

# Highly Sensitive and Specific SARS-CoV-2 Serological Assay Using a Magnetic Modulation Biosensing System

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**Table S1.** Data and characteristics of 85 SARS-CoV-2-positive and 79 SARS-CoV-2-negative samples tested using the 45-minute MMB-based SARS-CoV-2 IgG assay (MMB) and the 245-minute SARS-CoV-2 IgG ELISA test (ELISA). a. Data analysis and characteristics of 85 RT-qPCR SARS-CoV-2-positive serum samples taken from Israeli patients presenting to Sheba Medical Center, Israel. b. Data analysis and characteristics of 79 SARS-CoV-2-negative samples taken from Israeli patients before the COVID-19 outbreak (before September 2019).

a).

Sample #	ELISA	MMB	Days from PCR	Sample #	ELISA	MMB	Days from PCR
CO-854	Pos (2.11)	Pos (6.38)	31	CO-3869	Pos (1.45)	Pos (8.61)	107
CO-622	Pos (5.61)	Pos (43.91)	21	CO-3864	Pos (2.84)	Pos (33.59)	122
CO-623	Pos (3.33)	Pos (15.23)	21	CO-3813	Pos (2.66)	Pos (11.31)	127
CO-624	Pos (3.3)	Pos (8.12)	10	CO-3937	Pos (4.42)	Pos (22.76)	132
CO-625	Pos (2.56)	Pos (24.89)	15	CO-3936	Pos (3.57)	Pos (15.62)	113
CO-626	Neg (0.2)	Neg (3.25)	16	CO-3935	Neg (0.13)	Pos (54.35)	134
CO-636	Neg (0.55)	Neg (4.56)	N.A.	CO-3934	Pos (5.55)	Pos (41.63)	130
CO-627	Pos (1.21)	Pos (12.22)	8	CO-3933	Pos (2.47)	Pos (13.24)	103
CO-637	Pos (8.5)	Pos (25.12)	24	CO-3932	Pos (5.79)	Pos (46.14)	123
CO-843	Pos (1.62)	Pos (11.67)	18	CO-3931	Pos (3.63)	Pos (13.91)	141
CO-844	Pos (7.85)	Pos (25.42)	34	CO-3930	Pos (6.02)	Pos (21.71)	117
CO-846	Pos (11.54)	Pos (61.67)	25	CO-3929	Pos (4.77)	Pos (35.47)	123
CO-848	Pos (2)	Pos (7.59)	20	CO-3928	Pos (2.73)	Pos (22.2)	124
CO-849	Pos (11.51)	Pos (47.12)	35	CO-3927	Pos (7.37)	Pos (51.2)	108
CO-850	Pos (1.23)	Pos (6.08)	38	CO-3926	Pos (2.78)	Pos (19.57)	107
CO-851	Neg (0.49)	Pos (6.26)	27	CO-4302	Pos (4.25)	Pos (27.21)	129
CO-852	Pos (3.71)	Pos (21.07)	28	CO-7413	Pos (2.06)	Pos (25)	75
CO-853	Neg (0.44)	Neg (2.4)	27	CO-7419	Pos (2.72)	Pos (22)	N.A.

