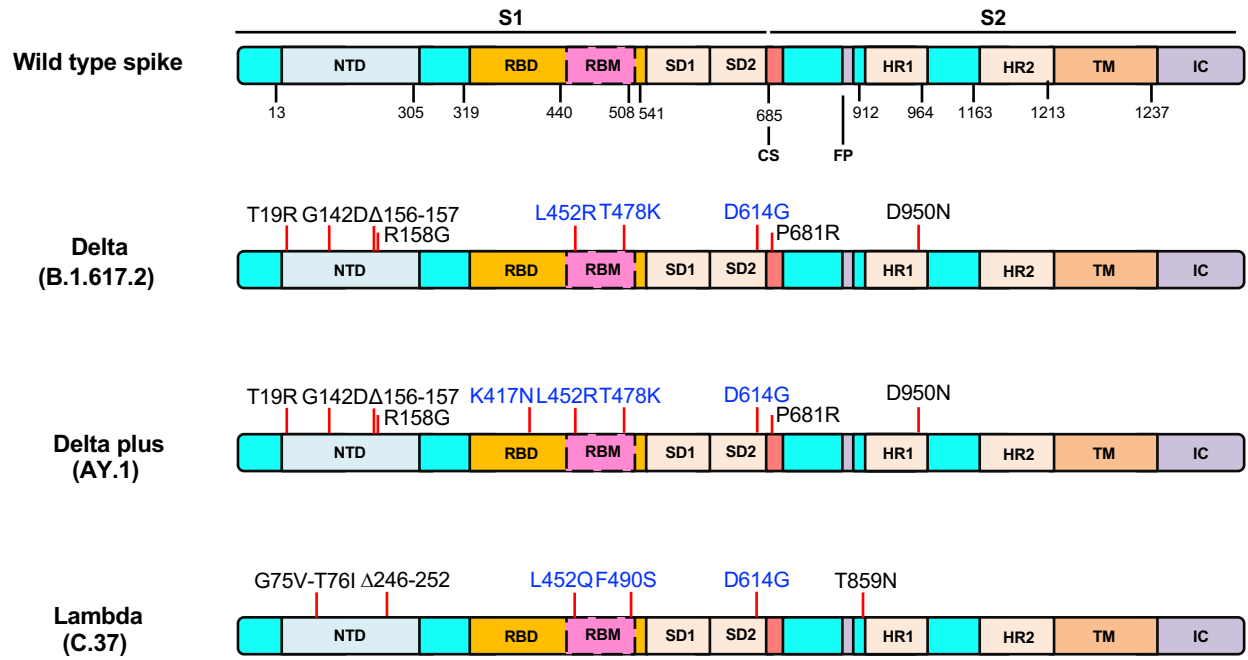
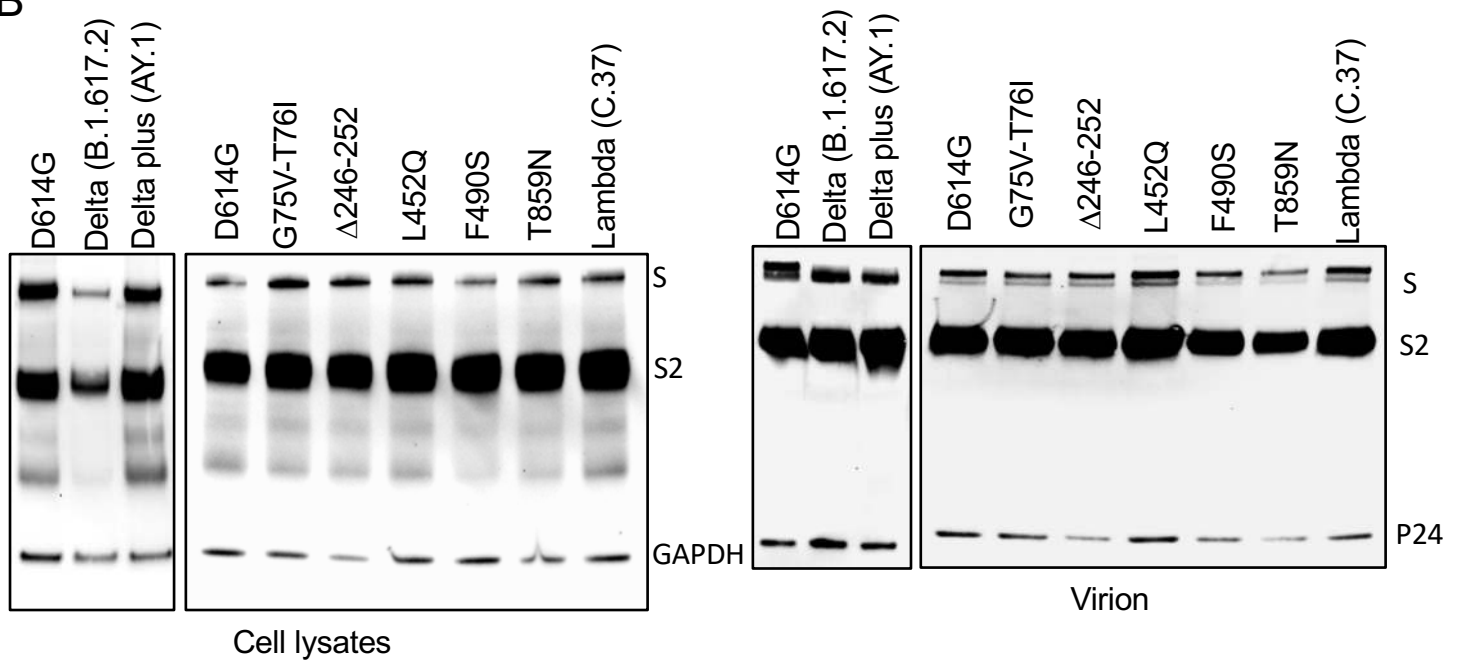


