SUPPLEMENTAL MATERIALS

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Roche Testing Procedures

The Roche assay was performed as per manufacturer's instructions, at a single laboratory, using the same lot of reagents. Samples with an anti-SARS-CoV-2 concentration above the measuring range (250 U/ml) were diluted by the Roche analyzer with Diluent Universal at 1:10, as per manufacturer's instructions. After dilution by the analyzer, the software automatically utilized the dilution value when calculating the sample concentration, up to the maximum value of 2500 U/ml. Heat inactivation was not performed. For quality control, we used PreciControl Anti-SARS-CoV-2. These were run at least once every 24 hours. Cutoffs were determined automatically by the analyzer software based on calibrated master curves. For calibration, the method is standardized against an internal Roche standard for anti-SARS-CoV-2 provided with the assay. A pre-defined master curve is adapted to the analyzer using the kit calibration reagents. Calibration is preformed once per reagent lot or following servicing of the analyzer.

MSD Testing Procedures

The MSD assay was performed as per the manufacturer's instructions, at a single laboratory. Samples were tested at a 1:5000 dilution, with the diluent provided by the manufacturer (Diluent 100). Heat inactivation was not performed. The assay includes a standard curve based on a reference standard that contains a pre-determined concentration of each antigen. It also includes three serological controls which contain a known concentration of IgG antibodies against the antigens of the assay. We ran the standard curve and serological controls on every plate to assess for quality control.

Variables	Full Cohort n=186	Interval between doses		P-value[1]
		Short Interval (17-28 days) n = 118	Long interval (42-49 days) n = 68	
Age (yr), mean (SD)	39 (10)	39 (9)	40 (10)	0.281
Female sex at birth, n (%)	84 (45)	52 (44)	32 (47)	0.693
Male Sex at birth, n (%)	102 (55)	66 (56)	36 (53)	
Ethnicity/Race[2]**,** n (%)				
White	159 (85)	95 (80)	64 (94)	
Asian	13 (7.0)	10 (8.5)	3 (4.4)	0.183
Other	3 (1.6)	3 (2.5)	0 (0)	
Educational level, n (%)				
Non-university certificate	107 (58)	59 (50)	48 (71)	
University Bachelor's degree	71 (38)	54 (46)	17 (25)	0.018
University Graduate degree	8 (4.3)	5 (4.2)	3 (4.4)	
Smoking History, n (%)				
Cigarette use	11 (5.9)	8 (6.8)	3 (4.4)	0.510
E-cigarette use	7 (3.8)	4 (3.4)	3 (4.4)	0.724
Past Medical History, n (%)				
Hypertension	13 (7.4)	8 (6.8)	5 (7.4)	0.655
Diabetes	2 (1.1)	1 (0.85)	1 (1.4)	0.894
Asthma	29 (16)	19 (16)	10 (15)	0.943
Chronic Lung disease	2 (1.1)	2 (1.7)	0 (0)	-
Chronic Heart Disease	1 (0.54)	1 (0.85)	0 (0)	-
Chronic Kidney disease	1 (0.54)	1 (0.85)	0 (0)	-
Liver disease	4 (2.2)	2 (1.7)	2 (2.9)	-
Malignancy	4 (2.2)	3 (2.5)	1 (1.4)	-
Immune suppressed	6 (3.2)	2 (1.7)	4 (5.9)	-
Mass and Height, mean (SD)				
Weight (kg)	85 (25)	85 (27)	85 (19)	0.928
Height (m)	1.7 (0.10)	1.7 (0.10)	1.7 (0.10)	0.810
BMI (kg/m^2)	30 (8.7)	30 (9.5)	29 (7.1)	0.902
Vaccine, n (%)				
Both BNT162b2	131 (70)	109 (92)	22 (32)	
Both mRNA-1273	54 (29)	9 (7.6)	45 (66)	
BNT162b2 (1st), mRNA-1273 (2nd)	0 (0)	0 (0)	0 (0)	<0.0001
mRNA-1273 (1st), BNT162b2 (2nd)	1 (0.54)	0 (0)	1 (1.47)	
Jan.1/21-to-1st vacc (d),[3] median (IQR)	7 (5-12)	6 (4-14)	8 (7-12)	<0.0001
Vacc dosing interval (d), mean (SD)	30 (10)	23 (2.5)	43 (1.6)	<0.0001
2nd dose-to-BS interval (d), mean (SD)	56 (29)	62 (27)	46 (30)	<0.0001
Serological Testing				
Ortho Reactive, n (%)	186 (100)	118 (100)	68 (100)	0.187
MSD Spike (Au/mL), gM (gSD)	109,821 (3.4)	96,394 (3.1)	137,902 (3.8)	0.001
Roche Spike (U/mL), mean (SD)	1909 (767)	1656 (766)	2346 (542)	<0.0001
MSD RBD (Au/mL), gM (gSD)	71,561 (4.1)	53,157 (4.1)	120,813 (3.7)	<0.0001
MSD NTD (Au/mL), gM (gSD)	1725 (4.6)	1462 (3.1)	2294 (7.6)	<0.0001
MSD N (Au/mL), gM (gSD)	196 (3.7)	152 (3.3)	303 (4.1)	<0.0001