<b>CO-3803</b>	Pos (1.18)	Pos (10.09)	112	<b>CO-7416</b>	Pos (4.37)	Pos (44)	N.A.
<b>CO-3801</b>	Pos (3.65)	Pos (19)	114	<b>CO-7396</b>	Pos (4.04)	Pos (37)	N.A.
<b>CO-3469</b>	Pos (1.64)	Pos (8.02)	119	<b>CO-7395</b>	Pos (2.97)	Pos (16)	N.A.
<b>CO-3372</b>	Pos (4.59)	Pos (11)	104	<b>CO-7389</b>	Pos (2.21)	Pos (18)	N.A.
<b>CO-3863</b>	Pos (2.31)	Pos (22.88)	127	<b>CO-7388</b>	Pos (3.21)	Pos (23)	N.A.
<b>CO-3862</b>	Pos (5.3)	Pos (28.46)	139	<b>CO-7387</b>	Pos (6.12)	Pos (22)	N.A.
<b>CO-3860</b>	Pos (6.23)	Pos (24.39)	117	<b>CO-7386</b>	Pos (4.99)	Pos (40)	N.A.
<b>CO-3858</b>	Pos (1.32)	Pos (6.8)	125	<b>CO-7412</b>	Pos (7.53)	Pos (59)	79
<b>CO-3857</b>	Pos (6.81)	Pos (38.13)	128	<b>CO-7411</b>	Pos (5.99)	Pos (28)	150
<b>CO-3856</b>	Pos (5.82)	Pos (16.74)	119	<b>CO-7410</b>	Pos (4.58)	Pos (49)	259
<b>CO-3854</b>	Neg (0.19)	Neg (3.77)	117	<b>CO-7408</b>	Pos (2.13)	Pos (18)	275
<b>CO-3851</b>	Neg (0.68)	Neg (2.23)	90	<b>CO-7407</b>	Pos (4.26)	Pos (8.1)	101
<b>CO-3812</b>	Pos (2.31)	Pos (6.67)	121	<b>CO-7404</b>	Pos (4.89)	Pos (33)	72
<b>CO-3811</b>	Pos (4.11)	Pos (19.35)	115	<b>CO-7402</b>	Pos (5.09)	Pos (30)	N.A.
<b>CO-3810</b>	Pos (3.96)	Pos (17.13)	121	<b>CO-7401</b>	Pos (4.25)	Pos (23)	N.A.
<b>CO-3808</b>	Pos (6.52)	Pos (23.56)	99	<b>CO-7397</b>	Pos (2.47)	Pos (16)	N.A.
<b>CO-3940</b>	Pos (3.51)	Pos (16.61)	139	<b>CO-7326</b>	Pos (2.07)	Pos (16)	76
<b>CO-3938</b>	Pos (3.84)	Pos (12)	127	<b>CO-7327</b>	Pos (6.62)	Pos (18)	264
<b>CO-3925</b>	Pos (6.73)	Pos (46.44)	95	<b>CO-7328</b>	Pos (6.7)	Pos (10)	267
<b>CO-3924</b>	Pos (5.72)	Pos (37.35)	107	<b>CO-7329</b>	Pos (5.04)	Pos (20)	267
<b>CO-3923</b>	Pos (6.1)	Pos (42.99)	143	<b>CO-7330</b>	Pos (3)	Pos (22)	267
<b>CO-3922</b>	Pos (1.68)	Neg (2.16)	58	<b>CO-7175</b>	Pos (2.17)	Pos (9.79)	54
<b>CO-3920</b>	Pos (5.62)	Pos (9.57)	121	<b>CO-6819</b>	Pos (2.81)	Pos (15)	120
<b>CO-3871</b>	Pos (1.51)	Pos (26.5)	90	<b>CO-7384</b>	Pos (2.73)	Pos (8.9)	N.A.
<b>CO-3919</b>	Pos (6.06)	Pos (47.1)	130				

b).

<b>Sample #</b>	<b>ELISA</b>	<b>MMB</b>	<b>Sample #</b>	<b>ELISA</b>	<b>MMB</b>
<b>5542/19</b>	Neg (0.31)	Neg (0.80)	<b>1397/19</b>	Neg (0.37)	Neg (3.38)
<b>5543/19</b>	Neg (0.44)	Neg (0.89)	<b>1438/19</b>	Equ (1.09)	Neg (3.57)
<b>5544/19</b>	Neg (0.32)	Neg (1.13)	<b>1263/19</b>	Neg (0.54)	Neg (2.53)
<b>5545/19</b>	Neg (0.31)	Neg (0.84)	<b>1266/19</b>	Neg (0.45)	Neg (3.37)
<b>5546/19</b>	Neg (0.31)	Neg (2.90)	<b>1278/19</b>	Neg (0.33)	Neg (3.56)
<b>5547/19</b>	Neg (0.38)	Neg (3.57)	<b>1301/19</b>	Neg (0.38)	Neg (1.34)
<b>5548/19</b>	Neg (0.32)	Neg (2.91)	<b>1312/19</b>	Neg (0.29)	Neg (3.86)
<b>5549/19</b>	Neg (0.37)	Neg (1.88)	<b>1334/19</b>	Neg (0.36)	Neg (3.68)
<b>5541/19</b>	Neg (0.37)	Neg (4.21)	<b>1343/19</b>	Neg (0.38)	Neg (2.04)
<b>5539/19</b>	Neg (0.33)	Neg (1.91)	<b>1346/19</b>	Neg (0.35)	Neg (3.15)
<b>5332/19</b>	Neg (0.24)	Neg (1.01)	<b>1391/19</b>	Neg (0.48)	Neg (3.13)
<b>5334/19</b>	Neg (0.2)	Neg (3.83)	<b>1158/19</b>	Neg (0.33)	Neg (2.68)
<b>5335/19</b>	Neg (0.31)	Neg (4.07)	<b>1053/19</b>	Neg (0.36)	Pos (12)

