Supplemental information

Immunogenicity of a third dose with mRNA-vaccines

in the ChAdOx1-S/BNT162b2 vaccination

regimen against SARS-CoV-2 variants

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Data S1. Figures S1-S3 and Tables S1-S16.

Figure S1. Humoral immune response by age, related to figures 2 and 4. Group A represents participants aged 18-49 years and group B those aged 50-59 years. Total anti-RBD Igs were expressed as BAU/ml and variant-specific neutralization capacity as NT50. Data are represented as median and interquartile range.
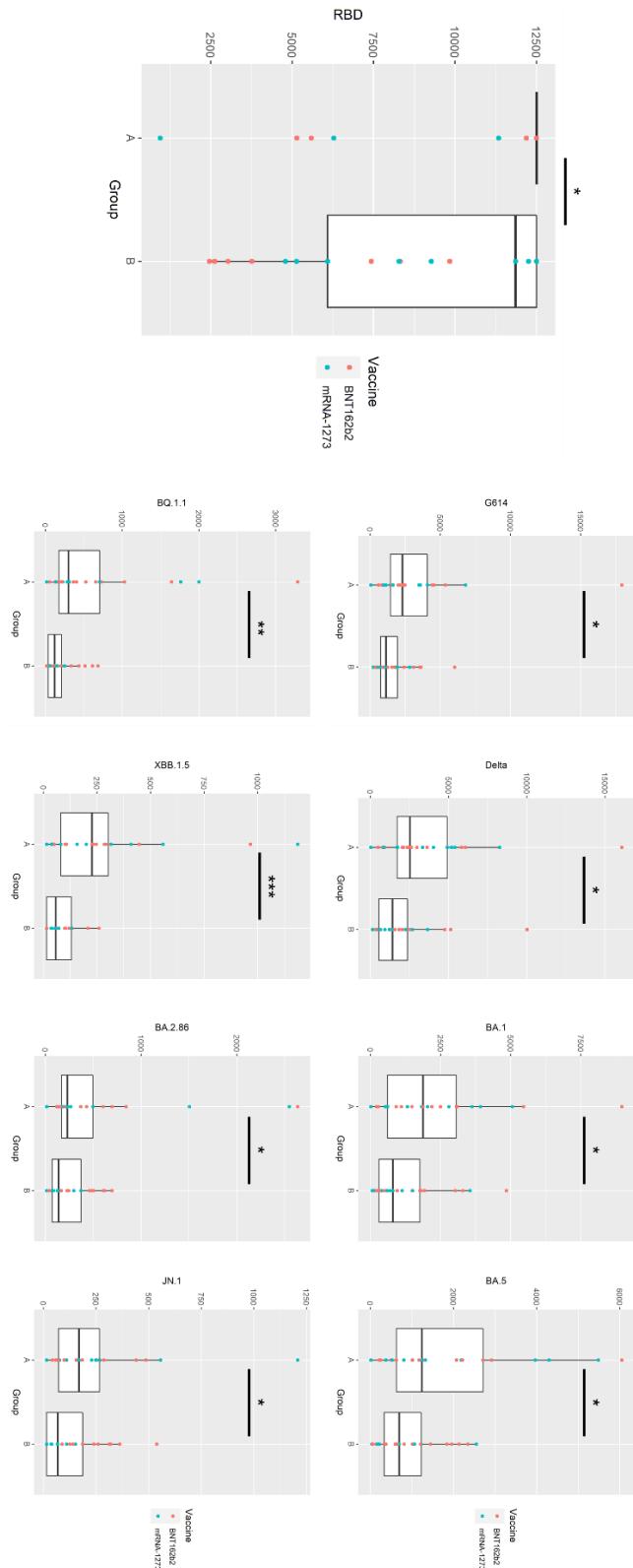
Figure S2. Humoral immune response by sex, related to figures 2 and 4. Total anti-RBD Igs were expressed as BAU/ml and variant-specific neutralization capacity as NT50. Data are represented as median and interquartile range.