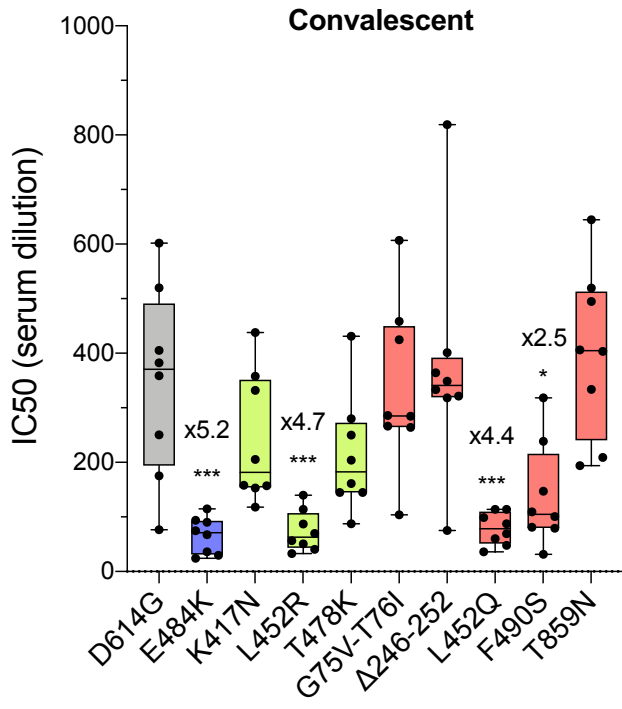
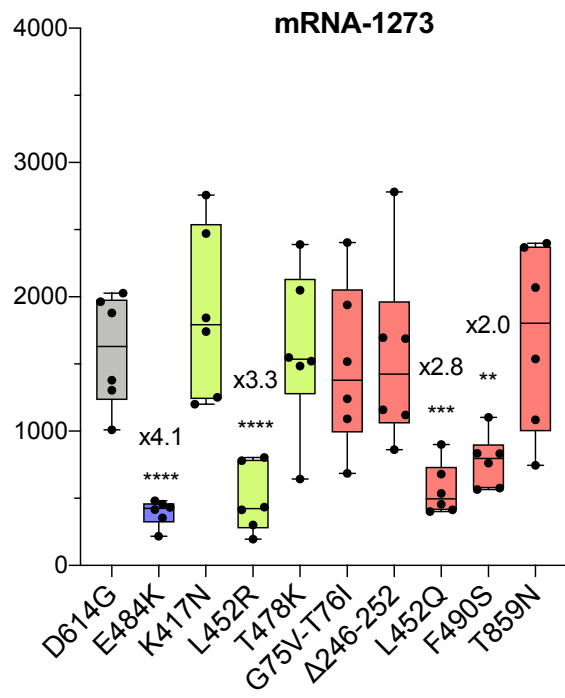
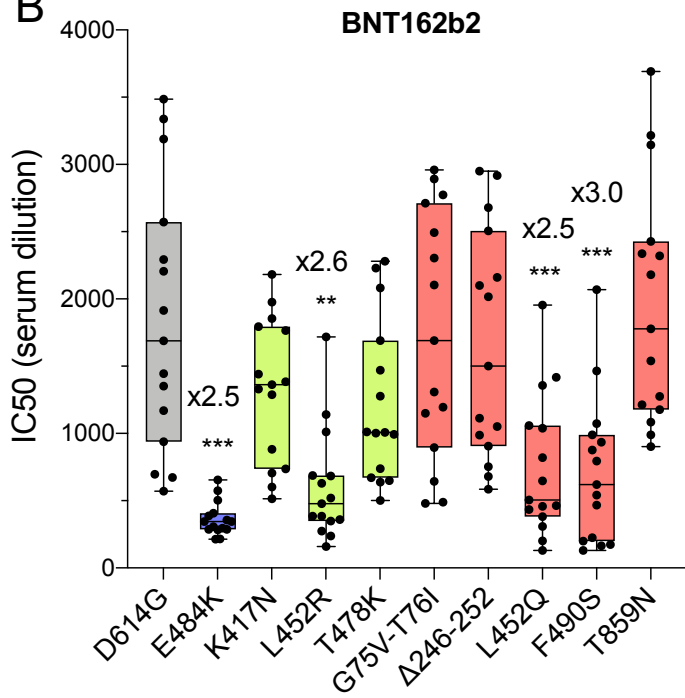
Figure 2