5538/19	Neg (0.32)	Neg (2.75)	1112/19	Neg (0.43)	Neg (2.4)
5537/19	Neg (0.32)	Neg (1.84)	1189/19	Neg (0.5)	Neg (2.25)
5530/19	Neg (0.24)	Neg (1.64)	1242/19	Neg (0.69)	Neg (1.79)
5531/19	Neg (0.43)	Neg (4.14)	1206/19	Neg (0.33)	Neg (1.5)
5528/19	Neg (0.3)	Neg (2.53)	1262/19	Neg (0.33)	Neg (1.2)
5527/19	Neg (0.32)	Neg (3.44)	1063/19	Neg (0.38)	Neg (2)
5521/19	Neg (0.35)	Neg (4.51)	1119/19	Pos (1.16)	Pos (10.08)
5522/19	Neg (0.5)	Neg (3.86)	2193/19	Neg (0.26)	Neg (1.11)
5525/19	Neg (0.27)	Neg (2.31)	2217/19	Neg (0.51)	Neg (1.36)
5524/19	Neg (0.46)	Neg (3.01)	2327/19	Neg (0.24)	Neg (1.5)
5523/19	Neg (0.4)	Neg (1.85)	2156/19	Neg (0.24)	Neg (1.11)
5320/19	Neg (0.49)	Neg (1.66)	2278/19	Neg (0.21)	Neg (1.21)
5517/19	Neg (0.46)	Neg (2.08)	2321/19	Neg (0.21)	Neg (1)
5516/19	Neg (0.28)	Neg (1.54)	2325/19	Neg (0.28)	Neg (0.95)
5509/19	Neg (0.36)	Neg (1.48)	2328/19	Neg (0.29)	Neg (1.17)
5515/19	Neg (0.32)	Neg (2.53)	2246/19	Neg (0.22)	Neg (1.11)
5508/19	Neg (0.43)	Neg (3.03)	2244/19	Neg (0.18)	Neg (1.10)
5507/19	Neg (0.74)	Neg (1.87)	2252/19	Neg (0.26)	Neg (2.5)
5514/19	Neg (0.27)	Neg (1.82)	2521/19	Neg (0.31)	Neg (2.2)
5506/19	Neg (0.73)	Neg (2.10)	2324/19	Neg (0.26)	Neg (4.1)
5513/19	Neg (0.46)	Neg (2.32)	2323/19	Neg (0.34)	Neg (1.3)
5518/19	Neg (0.36)	Neg (1.71)	2211/19	Neg (0.39)	Neg (2.66)
5526/19	Neg (0.37)	Neg (1.22)	2251/19	Neg (0.23)	Neg (1.9)
5511/19	Neg (0.51)	Neg (1.68)	2158/19	Neg (0.18)	Neg (1.8)
5512/19	Neg (0.53)	Neg (3.14)	2260/19	Neg (0.24)	Neg (1.6)
5505/19	Neg (0.54)	Neg (2.87)	2326/19	Neg (0.14)	Neg (2.4)
5510/19	Neg (0.39)	Neg (1.33)			

Using ELISA, values below 0.8 (ELISA absorbance units) are regarded as negative, values in the range of 0.8–1.1 are regarded as equivocal (which are clinically regarded as negative, but require further monitoring), and values above 1.1 are regarded as positive. Using the MMB-based assay, the ROC cutoff is 5.32. Thus, values below 5.32 (MMB normalized fluorescent signal) are regarded as negative, and values above 5.32 are regarded as positive. **Abbreviations:** ELISA, enzyme-linked immunosorbent assay; MMB, magnetic modulation biosensing; N.A., not applicable; Neg, negative; Pos, positive; Equ, Equivocal; ROC, receiver operating characteristic.