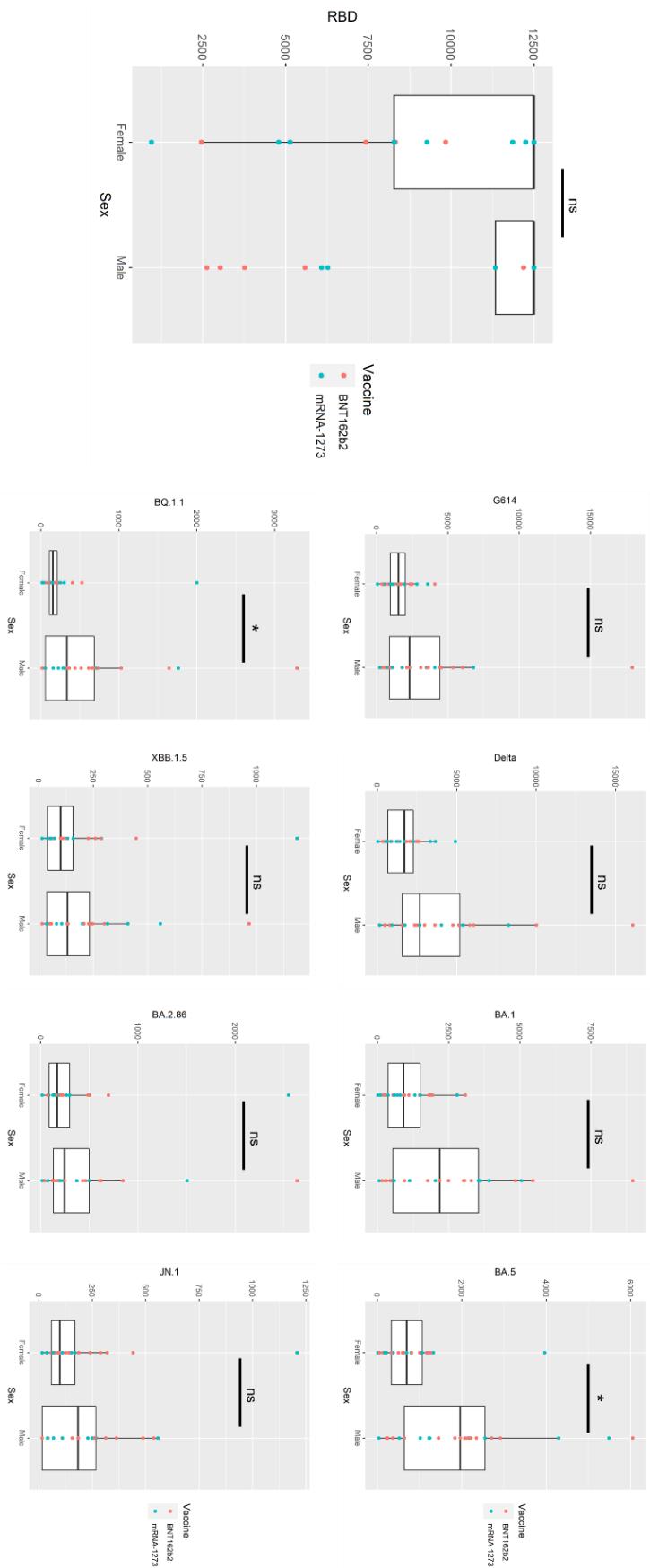
Figure S3. Heatmap showing the prevalence of the mutations of the lineages used in this study according to GISAID database, related to STAR Methods.
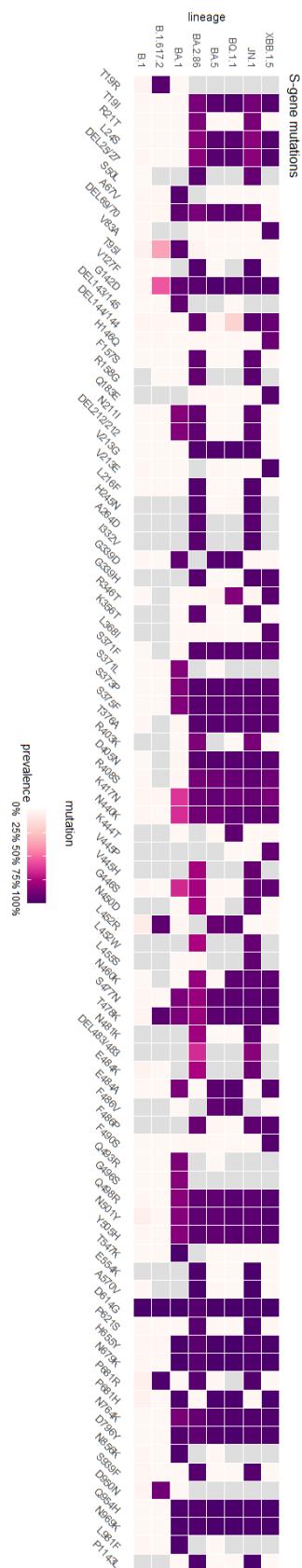
Table S1. Summary statistics for SARS-CoV-2 anti-spike Ig response comparing BNT162b2 and mRNA-1273 arms in participants with a negative result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	33	1381.9[990.0;1929.0]	28	1154.7[754.3;1767.6]	0.307
day 28	30	12220.6[11896.8;12553.1]	26	12243.3[11861.7;12637.2]	0.903
day 90	24	9157.7[7813.6;10733.1]	22	10431.8[9158.3;11882.3]	0.184
day 180	11	6613.1[3687.5;11859.9]	10	10302.9[8156.7;13013.8]	0.138