A

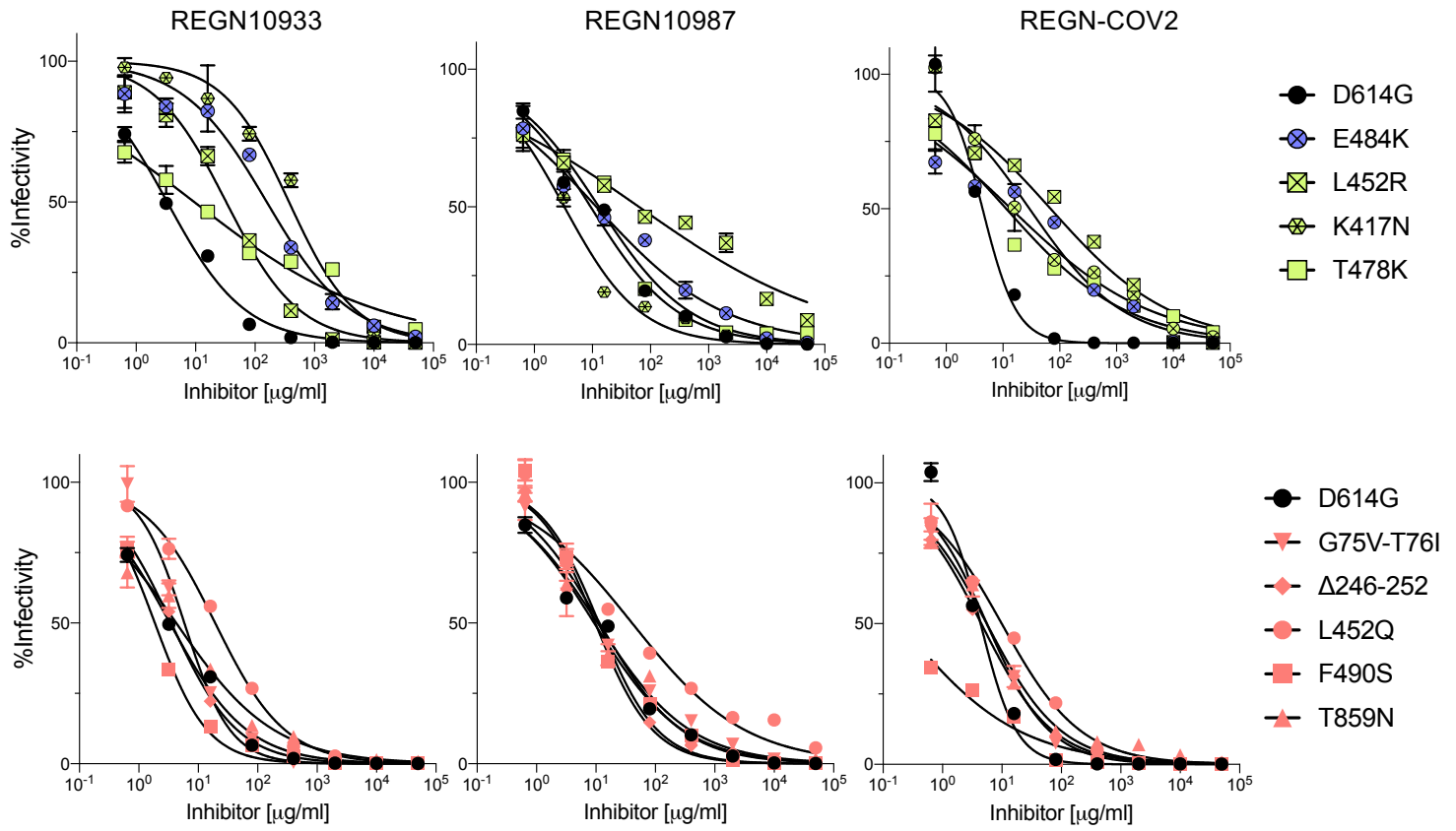


B



A**B**

A



| | IC50 (ng/ml) | | | | | | | | | |
|-----------|--------------|-----------|----------|-------|-------|-------|-------|-------|-------|-------|
| | D614G | G75V-T76I | Δ246-252 | L452Q | F490S | T859N | E484K | L452R | K417N | T478K |
| REGN10933 | 3.3 | 5.8 | 3.5 | 19.3 | 1.9 | 4.3 | 157.6 | 32.7 | 373.3 | 9.5 |
| REGN10987 | 9.3 | 10.7 | 9.7 | 40.5 | 11.2 | 11.5 | 11.7 | 63.3 | 3.3 | 13.0 |
| REGN-COV2 | 4.4 | 5.6 | 4.5 | 9.9 | 0.2 | 5.5 | 13.9 | 69.0 | 29.3 | 11.4 |

Table S1. Neutralization of variants by convalescent sera, BNT162b2 and mRNA-1273 elicited antibodies
7 days post-second vaccination.

| | Convalescent | | | | | |
|--------------|-----------------------------------|--------------------|-------------------|----------------------|----------------------|------------------|
| | IC ₅₀ (serum dilution) | | | | | |
| donor | D614G (B.1) | Alpha (B.1.1.7) | Beta (B.1.351) | Delta (B.1.617.2) | Delta plus (AY.1) | Lambda (C.37) |
| 1 | 251 | 312 | 69 | 56 | 112 | 94 |
| 2 | 176 | 223 | 91 | 84 | 121 | 124 |
| 3 | 77 | 140 | 68 | 185 | 132 | 29 |
| 4 | 406 | 375 | 38 | 51 | 68 | 121 |
| 5 | 602 | 146 | 57 | 3 | 77 | 27 |
| 6 | 383 | 416 | 119 | 34 | 142 | 199 |
| 7 | 520 | 556 | 100 | 119 | 89 | 187 |
| 8 | 359 | 273 | 25 | 94 | 100 | 61 |
| Mean (SD) | 346 (174) | 305 (142) | 71 (32) | 78 (56) | 105 (26) | 105 (66) |