Supplemental Table 1: Participant Characteristics and Serology Results, Overall and Classified by Vaccine Dosing Interval Group

[1] Compared using the Mann-Whitney U-test (serological testing), Pearson chi-square test (ethnicity, educational level, past medical history, sex, smoking history, vaccine type), Wilcoxon signed rank test ("Jan.1-to-1st vaccine"),

and independent sample t-test (age, weight, height, BMI). Ethnicity and education were assessed using 3-level variables

[2] Participants were asked "How would you describe your ethnicity or race?"; "Asian" includes *Southeast Asian, West Asian, Chinese, Japanese, Korean*

[3] Median date of first vaccine Jan. 8, 2021; mean date of first vaccine Jan. 11, 2021

n, number; SD, standard deviation; BMI, body mass index; Vacc, vaccine; BS, blood sampling date; d, day; MSD, Meso Scale Discovery; MSD, RBD, receptor binding domain; NTD, spike N terminal domain; N, nucleocapsid; gM, geographic mean; gSD, geometric standard deviation

Model	P value of interaction term
MSD Spike	0.842
MSD RBD	0.785
MSD NTD	0.300
MSD N	0.767
Roche Spike	0.968

Supplemental Table 2: Secondary Analyses Including an Interaction Terms for Vaccine Type and Dosing Interval Group

Analysis used an interaction term between vaccine-type and dosing interval-group for each model, adjusting for the second vaccine-to-blood sampling interval.

P value shown is for the interaction term.

MSD, Meso Scale Discovery; MSD, RBD, receptor binding domain; NTD, spike N terminal domain; N, nucleocapsid

Antibody tests	Coeff (β)	Test for regression coefficients	
		t-test	P-value
MSD Spike	40638.88	2.210	0.029
MSD RBD	61641.72	3.857	<0.0001
MSD NTD	1608.11	3.580	<0.0001
MSD N	533.93	1.411	0.160
Roche Spike	723.43	7.199	<0.0001

Supplemental Table 3: Results of Regression Model including covariates of Educational Level and Ethnicity/Race

