Supplementary Information

Title: Test performance evaluation of SARS-CoV-2 serological assays

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Assay	Supplier	Product	Antigen*	Format **	Lot(s)	Product Number	Distributor	Kit Acquisition for Study	Performance Notes
LFAs	**BioMedomics** Inc, Morrisville, NC, USA	COVID-19 IgM-IgG Rapid Test	RBD	1	2020032103	51-002-20	Henry Schein, Melville, NY, USA	Provided by Distributor Free of Charge	Some control band inconsistency
	Bioperfectus Technologies Co Ltd, Jiangsu, China	PerfectPOC Novel Corona Virus (SARS-CoV-2) IgM/IgG Rapid Test Kit	NP, SP	1	20200313, 20200313, 20210312	SC30201W	--------	Provided by Supplier Free of Charge	Extra diluent necessary
	Decombio Biotechnology Co Ltd, Beijing, China	Novel Coronavirus (SARS-CoV-2) IgM/IgG Combo Rapid Test-Cassette	--------	1	--------	--------	--------	Provided by Supplier Free of Charge	Some control band inconsistency
	DeepBlue Medical Technology Co Ltd, Anhui, China	COVID-19 (SARS-CoV-2) IgG/IgM Antibody Test Kit (Colloidal Gold)	--------	1	20200305	--------	--------	Donated by John Hering, who purchased from supplier	Extra diluent necessary, Some control band inconsistency
	Innovita Biological Technology Co Ltd, Qian'an, China	Novel Coronavirus (2019-nCoV) Ab Test (Colloidal Gold)	NP, SP	2	20200304	--------	20/20 GeneSystems, Rockville, MD, USA	Purchased from Distributor	Some band smearing
	Premier Biotech, Minneapolis, MN, USA	COVID-19 IgG/IgM Rapid Test Cassette	--------	1	COV2003071	INGM-MC42S	--------	Purchased from Supplier	Some band smearing
	Sure Biotech, New York, NY, USA; Wan Chai, Hong Kong	SARS-CoV-2 IgM/IgG Antibody Rapid Test	NP, SP	1	COV1252003B	VC012103	--------	Provided by Supplier Free of Charge	--------
	UCP Biosciences, San Jose, CA, USA	Coronavirus IgG/IgM Antibody (COVID-19) Test Cassette	--------	1	SMP2020031 2, SMP2020031 3	U-CoV-102	--------	Provided by Supplier Free of Charge	Extra diluent necessary
	VivaChek Biotech Co, Hangzhou, China	VivaDiag™ SARS-CoV-2 IgM/IgG Rapid Test (COVID-19 IgM/IgG Rapid Test)	--------	1	E2003002	VID35-08-011	Everest Links Pte Ltd, Singapore	Purchased from Distributor	Some band smearing
	Wondfo Biotech Co Ltd, Guangzhou, China	SARS-CoV-2 Antibody Test (Lateral Flow Method)	--------	3	W19500318	W195	--------	Donated by David Friedberg, who purchased from supplier	Some band smearing

Assay	Supplier	Product	Antigen*	Format**	Lot(s)	Product Number	Distributor	Kit Acquisition for Study	Performance Notes
MGH LFAs	**SD Biosensor**, Suwon-si, Gyeonggi-doz, Republic of Korea	STANDARD Q COVID-19 IgM/IgG Duo	NP	2	QCO102000 6	Q-NCOV-01D	Henry Schein, Melville, NY, USA	Provided by Distributor Free of Charge	--------
	Biolidics Limited, Mapex, Singapore	2019-nCoV IgG/IgM antibody detection kit	NP, RBD	1	V2020 0330	CBB-F015016 -V	--------	Purchased from Supplier	--------
	Biomedomics Inc, Morrisville, NC, USA	COVID-19 IgM and IgG Rapid Test	RBD	1	20200 22702 20200 32103	51-002-20	Henry Schein, Melville, NY, USA	Lot 1 provided by Distributor Free of Charge; Lot 2 purchased from Supplier	--------
ELISAs	**Epitope** Diagnostics, San Diego, CA, USA	KT-1033 EDI™ Novel Coronavirus COVID-19 **IgM** ELISA Kit	NP	--	P630C	KT-1032	--------	Purchased from Supplier	--------
	Epitope Diagnostics, San Diego, CA, USA	KT-1032 EDI™ Novel Coronavirus COVID-19 **IgG** ELISA Kit	NP	--	P637U	KT-1033	--------	Purchased from Supplier	--------
	In-House ELISA	Peroxidase AffiniPure Goat Anti-human IgG (F(ab')2 specific) secondary antibody (Jackson ImmunoResearch)	RBD	--	14657 6	109-035-097	Adapted from Krammer Lab, Icahn School of Medicine at Mt. Sinai, New York, NY, USA	Lab-developed test	--------