| | | BNT162b2 | | | | | |
|-----------|------------------------|-----------------------------------|-----------------|----------------|-------------------|-------------------|---------------|
| | | IC ₅₀ (serum dilution) | | | | | |
| donor | Days post last vaccine | D614G (B.1) | Alpha (B.1.1.7) | Beta (B.1.351) | Delta (B.1.617.2) | Delta plus (AY.1) | Lambda (C.37) |
| 1 | 7 | 1915 | 1994 | 877 | 914 | 575 | 834 |
| 2 | 7 | 697 | 615 | 228 | 231 | 169 | 191 |
| 3 | 7 | 2572 | 2026 | 1366 | 950 | 1088 | 1244 |
| 4 | 7 | 939 | 925 | 145 | 507 | 171 | 123 |
| 5 | 7 | 1445 | 1717 | 161 | 416 | 361 | 167 |
| 6 | 7 | 2205 | 2069 | 413 | 370 | 614 | 935 |
| 7 | 7 | 1689 | 1259 | 918 | 560 | 1769 | 735 |
| 8 | 7 | 3189 | 2676 | 1045 | 1095 | 762 | 1032 |
| 9 | 7 | 1352 | 1720 | 456 | 594 | 363 | 451 |
| 10 | 7 | 1170 | 1355 | 604 | 669 | 796 | 635 |
| 11 | 7 | 672 | 729 | 219 | 398 | 592 | 238 |
| 12 | 7 | 571 | 841 | 364 | 441 | 480 | 259 |
| 13 | 7 | 3338 | 3099 | 1245 | 1463 | 1241 | 926 |
| 14 | 7 | 3486 | 3181 | 591 | 1042 | 685 | 1200 |
| 15 | 7 | 2294 | 2257 | 654 | 1092 | 1138 | 1888 |
| Mean (SD) | | 1835 (986) | 1764 (822) | 619 (394) | 716 (354) | 720 (436) | 724 (502) |

| | | mRNA-1273 | | | | | |
|-----------|------------------------|-----------------------------------|-----------------|----------------|-------------------|-------------------|---------------|
| | | IC ₅₀ (serum dilution) | | | | | |
| donor | Days post last vaccine | D614G (B.1) | Alpha (B.1.1.7) | Beta (B.1.351) | Delta (B.1.617.2) | Delta plus (AY.1) | Lambda (C.37) |
| 1 | 7 | 1380 | 1186 | 532 | 500 | 382 | 472 |
| 2 | 7 | 1963 | 1852 | 362 | 614 | 731 | 1185 |
| 3 | 7 | 1010 | 833 | 351 | 273 | 1055 | 209 |
| 4 | 7 | 1305 | 779 | 298 | 419 | 234 | 427 |
| 5 | 7 | 1879 | 2395 | 535 | 638 | 411 | 880 |
| 6 | 7 | 2028 | 1990 | 322 | 568 | 615 | 946 |
| Mean (SD) | | 1594 (419) | 1506 (668) | 400 (106) | 502 (138) | 571 (296) | 687 (373) |

Table S2. Neutralization of viruses by sera from BNT162b2, mRNA-1273 and Ad26.COVS vaccinated individuals.