**Table S2.** Data and characteristics of 10 Israeli patients vaccinated with two doses of BNT162b2 mRNA (Pfizer-BioNTech) 21 days apart.

Sample #	Age	Administration of first dose	Serology test (IgG)	ELISA	MMB
70001			29/12/2020	Neg (0.24)	Neg (4.53)
A 70302	24	20/12/2020	04/01/2021	Pos (3.31)	Pos (22.76)
70720			11/01/2021	Pos (4.32)	Pos (53.32)

				71267	19/01/2021	Pos (6.79)	Pos (107.83)
				70004	29/12/2020	Neg (0.41)	Pos (6.95)
				70324	04/01/2021	Pos (2.01)	Pos (20.18)
				70715	11/01/2021	Pos (1.89)	Pos (58.58)
				71144	18/01/2021	Pos (7.08)	Pos (88.83)
				70005	29/12/2020	Neg (0.27)	Pos (9.04)
				70316	04/01/2021	Equ (0.93)	Pos (7.5)
				70706	11/01/2021	Pos (1.41)	Pos (17.18)
				71207	19/01/2021	Pos (7.14)	Pos (50.41)
				70006	29/12/2020	Neg (0.38)	Pos (11.11)
				70293	04/01/2021	Pos (4.06)	Pos (21.52)
				70670	11/01/2021	Pos (3.54)	Pos (28.68)
				71125	18/01/2021	Pos (7.55)	Pos (29.26)
				70023	29/12/2020	Neg (0.29)	Pos (6.5)
				70300	04/01/2021	Pos (3.98)	Pos (26.3)
				70688	11/01/2021	Pos (5.1)	Pos (44.82)
				71148	18/01/2021	Pos (7.05)	Pos (41.02)
				70034	29/12/2020	Neg (0.24)	Pos (7.34)
				70299	04/01/2021	Pos (1.23)	Pos (8.84)
				70683	11/01/2021	Pos (1.58)	Pos (14.36)
				71139	18/01/2021	Pos (6.78)	Pos (49.35)
				70018	29/12/2020	Neg (0.29)	Neg (2.9)
				70296	04/01/2021	Neg (0.48)	Neg (4.35)
				70679	11/01/2021	Pos (1.23)	Pos (8.07)
				72284	N/A	Pos (7.38)	Pos (42.05)
				70044	29/12/2020	Neg (0.38)	Neg (4.07)
				70493	06/01/2021	Pos (2.33)	Pos (12.44)
				70895	13/01/2021	Pos (4.13)	Pos (16.53)
				71383	21/01/2021	Pos (6.08)	Pos (20.68)
				70048	29/12/2020	Neg (0.23)	Neg (3.32)
				70310	04/01/2021	Pos (2.03)	Pos (15.84)
				70701	11/01/2021	Pos (4.1)	Pos (32.2)
				71170	18/01/2021	Pos (7.12)	Pos (35.83)
				70075	30/12/2020	Neg (0.22)	Pos (17.56)
				70375	05/01/2021	Pos (3.71)	Pos (22.13)
				70887	13/01/2021	Pos (4.78)	Pos (30.15)
				71461	21/01/2021	Pos (7.66)	Pos (37.82)

The samples were obtained 7, 14, 21, and 28 days following the administration of the first dose. All patients received their second dose 21 days following the first dose. Using ELISA, values below 0.8 (ELISA absorbance units) are regarded as negative, values in the range of 0.8–1.1 are regarded as equivocal (which are clinically regarded as negative but

require further monitoring), and values above 1.1 are regarded as positive. Using the MMB-based assay, the ROC cutoff is 5.32. Thus, values below 5.32 (MMB normalized fluorescent signal) are regarded as negative and values above 5.32 are regarded as positive. Abbreviations: Neg, negative; Pos, positive; Equ, equivocal.