*Antigen:
NP = Nucleocapsid protein
SP = Spike protein
RBD = Receptor binding domain, Spike protein

**LFA Test Cartridge Format:
1: Single lane, separate IgM and IgG bands
2: Separate IgM and IgG lanes
3: Single lane, single band for both IgM and IgG

Supplementary Table 1. Immunoassay Kit and Manufacturer Information. Bold signifies labels used in text and figures.

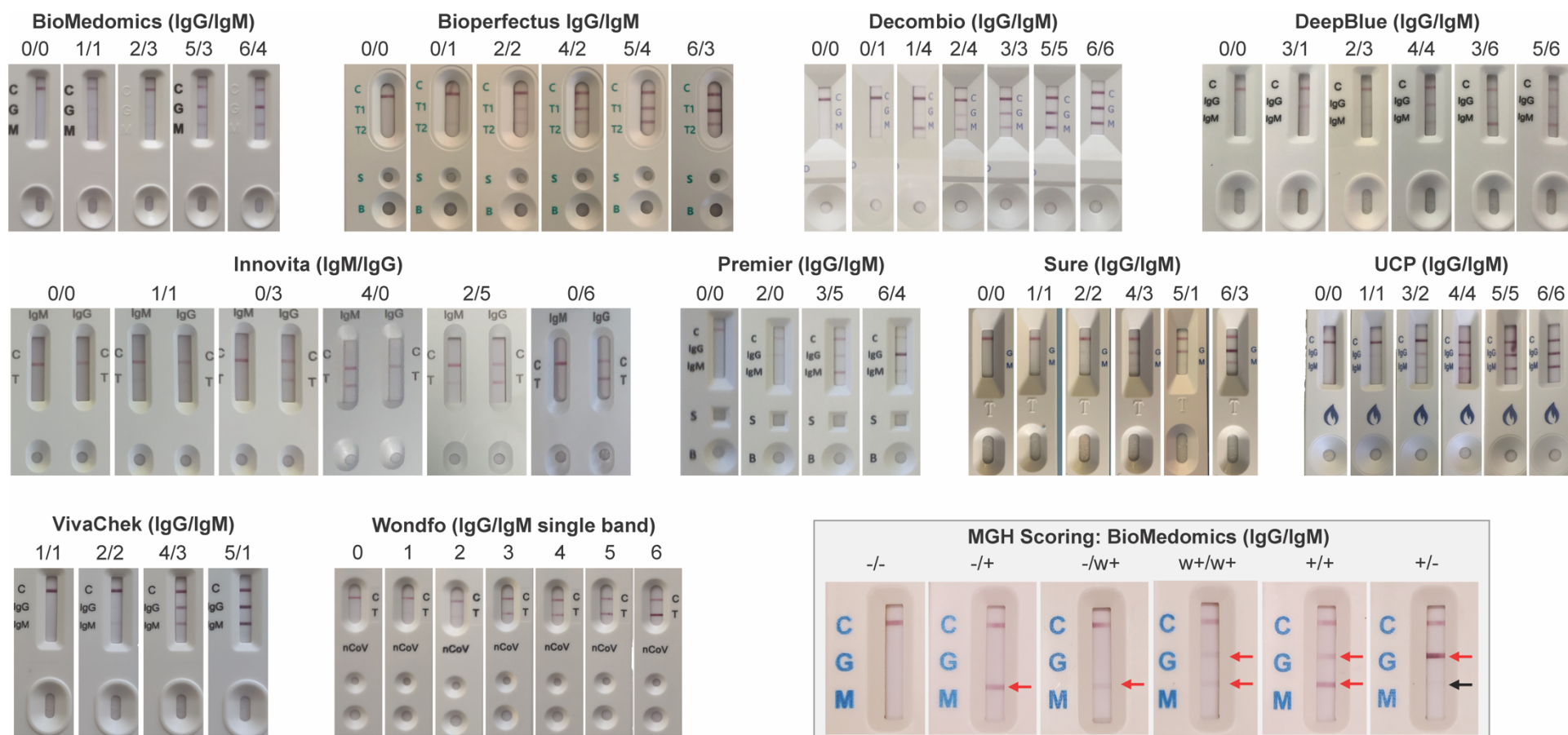|	IgG			IgM		
Supplier	n	Positive Kappa Correlation	Weighted Kappa Correlation	n	Positive Kappa Correlation	Weighted Kappa correlation
BioMedomics	284	0.9649	0.9580	284	0.8238	0.8253
Bioperfectus	275	0.9586	0.9488	275	0.9130	0.8631
DecomBio	282	0.9761	0.9529	282	0.9845	0.9660
DeepBlue	287	0.9547	0.8971	287	0.9213	0.9377
Innovita	250	0.9588	0.8490	249	0.8084	0.8029
Premier	286	0.9718	0.9880	286	0.9680	0.9340
Sure	286	0.9908	0.9665	286	0.9300	0.7967
UCP	286	0.9565	0.9574	286	1.0000	0.9484
VivaChek	266	0.9912	0.9669	266	0.9332	0.9439
Wondfo*	271	0.9916	0.9542	-	-	-