Units: BAU/mL

Table S2. Comparative neutralization of SARS-CoV-2 ancentral G614 variant in BNT162b2 and mRNA-1273 arms in participants with a negative result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	33	113.7[75.0;172.4]	28	129.4[73.0;229.5]	0.728
day 28	30	1929.0[1499.7;2481.3]	26	2693.3[2025.8;3580.7]	0.080
day 90	24	1010.7[707.3;1444.3]	22	1794.9[1223.9;2632.4]	0.038
day 180	11	980.2[405.9;2367.0]	10	1522.0[771.7;3002.0]	0.597

Units: neutralising titer 50 (NT_{50})

Table S3. Comparative neutralization of SARS-CoV-2 Delta variant (B.1.617.2 lineage) in BNT162b2 and mRNA-1273 arms in participants with a negative result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	33	74.0[45.5;120.4]	28	116.1[63.8;211.3]	0.330
day 28	30	2027.1[1539.5;2669.1]	26	2926.7[2196.2;3900.0]	0.022
day 90	24	852.3[538.6;1348.7]	22	1500.2[934.2;2409.3]	0.073
day 180	11	811.8[291.7;2259.1]	10	1525.1[615.4;3779.6]	0.342

Units: neutralising titer 50 (NT_{50})

Table S4. Comparative neutralization of SARS-CoV-2 Omicron variant (BA.1 lineage) in BNT162b2 and mRNA-1273 arms in participants with a negative result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	33	36.1[22.4;58.3]	28	42.2[22.2;80.1]	0.839
day 28	30	725.0[493.4;1065.2]	26	1368.3[934.9;2002.6]	0.009
day 90	24	508.4[327.0;790.5]	22	904.9[534.4;1532.4]	0.106
day 180	11	471.1[150.4;1475.6]	10	852.4[344.3;2110.2]	0.342

Units: neutralising titer 50 (NT_{50})

Table S5. Summary statistics for SARS-CoV-2 anti-spike Ig response comparing BNT162b2 and mRNA-1273 arms in participants with a positive result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

		BNT162b2		mRNA-1273	p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	8	9764.0[6851.6;13914.5]	6	9630.4[6148.9;15083.2]	0.830
day 28	10	12326.2[11942.0;12722.8]	7	12500[12500;12500]	0.473
day 90	13	10685.5[8788.5;12992.0]	9	11631.8[10324.9;13104.1]	0.640
day 180	18	9567.8[7467.6;12258.8]	11	10755.2[8739.9;13235.2]	0.652

Units: BAU/mL

Table S6. Comparative neutralization of SARS-CoV-2 ancestral G614 variant in BNT162b2 and mRNA-1273 arms in participants with a positive result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

		BNT162b2		mRNA-1273	p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	8	1530.9[709.0;3305.3]	6	1054.1[436.4;2546.4]	0.561
day 28	10	4458.6[3199.8;6212.5]	7	5192.5[2981.1;9044.2]	0.526
day 90	13	2435.5[1494.1;3970.0]	9	3422.3[1558.0;7517.1]	0.841
day 180	18	2123.9[1363.7;3307.6]	11	1624.1[1015.8;2596.7]	0.432

Units: neutralising titer 50 (NT_{50})

Table S7. Comparative neutralization of SARS-CoV-2 Delta variant (B.1.617.2 lineage) in BNT162b2 and mRNA-1273 arms in participants with a positive result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	8	2203.0[1227.3;3954.4]	6	1239.9[492.0;3124.9]	0.175
day 28	10	5512.8[4261.2;7132.1]	7	5347.3[2973.3;9616.7]	0.733
day 90	13	2633.7[1818.2;3815.2]	9	2897.0[1922.1;4366.2]	0.841
day 180	18	2694.9[1762.9;4119.7]	11	1699.5[1082.1;2669.0]	0.157

Units: neutralising titer 50 (NT_{50})

Table S8. Comparative neutralization of SARS-CoV-2 Omicron variant (BA.1 lineage) in BNT162b2 and mRNA-1273 arms in participants with a positive result of IgG to SARS-CoV-2 nucleocapsid, related to Figure 2.

