SARS-CoV-2 Antibody Responses Do Not Predict COVID-19 Disease Severity

William S. Phipps, MD,[1,] Jeffrey A. SoRelle, MD,[1,*] Quan-Zhen Li, MD, PhD,[2,3,*] Lenin Mahimainathan, PhD, MB(ASCP),[1] Ellen Araj, MD,[1] John Markantonis, DO,[1] Chantale Lacelle, PhD,[1] Jyoti Balani, MD,[1] Hiren Parikh, MT(ASCP),[1] E. Blair Solow, MD, MSCS,[2] David R. Karp, MD, PhD,[2] Ravi Sarode, MD,[1,4] and Alagarraju Muthukumar, PhD, DABCC[1]*

From the Departments of [1]Pathology, [2]Internal Medicine (Rheumatic Diseases), [3]Immunology, Microarray Core Facility, and [4]Internal Medicine (Hematology/Oncology), University of Texas Southwestern Medical Center, Dallas.

Key Words: Microbiology; Immunology; Global health; SARS-CoV-2; COVID-19; Coronavirus

Am J Clin Pathol 2020;XX:0–0

DOI: 10.1093/AJCP/AQAA123

ABSTRACT

Objectives: *Initial reports indicate adequate performance of some serology-based severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) assays. However, additional studies are required to facilitate interpretation of results, including how antibody levels impact immunity and disease course.*

Methods: *A total of 967 subjects were tested for IgG antibodies reactive to SARS-CoV-2, including 172 suspected cases of SARS-CoV-2, 656 plasma samples from healthy donors, 49 sera from patients with rheumatic disease, and 90 specimens from individuals positive for polymerase chain reaction (PCR)–based respiratory viral panel. A subgroup of SARS-CoV-2 PCR-positive cases was tested for IgM antibodies by proteome array method.*

Results: *All specificity and cross-reactivity specimens were negative for SARS-CoV-2 IgG antibodies (0/795, 0%). Positive agreement of IgG with PCR was 83% of samples confirmed to be more than 14 days from symptom onset, with less than 100% sensitivity attributable to a case with severe immunosuppression. Virus-specific IgM was positive in a higher proportion of cases less than 3 days from symptom onset. No association was observed between mild and severe disease course with respect to IgG and IgM levels.*

Conclusions: *The studied SARS-CoV-2 IgG assay had 100% specificity and no adverse cross-reactivity. Measures of IgG and IgM antibodies did not predict disease severity in our patient population.*

Key Points

- PCR-confirmed cases of COVID-19 demonstrate high rates of seroconversion beyond 14 days of symptoms unless a patient is severely immunosuppressed.
- Testing for IgG against SARS-CoV-2 nucleocapsid protein can be performed with high specificity, including in the setting of prior respiratory infection or underlying rheumatic disease.
- Index values of IgG and IgM antibodies did not appear to predict disease severity.

As the COVID-19 global pandemic continues,[1] a major priority is the application of serologic testing to determine the scale and rate of exposures. The coronavirus disease 2019 (COVID-19) pathogen, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), is an enveloped, positive-sense, single-stranded RNA Betacoronavirus with an approximately 30 kilobase genome.[2] The molecular detection of SARS-CoV-2 is based on targeting the viral genome (eg, Orf1a/b, E, S, N genes) by polymerase chain reaction (PCR)[3-7] and is currently the gold standard to diagnose acute infection.[3] Cellular and humoral immunity resolve the infection, which can be detected by the formation of antibodies specific for the virus.

Various serologic assays have recently acquired the Food and Drug Administration's emergency use authority for SARS-CoV-2 antibody testing in COVID-19 patients, but the interpretation of antibody data and their clinical significance remains challenging. Understanding the time course of antibody response and potential reasons for

apparent failure of seroconversion are essential. Further, before assessing whether specific antibodies ameliorate SARS-CoV-2 infection or prevent reinfection, confidence in the analytical specificity of the test is required. Antibody assays in general are frequently susceptible to nonspecific reactivity, leading to false positives. This can have dramatic effects when the incidence of exposure is low. Thus, a high positive predictive value gained from minimal cross-reactivity towards other pathogen or autoimmune-associated antibodies is critical.

Long et al[8] have described a variable antiviral IgM and IgG immune response to SARS-CoV-2 infection in a Chinese population in which seroconversion in a group of 285 patients from 3 hospitals showed IgG positivity for all cases beyond 17 to 19 days. Bryan et al[9] demonstrated timing of seroconversion for an Idaho population. Additional studies are lacking for the US population. The goals of this study were to ascertain key performance metrics of analytical specificity and cross-reactivity for a SARS-CoV-2 IgG serologic assay, perform a detailed cross-sectional and serial assessment of IgG and IgM antibody responses in suspected COVID-19 patients, and determine their relation to disease severity.