*Wondfo kits detect combined IgG and IgM.

Supplementary Table 2. **Reader Agreement on Immunochromatographic Lateral Flow Assays (LFAs).** Cohen's Kappa correlations were calculated for scores of the IgG band (left) and IgM band (right) of each LFA. The LFA manufactured by Wondfo has a single band for IgG and IgM detection and is displayed here as IgG for convenience. Positive Kappa Correlation: unweighted inter-reader agreement on positive (LFA score > 0) vs. negative (LFA score = 0) reads. Weighted Kappa Correlation: inter-reader agreement on LFA score (0-6), weighted by the square of the difference in reads. All correlations were calculated with the *irr* package version 0.84.1 in R version 3.6.1 using RStudio.
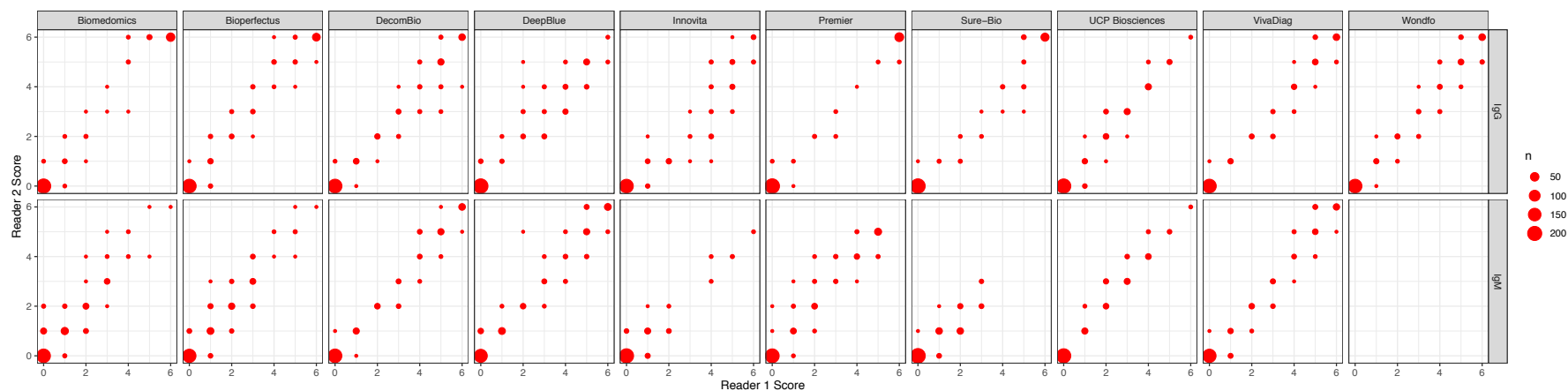
MGH Serology Test Performance Evaluation