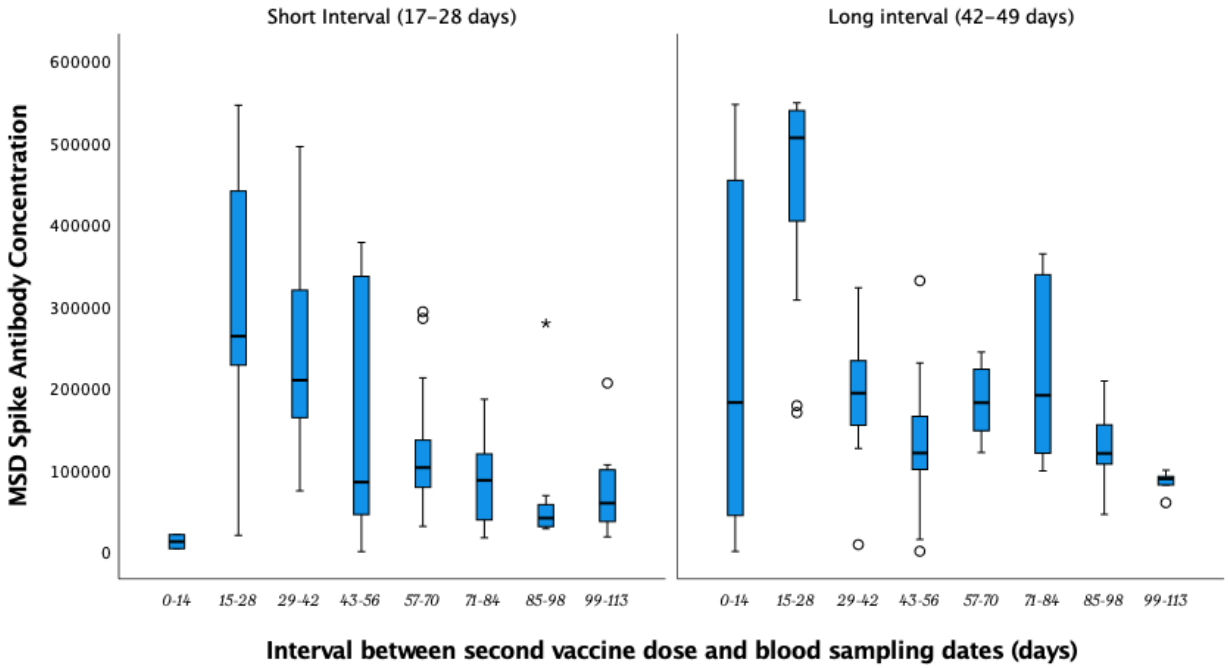
Multiple linear regression for the association between vaccine dosing interval group and quantitative antibodies, adjusted for second vaccine-to-blood sampling interval (days), educational level and race.
Coeff (β): is the regression coefficient of the dosing interval (long interval versus short interval [reference])
For ethnicity/race data, participants were asked "How would you describe your ethnicity or race?"
MSD, Meso Scale Discovery; MSD, RBD, receptor binding domain; NTD, spike N terminal domain; N, nucleocapsid; gM, geographic mean

Antibody tests	BNT162b2 (Pfizer) vaccine (n= 124)				mRNA-1273 (Moderna) vaccine (n=48)			
	Antibody Concentration		Test for regression coefficient		Antibody Concentration		Test for regression coefficient	
	Short Interval, gM	Long Interval, gM	T-test	P-value	Short Interval, gM	Long Interval, gM	T-test	P-value
MSD Spike	97,545	154,498	1.667	0.098	142,524	146,247	0.327	0.745
MSD RBD	54,441	127,073	3.041	0.003	86513	113,242	1.123	0.267
MSD NTD	1487	2,811	3.383	0.001	2392	2086	0.543	0.589
MSD N	147	401	2.580	0.011	251	225	0.020	0.984
Roche Spike	1390	2475	4.457	<0.0001	1991	2471	3.209	<0.0001

Supplemental Table 4: Adjusted Linear Regression models comparing vaccine dosing intervals within strata defined by vaccine type.

Model was adjusted for second vaccine-to-blood sampling interval (days)