	BNT162b2		mRNA-1273		p-value
	n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
day 0	8	1390.8[738.8;2618.1]	6	1123.9[395.7;3192.2]	0.948
day 28	10	3019.9[1963.0;4645.9]	7	3338.2[1807.1;6166.9]	0.733
day 90	13	1825.2[1150.7;2895.0]	9	1634.4[1052.2;2538.6]	0.504
day 180	18	1480.3[880.8;2487.9]	11	1258.3[735.5;2152.8]	0.544

Units: neutralising titer 50 (NT_{50})

Table S9. Comparative humoral immune response against SARS-CoV-2 variants G614, Delta, and BA.1 between participants with a negative and positive result of IgG to SARS-CoV-2 nucleocapsid in BNT162b2 arm, related to Figure 2.

		N-*		N+**		p-value
		n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
Day 0	**RBD-Igs**	33	1381.9 [990.0; 1929.0]	8	9764.0 [6851.6- 13914.5]	6.3×10^{-5}
	G614	33	113.7 [75.0; 172.4]	8	1530.9 [709.0; 3305.3]	5.6×10^{-5}
	Delta	33	74.0 [45.5; 120.4]	8	2203.0 [1227.3; 3954.4]	3.1×10^{-5}
	BA.1	33	36.1 [22.4; 58.3]	8	1390.8 [738.8; 2618.1]	1.6×10^{-5}
Day 28	**RBD-Igs**	30	12220.6 [11896.8; 12553.2]	10	12326.2 [11942.0; 12722.8]	0.952
	G614	30	1929.0 [1499.7; 2481.3]	10	4458.6 [3199.8; 6212.5]	4.3×10^{-4}
	Delta	30	2027.1 [1539.5; 2669.1]	10	5512.8 [4261.2; 7132.1]	4.0×10^{-5}
	BA.1	30	725.0 [493.4; 1065.2]	10	3019.9 [1963.0; 4645.9]	4.0×10^{-5}
Day 90	**RBD-Igs**	24	9157.7 [7813.6; 10733.1]	13	10685.5 8788.5; 12992.0]	0.1046
	G614	24	1010.7 [707.3; 1444.3]	13	2435.5 [1494.1; 3970.0]	0.004
	Delta	24	852.3 [538.6; 1348.7]	13	2633.8 [1818.2; 3815.2]	3.2×10^{-4}
	BA.1	24	508.4 [327.0; 790.5]	13	1825.2 [1150.7; 2895.0]	5.0×10^{-4}
Day 180	**RBD-Igs**	11	6613.1 [3687.5; 11859.9]	18	9567.8 [7467.6; 12258.8]	0.092
	G614	11	980.2 [405.9; 2367.0]	18	2123.9 [1363.8; 3307.7]	0.204
	Delta	11	811.8 [291.7; 2259.1]	18	2694.9 [1762.9; 4119.7]	0.032
	BA.1	11	471.1 [150.4; 1475.6]	18	1480.4 [880.8; 2487.9]	0.061

Units: neutralising titer 50 (NT_{50})

*N-: negative result of IgG to SARS-CoV-2 nucleocapsid

**N+: positive result of IgG to SARS-CoV-2 nucleocapsid

Table S10. Comparative humoral immune response against SARS-CoV-2 variants G614, Delta, and BA.1 between participants with a negative and positive result of IgG to SARS-CoV-2 nucleocapsid in mRNA-1273 arm, related to Figure 2.