Assay	Total N	IgM positive	%	95% CI Lower	95% CI Upper	Total N	IgG positive	%	95% CI Lower	95% CI Upper	Total N	IgM or IgG positive	%	95% CI Lower	95% CI Upper
LFAs															
SD Biosensor															
1-5 days	7	0	0.00	0.00	40.96	7	1	14.29	0.36	57.87	7	1	14.29	0.36	57.87
6-10 days	15	6	40.00	16.34	67.71	15	5	33.33	11.82	61.62	15	7	46.67	21.27	73.41
11-15 days	19	15	78.95	54.43	93.95	19	16	84.21	60.42	96.62	19	17	89.47	66.86	98.70
>16 days	7	6	85.71	42.13	99.64	7	6	85.71	42.13	99.64	7	6	85.71	42.13	99.64
Pre-COVID-19	60	0				60	1				60	1			
Biolidics															
1-5 days	7	0	0.00	0.00	40.96	7	0	0.00	0.00	40.96	7	0	0.00	0.00	40.96
6-10 days	15	2	13.33	1.66	40.46	15	8	53.33	26.59	78.73	15	8	53.33	26.59	78.73
11-15 days	19	9	47.37	24.45	71.14	19	16	84.21	60.42	96.62	19	16	84.21	60.42	96.62
>16 days	7	4	57.14	18.41	90.10	7	6	85.71	42.13	99.64	7	6	85.71	42.13	99.64
Pre-COVID-19	60	0				60	0				60	0			
BioMedomics															
1-5 days	7	1	14.29	0.36	57.87	7	0	0.00	0.00	40.96	7	1	14.29	0.36	57.87
6-10 days	15	6	40.00	16.34	67.71	15	6	40.00	16.34	67.71	15	7	46.67	21.27	73.41
11-15 days	19	14	73.68	48.80	90.85	19	14	73.68	48.80	90.85	19	15	78.95	54.43	93.95
>16 days	7	6	85.71	42.13	99.64	7	6	85.71	42.13	99.64	7	6	85.71	42.13	99.64
Pre-COVID-19	60	0				60	0				60	0			