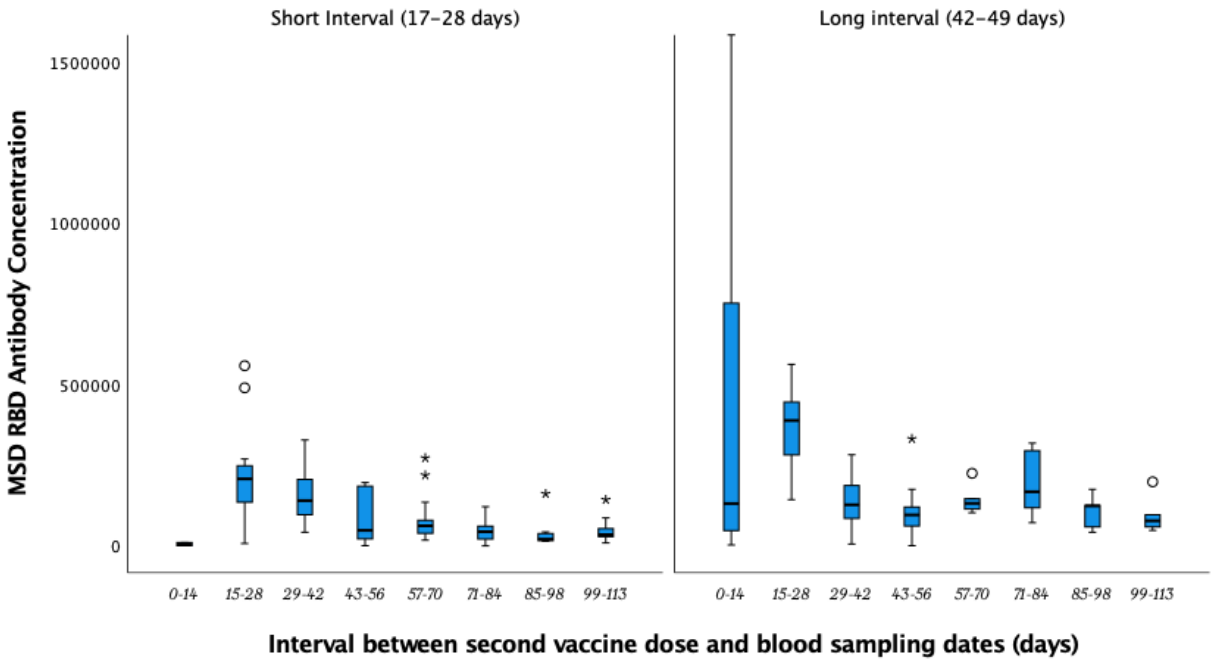
MSD, Meso Scale Discovery; MSD, RBD, receptor binding domain; NTD, spike N terminal domain; N, nucleocapsid; gM, geographic mean



Supplemental Figure 1: Box plots of the MSD Spike antibody concentration (Au/mL), stratified by the second vaccine dose-to-blood sampling interval

Black line is the median value, boxes are the inter-quartile range (IQR), whiskers include the full range of values (restricted to a maximum of IQR*1.5 above the 75th percentile, above which outliers are represented as individual data points)