| BNT162b2 | | | | | | | | | | | |
|-----------------|------------------------|--------------|-----|-----|---------------------------|-----------------------------------|-----------|----------|-----------|------------|-----------|
| donor | Days post last vaccine | Anti-N ELISA | Age | Sex | Comorbidities | IC ₅₀ (serum dilution) | | | | | |
| | | | | | | D614G | Alpha | Beta | Delta | Delta plus | Lambda |
| 1 | 84 | - | 39 | F | None | 575 | 427 | 51 | 141 | 215 | 167 |
| 2 | 52 | - | 23 | F | None | 1338 | 1055 | 82 | 314 | 296 | 101 |
| 3 | 101 | - | 26 | F | Asthma | 1101 | 829 | 258 | 362 | 598 | 209 |
| 4 | 109 | - | 33 | F | None | 562 | 750 | 138 | 243 | 186 | 111 |
| 5 | 60 | - | 35 | F | Hypothyroidism, Psoriasis | 1024 | 930 | 53 | 239 | 391 | 284 |
| 6 | 81 | - | 42 | F | Asthma | 258 | 279 | 32 | 103 | 248 | 39 |
| 7 | 108 | - | 26 | F | None | 580 | 485 | 247 | 95 | 133 | 396 |
| 8 | 107 | - | 24 | M | None | 372 | 520 | 104 | 77 | 147 | 78 |
| 9 | 110 | - | 35 | M | None | 445 | 362 | 60 | 148 | 67 | 95 |
| Mean (SD) | 90 (22) | | 31 | | | 695 (369) | 626 (272) | 114 (85) | 191 (102) | 253 (161) | 164 (114) |

| mRNA-1273 | | | | | | | | | | | |
|------------------|------------------------|--------------|-----|-----|---------------|-----------------------------------|-----------|----------|----------|------------|----------|
| donor | Days post last vaccine | Anti-N ELISA | Age | Sex | Comorbidities | IC ₅₀ (serum dilution) | | | | | |
| | | | | | | D614G | Alpha | Beta | Delta | Delta plus | Lambda |
| 1 | 89 | - | 26 | M | None | 984 | 1043 | 108 | 173 | 364 | 257 |
| 2 | 92 | - | 53 | M | None | 972 | 703 | 237 | 207 | 239 | 273 |
| 3 | 61 | - | 67 | M | Prediabetes | 774 | 544 | 87 | 68 | 264 | 139 |
| 4 | 93 | - | 33 | F | None | 509 | 443 | 58 | 209 | 82 | 91 |
| 5 | 44 | - | 32 | M | None | 856 | 579 | 273 | 203 | 365 | 258 |
| 6 | 100 | - | 29 | F | None | 1038 | 1014 | 305 | 295 | 312 | 274 |
| 7 | 52 | - | 33 | F | None | 990 | 968 | 145 | 322 | 213 | 152 |
| 8 | 105 | - | 55 | F | Asthma | 537 | 485 | 246 | 184 | 160 | 391 |
| Mean (SD) | 80 (24) | | 41 | | | 833 (209) | 722 (249) | 182 (94) | 208 (77) | 250 (99) | 229 (96) |

| Ad26.COVS | | | | | | | | | | | |
|------------------|------------------------|--------------|-----|-----|---------------|-----------------------------------|-----------|---------|---------|------------|---------|
| donor | Days post last vaccine | Anti-N ELISA | Age | Sex | Comorbidities | IC ₅₀ (serum dilution) | | | | | |
| | | | | | | D614G | Alpha | Beta | Delta | Delta plus | Lambda |
| 1 | 57 | - | 42 | F | None | 46 | 55 | 22 | 31 | 41 | 21 |
| 2 | 58 | - | 28 | F | None | 133 | 101 | 5 | 28 | 46 | 47 |
| 3 | 66 | - | 36 | F | None | 500 | 130 | ND | ND | ND | ND |
| 4 | 92 | - | 33 | F | None | 333 | 257 | 23 | 31 | 8 | 24 |
| 5 | 87 | - | 39 | F | Prediabetes | 244 | 205 | 19 | 42 | 31 | 36 |
| 6 | 72 | - | 32 | M | None | 268 | 308 | 79 | 34 | 63 | 59 |
| 7 | 92 | - | 39 | F | None | 251 | 377 | 44 | 46 | 38 | 70 |
| 8 | 71 | - | 75 | F | None | 298 | 648 | ND | 7 | ND | ND |
| 9 | 105 | - | 30 | M | None | 38 | 45 | 18 | 37 | 31 | 13 |
| 10 | 115 | - | 33 | F | None | 98 | 194 | 50 | 15 | 68 | 20 |
| Mean (SD) | 82 (20) | | 39 | | | 221 (144) | 232 (182) | 33 (24) | 30 (12) | 41 (19) | 36 (21) |