Supplementary Table 3. Assay performance on validation cohort performed at MGH using positivity thresholds based on concordance studies to an MGH-group in-house ELISA. Comparison of MGH and UCSF percent positivity at different positivity thresholds is performed in Supplementary Figure 4. Note, the one negative patient included in the >16-day timepoint was immunocompromised.
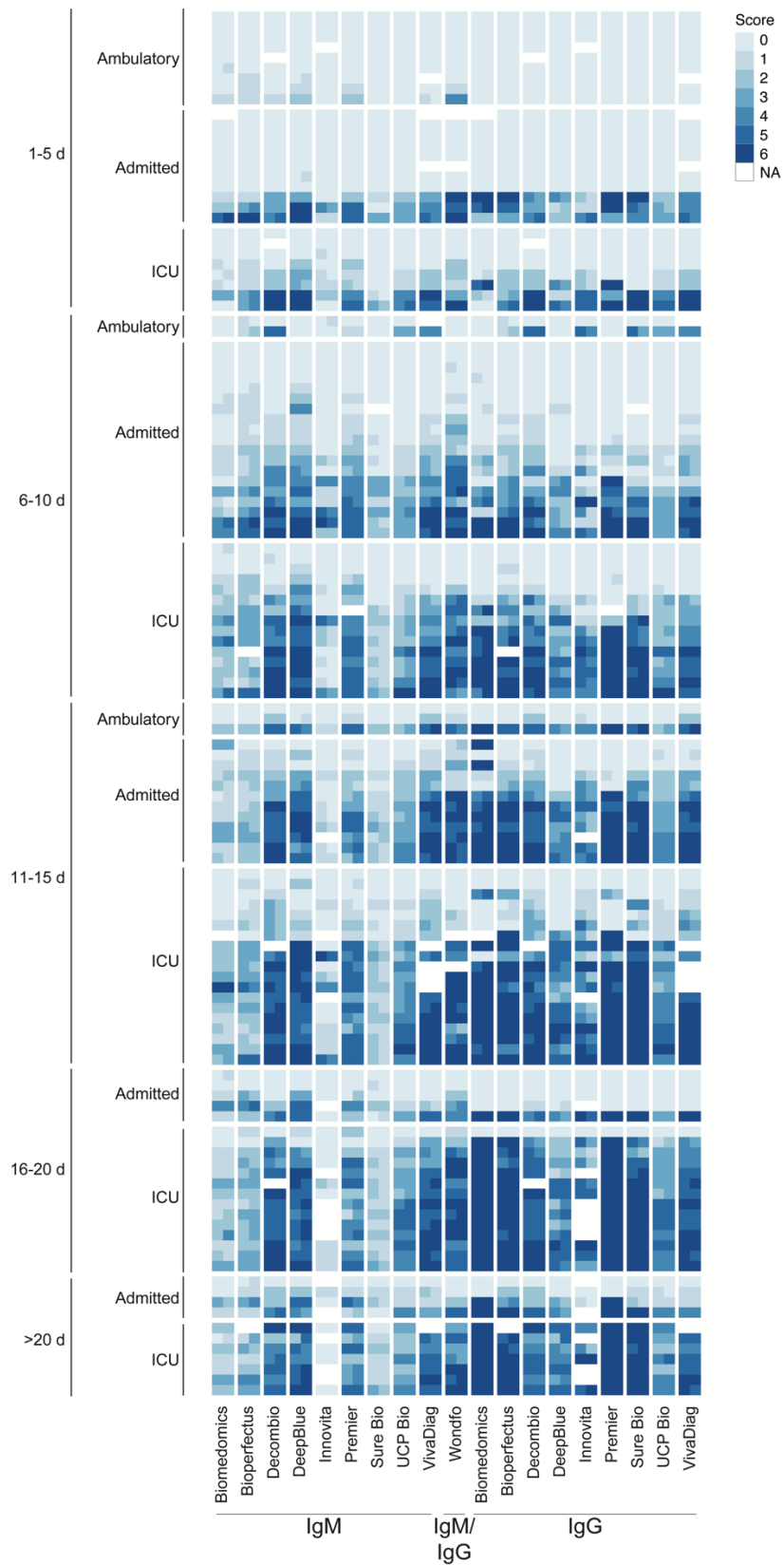
Supplementary Figure 1: Representative images of LFA scoring. The intensity score is noted at the top of each image for IgG and IgM separately (unless stated otherwise).

Supplementary Figure 2: Comparison of Reader 1 and Reader 2 LFA scores.
The size of each point signifies the number of tests with the indicated reader 1-to-reader 2 score combination. The LFA manufactured by Wondfo has a single band for IgG and IgM detection and is displayed here as IgG for convenience.

Supplementary Figure 3: LFA scores by serological assay according to highest-level clinical care received by the patient.