			N-*		N+**	p-value
		n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
Day 0	RBD-Igs	28	1154.7 [754.3; 1767.6]	6	9630.5 [6148.9; 15083.2]	7.4 x 10-4
	G614	28	129.4 [73.0; 229.5]	6	1054.1 [436.4; 2546.4]	0.003
	Delta	28	116.1 [63.8; 211.3]	6	1239.9 [492.0; 3124.9]	0.002
	BA.1	28	42.2 [22.2; 80.1]	6	1123.9 [395.7; 3192.2]	9.1 x 10-4
Day 28	RBD-Igs	26	12243.3 [11861.7; 12637.2]	7	12500.0 [12500.0; 12500.0]	0.377
	G614	26	2693.3 [2025.8; 3580.7]	7	5192.5 [2981.2; 9044.2]	0.043
	Delta	26	2926.7 [2196.2; 3900.0]	7	5347.3 [2973.3; 9616.7]	0.099
	BA.1	26	1368.3 [934.9; 2002.6]	7	3338.3 [1807.1; 6166.9]	0.043
Day 90	RBD-Igs	22	10431.8 [9158.3; 11882.3]	9	11631.8 [10324.9; 13104.1]	0.244
	G614	22	1795.0 [1223.9; 2632.4]	9	3422.3 [1558.0; 7517.1]	0.094
	Delta	22	1500.2 [934.2; 2409.3]	9	2897.0 [1922.1; 4366.2]	0.023
	BA.1	22	904.9 [534.4; 1532.4]	9	1634.4 [1052.2; 2538.6]	0.078
Day 180	RBD-Igs	10	10302.9 [8156.7; 13013.8]	11	10755.2 [8740.0; 13235.2]	0.717
	G614	10	1522.0 [771.7; 3002.0]	11	1624.1 [1015.8; 2596.7]	0.918
	Delta	10	1525.1 [615.4; 3779.6]	11	1699.5 [1082.1; 2669.0]	0.972
	BA.1	10	852.4 [344.3; 2110.2]	11	1258.3 [735.5; 2152.8]	0.704

Units: neutralising titer 50 (NT_{50})

*N-: negative result of IgG to SARS-CoV-2 nucleocapsid

**N+: positive result of IgG to SARS-CoV-2 nucleocapsid

Table S11. Comparative neutralization of SARS-CoV-2 Omicron sublineages (BA4/BA.5, BQ.1.1, and XBB.1.5/XBB.1.9) in BNT162b2 and mRNA-1273 arms at day 180, related to Figure 4.

			BNT162b2		mRNA-1273	
		n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	**p-value**
N-*	**BA.4/BA.5**	11	317.7 [117.9; 855.8]	10	648.4 [257.0; 1636.1]	0.245
	BQ.1.1	11	76.8 [32.5; 181.5]	10	166.6 [72.4; 383.1]	0.289
	XBB.1.5/XBB.1.9	11	36.9 [18.4; 74.0]	10	66.8 [35.1; 127.1]	0.180
	BA.2.86	11	101.5 [39.1; 263.2]	10	156.4 [59.8; 408.9]	0.457
	JN.1	11	47.3 [25.0; 89.4]	10	90.6 [30.6; 268.6]	0.205
N+**	**BA.4/BA.5**	18	1327.8 [778.8; 2263.7]	11	970.1 [502.3; 1873.6]	0.514
	BQ.1.1	18	263.9 [136.6; 510.0]	11	199.6 [64.2; 620.7]	0.574
	XBB.1.5/XBB.1.9	18	150.1 [84.6; 266.1]	11	117.9 [51.8; 268.3]	0.458
	BA.2.86	18	249.9 [137.7; 453.4]	11	245.7 [103.8; 581.7]	0.928
	JN.1	18	123.3 [68.9; 220.7]	11	129.6 [59.8; 281.2]	0.822

Units: neutralising titer 50 (NT_{50})

*N-: negative result of IgG to SARS-CoV-2 nucleocapsid

**N+: positive result of IgG to SARS-CoV-2 nucleocapsid

Table S12. Comparative neutralization of SARS-CoV-2 Omicron sublineages (BA4/BA.5, BQ.1.1, and XBB.1.5/XBB.1.9) between N- and N+ subjects in BNT162b2 and mRNA-1273 arms at day 180, related to Figure 4.

			N-*		N+**	p-value
		n	Geometric mean [95% CI]	n	Geometric mean [95% CI]	
BNT162b2	**BA.4/BA.5**	11	317.7 [117.9; 855.8]	10	1327.8 [778.8; 2263.7]	0.004
	BQ.1.1	11	76.8 [32.5; 181.5]	10	263.9 [136.6; 510.0]	0.026
	XBB.1.5/XBB.1.9	11	36.9 [18.4; 74.0]	10	150.1 [84.6; 266.1]	0.005
	BA.2.86	11	101.5 [39.1; 263.2]	10	249.9 [137.7; 453.4]	0.137
	JN.1	11	47.3 [25.0; 89.4]	10	123.3 [68.9; 220.7]	0.032
mRNA-1273	**BA.4/BA.5**	18	648.4 [257.0; 1636.1]	11	970.1 [502.3; 1873.6]	0.605
	BQ.1.1	18	166.6 [72.4; 383.1]	11	199.6 [64.2; 620.7]	0.860
	XBB.1.5/XBB.1.9	18	66.8 [35.1; 127.1]	11	117.9 [51.8; 268.3]	0.341
	BA.2.86	18	156.4 [59.8; 408.9]	11	245.7 [103.8; 581.7]	0.647
	JN.1	18	90.6 [30.6; 268.6]	11	129.6 [59.8; 281.2]	0.972