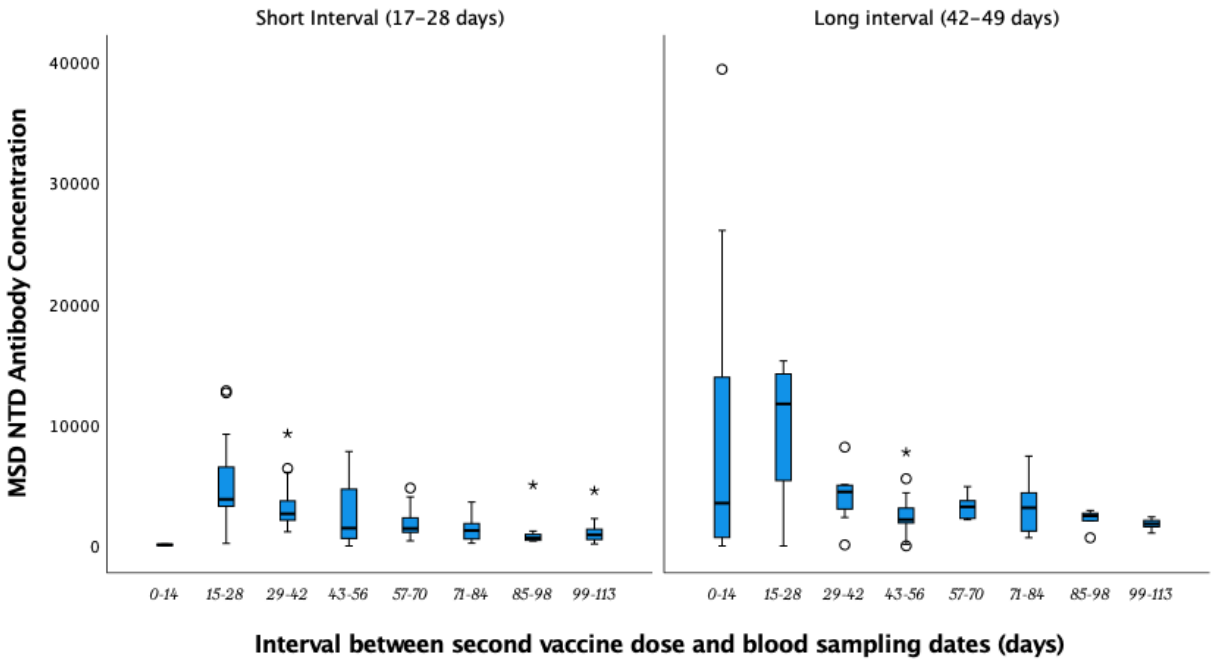
MSD, Meso Scale Discovery



Supplemental Figure 2: Box plots of the MSD RBD antibody concentration (Au/mL), stratified by the second vaccine dose to blood sampling interval

Black line is the median value, boxes are the inter-quartile range (IQR), whiskers include the full range of values (restricted to a maximum of IQR*1.5 above the 75th percentile, above which outliers are represented as individual data points)

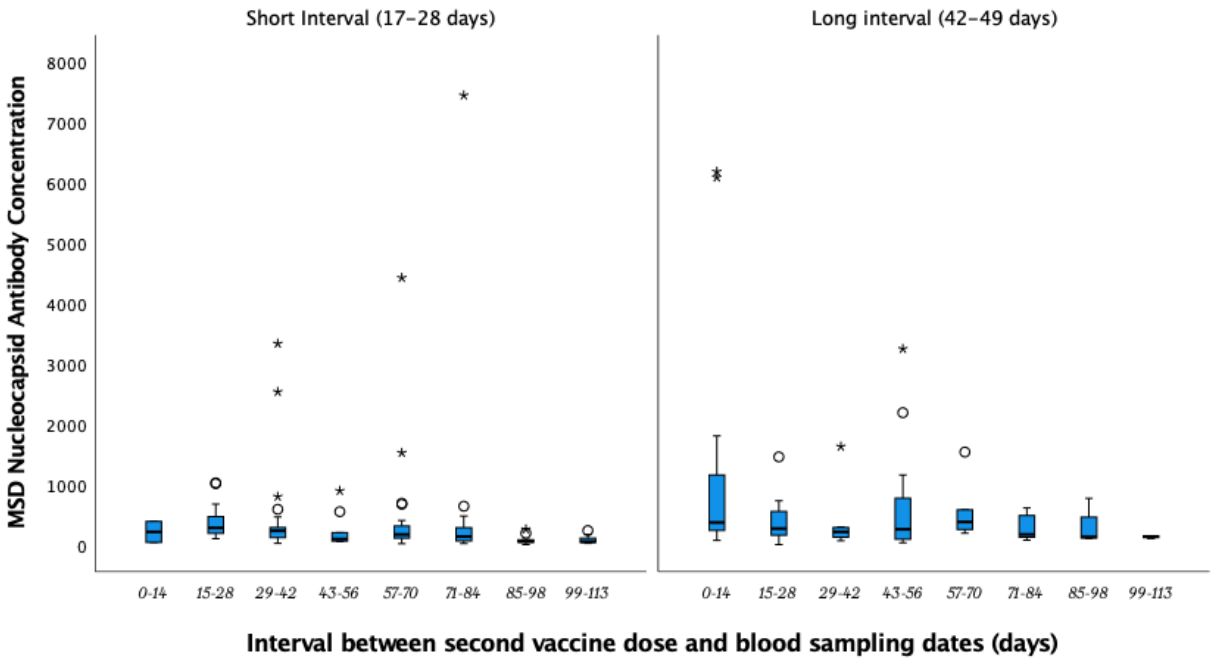
MSD, Meso Scale Discovery; MSD, RBD, receptor binding domain