Materials and Methods

Patient Samples

This study was approved by the University of Texas Southwestern Institutional Review Board. A total of 967 individuals (995 total specimens) were included in this study, including 656 healthy controls, 29 patients with systemic lupus erythematosus, 20 with rheumatoid arthritis, 90 with previous positive respiratory viral PCR panel and/or cytomegalovirus (CMV) IgG, and 172 suspected cases of COVID-19 **Figure 1**. Suspected cases comprised instances of new onset or acutely worsening fever, respiratory, or GI symptoms (applied to all PCR positive cases, and 55 of 96 PCR-negative cases), reported exposure to a COVID-19 patient, and/or requiring rule-out testing preprocedure.

SARS-CoV-2 IgG Testing

SARS-CoV-2 IgG (Abbott 06R86) testing was performed on the Abbott ARCHITECT i2000SR in accordance with manufacturer's specifications. The test is a chemiluminescent microparticle immunoassay (CMIA) for qualitative detection of IgG antibodies against SARS-CoV-2 nucleocapsid protein (NCP) in human serum and plasma. Strength of response in relative light units reflects quantity of IgG present, and is compared to a calibrator

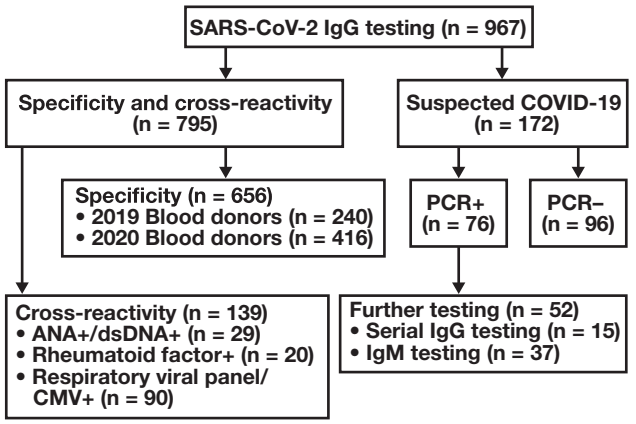

Figure 1 Study cases. Nine-hundred and sixty-seven unique individuals provided samples for SARS-CoV-2 IgG testing, including 15 with serial samples available. IgM testing was performed on a group of 37 specimens (17 IgG+, 20 IgG–). CMV, cytomegalovirus; PCR, polymerase chain reaction.

to determine the calculated index (specimen/calibrator [S/C]) for a sample (with positive at 1.4 or greater).

SARS-CoV-2 IgM Testing

IgM antibody reactivity against SARS-CoV-2 NCP was measured using a laboratory-developed proteome array as described previously.[10] Briefly, NCP expressed in baculovirus insect cells (Sino Biological) and in *Escherichia coli* (Creative Diagnostics) along with control proteins (human IgM and anti–human IgM) were printed onto nitrocellulose membrane–coated slides (Grace Bio) in sextuple using a Nanoplotter NP2.1 inkjet printer (Gesim). Patient serum samples were diluted 1:100 and incubated with the antigens on the array and the IgM antibody specificities detected with Cy5-conjugated anti–human IgM (1:1,000, Jackson ImmunoResearch). The array was scanned using Genepix 4400A scanner (Molecular Device) at wavelength 635 nm. The resulting images were analyzed using Genepix Pro 7.0 software (Molecular Devices). The median of the signal intensity for each spot was calculated and the local background around the spot subtracted, and data obtained from sextuple spots were averaged. The background subtracted signal intensity was normalized to the average intensity of the total human IgM (internal positive control) to generate normalized signal intensity (NSI). Samples with NSI of 25 or higher were considered positive for IgM. The NSI of NCP IgM was used to generate heat maps using Cluster and Treeview software (http://bonsai.hgc.jp/~mdehoon/software/cluster/index.html).

DOI: 10.1093/ajcp/aqaa123

Analytical Specificity

Specificity was evaluated using 240 banked plasma samples collected prior to the COVID-19 pandemic (blood donors September through November 2019) and samples from an additional 416 healthy donors without recent illness collected from March to April 2020.