Units: neutralising titer 50 (NT_{50})

*N-: negative result of IgG to SARS-CoV-2 nucleocapsid

**N+: positive result of IgG to SARS-CoV-2 nucleocapsid

Table S13. Neutralizing response by NT_{50} categories against different SARS-CoV-2 variants in N- subjects of BNT162b2 arm at day 180, related to Figure 4.

	BNT162b2 (n=11)															
	G614		Delta		BA.1		BA.5		BQ.1.1		XBB.1.5		BA.2.86		JN.1	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
<100, n (%)	1	9.1	1	9.1	1	9.1	2	18.2	6	54.5	8	72.7	6	54.5	8	72.7
>100 to <300, n (%)	1	9.1	1	9.1	4	36.4	2	18.2	4	36.4	3	27.3	2	18.2	3	27.3
>300 to <1000, n (%)	3	27.3	4	36.4	3	27.3	5	45.5	1	9.1	0	0.0	3	27.3	0	0.0
≥1000, n (%)	6	54.5	5	45.5	3	27.3	2	18.2	0	0.0	0	0.0	0	0.0	0	0.0

Table S14. Neutralizing response by NT_{50} categories against different SARS-CoV-2 variants in N- subjects of mRNA-1273 arm at day 180, related to Figure 4.

	mRNA-1273 (n=10)															
	G614		Delta		BA.1		BA.5		BQ.1.1		XBB.1.5		BA.2.86		JN.1	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
<100, n (%)	0	0.0	0	0.0	1	10.0	1	10.0	2	20.0	5	50.0	2	20.0	4	40.0
>100 to <300, n (%)	1	10.0	1	10.0	1	10.0	1	10.0	4	40.0	5	50.0	5	50.0	3	30.0
>300 to <1000, n (%)	2	20.0	3	30.0	4	40.0	3	30.0	4	40.0	0	0.0	3	30.0	3	30.0
≥1000, n (%)	7	70.0	6	60.0	4	40.0	5	50.0	0	0.0	0	0.0	0	0.0	0	0.0

Table S15. Neutralizing response by NT_{50} categories against different SARS-CoV-2 variants in N+ subjects of BNT162b2 arm at day 180, related to Figure 4.

	BNT162b2 (n=18)															
	G614		Delta		BA.1		BA.5		BQ.1.1		XBB.1.5		BA.2.86		JN.1	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
<100, n (%)	0	0.0	0	0.0	0	0.0	1	5.6	2	11.1	5	27.8	2	11.1	8	44.4
>100 to <300, n (%)	0	0.0	0	0.0	2	11.1	0	0.0	8	44.4	8	44.4	9	50.0	6	33.3
>300 to <1000, n (%)	3	16.7	2	11.1	3	16.7	3	16.7	6	33.3	5	27.8	5	27.8	4	22.2
≥1000, n (%)	15	83.3	16	88.9	13	72.2	14	77.8	2	11.1	0	0.0	2	11.1	0	0.0

Table S16. Neutralizing response by NT_{50} categories against different SARS-CoV-2 variants in N+ subjects of mRNA-1273 arm at day 180, related to Figure 4.

	mRNA-1273 (n=11)															
	G614		Delta		BA.1		BA.5		BQ.1.1		XBB.1.5		BA.2.86		JN.1	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
<100, n (%)	0	0.0	0	0.0	0	0.0	0	0.0	3	27.3	5	45.5	2	18.2	3	27.3
>100 to <300, n (%)	0	0.0	0	0.0	0	0.0	2	18.2	4	36.4	4	36.4	4	36.4	7	63.6
>300 to <1000, n (%)	4	36.4	3	27.3	4	36.4	2	18.2	1	9.1	1	9.1	4	36.4	0	0.0
≥1000, n (%)	7	63.6	8	72.7	7	63.6	7	63.6	3	27.3	1	9.1	1	9.1	1	9.1