**Table S3.** Data and characteristics of 25 Israeli patient's RT-qPCR SARS-CoV-2-positive samples that were taken from a large cohort study and were identified as borderline negatives by the 245-minute ELISA test.

Sample #	ELISA	MMB	Days from PCR
CO-926	Equ (1.01)	Pos (7.3)	42
CO-1486	Equ (1.02)	Pos (9.74)	44
CO-1472	Equ (0.98)	Pos (9.73)	42
CO-1488	Equ (1.05)	Pos (13.48)	42
CO-2301	Equ (0.94)	Neg (3)	72
CO-3362	Equ (0.98)	Neg (4.07)	72
CO-1436	Equ (0.97)	Pos (9.5)	38
CO-3368	Equ (1.01)	Pos (9.71)	104
CO-3573	Equ (0.92)	Pos (8.35)	109
CO-3673	Equ (0.92)	Neg (4.53)	123
CO-3675	Equ (0.94)	Neg (4.36)	112
CO-7409	Equ (0.99)	Pos (11.9)	149
CO-7128	Equ (0.9)	Pos (11.1)	N.A.
CO-6739	Equ (1.07)	Neg (2.79)	N.A.
CO-6718	Equ (1.05)	Pos (8.4)	73
CO-6722	Equ (0.97)	Pos (5.83)	66
CO-7147	Equ (0.96)	Neg (3.52)	78
CO-6655	Equ (1.02)	Neg (4.43)	N.A.
CO-4971	Equ (0.95)	Neg (4)	N.A.
CO-6757	Equ (1.06)	Pos (8.4)	62
CO-6759	Equ (1.06)	Pos (7.4)	132
CO-7063	Equ (1.08)	Equ (5.32)	159
CO-6635	Equ (1.01)	Pos (7.48)	N.A.
CO-7087	Equ (1.03)	Neg (2.52)	236
CO-7105	Equ (1.04)	Neg (2.16)	N.A.
CO-926	Equ (1.01)	Pos (7.3)	42
CO-1486	Equ (1.02)	Pos (9.74)	44
CO-1472	Equ (0.98)	Pos (9.73)	42
CO-1488	Equ (1.05)	Pos (13.48)	42

The samples were tested using the 45-minute MMB-based SARS-CoV-2 IgG assay (MMB). Using ELISA, values below 0.8 (ELISA absorbance units) are regarded as negative, values in the range of 0.8–1.1 are regarded as equivocal (which are clinically regarded as negative but require further monitoring), and values above 1.1 are regarded as positive. Using the MMB-based assay, the ROC cutoff is 5.32. Thus, values below 5.32 (MMB normalized fluorescent signal) are regarded as negative and values above 5.32 are regarded as positive. **Abbreviations:** N.A., not applicable; Neg, negative; Pos, positive; Equ, equivocal.

**Table S4.** Characteristics of anti-SARS-CoV-2 serological assays using WHO International Standard recombinant human immunoglobulin (Ig)G anti-SARS CoV-2 S1 antibody: Manufacturer, target antigen, method, dynamic range, and cutoff value in BAU/mL.

Manufacturer name	Target antigen	Method	Dynamic range (BAU/mL)	Cutoff (BAU/mL)
MMB	S-RBD	MMB	1.14–125	8.4
RBD-ELISA	S-RBD	ELISA	4.35–250	24.2
Euroimmun	S1	ELISA	3.2–384	25.6 (25.6–35.2 borderline)
Diasorin	Trimeric S	CLIA	4.8–2080	33.8
Thermo Fisher	S1	FEIA	2.8–816	28 (28–40 borderline)
Roche	S-RBD	ECLIA	0.40–243	0.823
Snibe	S-RBD	CLIA	0.78–433	4.33
Mindary	S-RBD	CLIA	3.65–1216	12.16

BAU, binding antibody unit; CLIA, chemiluminescence immunoassay; ECLIA, electrochemiluminescence immunoassay; ELISA, enzyme-linked immunosorbent assay; FEIA, fluorescence enzyme immunoassay