Supplemental Figure 3: Box plots of the MSD NTD antibody concentration (Au/mL), stratified by the second vaccine dose-to-blood sampling interval

Black line is the median value, boxes are the inter-quartile range (IQR), whiskers include the full range of values (restricted to a maximum of IQR*1.5 above the 75th percentile, above which outliers are represented as individual data points)

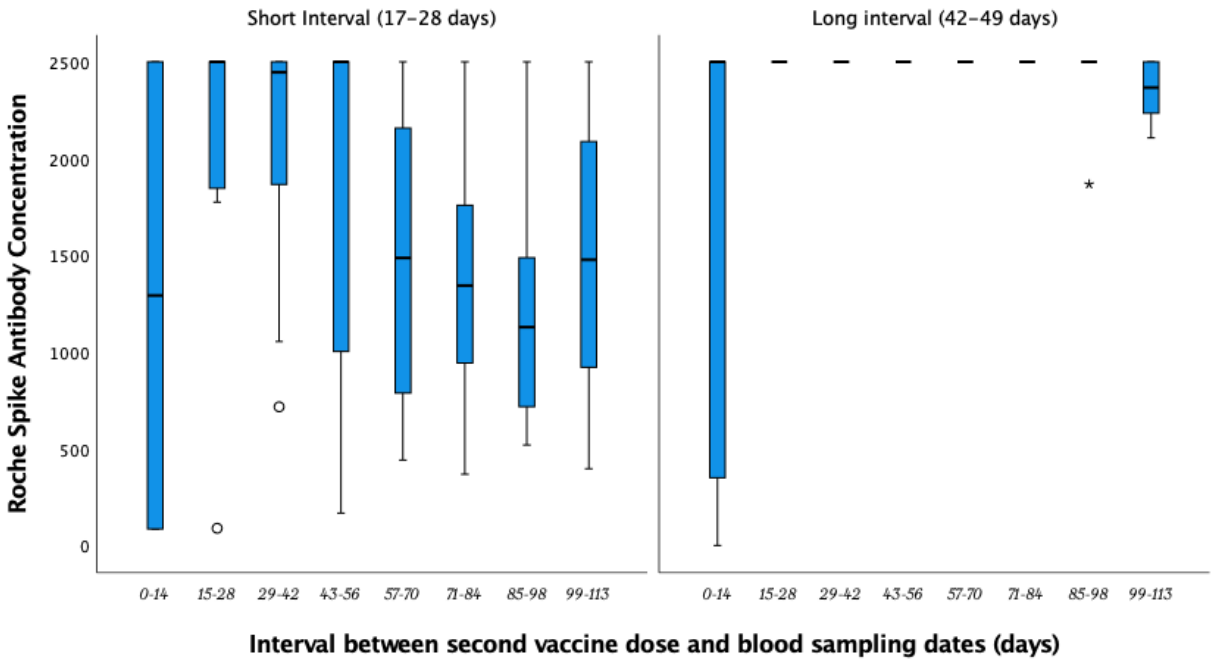
MSD, Meso Scale Discovery; NTD, spike N terminal domain



Supplemental Figure 4: Box plots of the MSD Nucleocapsid antibody concentration (Au/mL), stratified by the second vaccine dose-to-blood sampling interval

Black line is the median value, boxes are the inter-quartile range (IQR), whiskers include the full range of values (restricted to a maximum of IQR*1.5 above the 75th percentile, above which outliers are represented as individual data points)

MSD, Meso Scale Discovery



Supplemental Figure 5: Box plots of the Roche Spike antibody concentration (U/mL), stratified by the second vaccine dose-to-blood sampling interval

The maximum value of 2500 U/mL occurred in 35 (30%) samples of the short interval, and 60 (88%) samples of the long interval group

Black line is the median value, boxes are the inter-quartile range (IQR), whiskers include the full range of values (restricted to a maximum of IQR*1.5 above the 75th percentile, above which outliers are represented as individual data points)