Cross-Reactivity Studies

Cross-reactivity specimens related to prior respiratory illness and/or CMV IgG positivity (90 total samples) were collected by cross referencing banked serum in the HLA lab (January 1, 2015- September 30, 2019) with patients who had previously tested positive for CMV IgG, influenza A/B, respiratory syncytial virus (RSV), or an endemic coronavirus (NL63, 229E, OC43, or HKU1) by viral molecular tests. As the patients may have been immunosuppressed, we included only those specimens having normal or high levels of total IgG (measured alongside SARS-CoV-2) with no infusion of intravenous immunoglobulin in the preceding 3 months. Likewise, we tested 29 samples from lupus patients (collected 2004-2007) who were positive for multiple autoantibodies (100% ANA, 62% anti-dsDNA, 75% anti-U1RNP, 55% anti-Sm, 34% anti-Ro52, and 24% anti-La) and an additional 20 samples (collected 2011-2014) from rheumatoid arthritis patients positive for rheumatoid factor (85% were also anti–cyclic citrullinated peptide positive).

Agreement With PCR-Based Testing

Agreement with PCR-based molecular testing was determined using 172 plasma samples collected (147 lithium heparin, 13 EDTA, and 12 sodium citrate) from suspected COVID-19 cases with prior or same-day PCR-based nasopharyngeal swab testing on the m2000 Abbott RealTi*m*e SARS CoV-2 assay or the Abbott ID NOW COVID-19 assay. Testing using the m2000 platform was performed either alone, or as reflex testing in the setting of a negative result using the ID NOW platform. Patient charts were reviewed to determine time between symptom onset (fever, respiratory symptoms, or gastrointestinal complaints) and severity of condition (whether or not intensive care was required). A subgroup of 37 PCR-positive cases (17 IgG positive, 20 IgG negative) selected based on sample availability were additionally evaluated for SARS-CoV-2 IgM.

Serial Patient Monitoring

For 15 PCR-positive cases, 2 to 6 serial measurements were performed using available residual plasma samples. IgG levels and seroconversion based on the calculated index (S/C) were tracked over time.

Statistics

The calculated index (S/C, IgG level) was provided by the instrument. When multiple values of IgG S/C were compared, a mean and standard deviation were calculated. Student *t* test was used to compare 2 groups of nonparametrically distributed data, and a *P* value less than .05 was considered significant.

Results

Analytical Specificity

The SARS-CoV-2 IgG assay was calibrated followed by an imprecision study performed over a period of 5 consecutive days and was found to be acceptable. Analytical specificity of the assay was evaluated with samples from healthy blood donors, and none of these samples (0/656) were positive for virus-specific IgG ▮Table 1▮; the mean index value was 0.04, well below the cutoff of 1.4 for a positive index value.

Cross-Reactivity Studies

The cross-reactivity results for respiratory viral infection appear in Table 1. None of the 23 CMV IgG positive samples were COVID-19 IgG positive (0/23, 0%). No cases associated with prior influenza A+ (n = 8), influenza

▮Table 1▮
SARS-CoV-2 IgG Results in Healthy Donors and Cases of Previous Respiratory Viral Infection

Sample Type	IgG, AU/mL
Blood donors	
Mean ± SD	0.039 ± 0.065
No. of positive tests (%)	0/656 (0%)
CMV IgG+	
Mean ± SD	0.07 ± 0.067
No. of positive tests (%)	0/23 (0%)
Influenza A+	
Mean ± SD	0.13 ± 0.19
No. of positive tests (%)	0/8 (0%)
Influenza B+	
Mean ± SD	0.11 ± 0.17
No. of positive tests (%)	0/7 (0%)
RSV+	
Mean ± SD	0.035 ± 0.029
No. of positive tests (%)	0/6 (0%)
Coronavirus+	
Mean ± SD	0.050 ± 0.079
No. of positive tests (%)	0/47 (0%)

AU, absorbance unit; CMV, cytomegalovirus; RSV, respiratory syncytial virus; SD, standard deviation.

DOI: 10.1093/ajcp/aqaa123

B+ (n = 7), RSV+ (n = 6), or all 4 types of human coronavirus (n = 47) demonstrated cross-reactivity. Similarly, none of the sera associated with clinically significant levels of autoantibodies produced a positive antiviral IgG test result (0/49, 0%) Table 2. Highest mean of S/C ratio observed was 0.05 for human coronaviruses and 0.08 for rheumatic diseases.

Cross-Sectional Data for SARS-CoV-2 IgG and IgM

Of 172 potential COVID-19 cases included in the study described, 76 were confirmed positive by PCR methods. Overall, 29 of 76 (38%) tested positive for SARS-CoV-2 IgG. The time course of symptom onset revealed increasing IgG positivity rates Table 3 from less than 3 days (1/15, 7%) to 3 to 7 days (8/27, 30%) and 8 to 13 days (5/15, 33%), to being the highest after 14 days (5/6, 83%). IgG positivity was high (10/13, 77%) for patients with indeterminate time from symptom onset. IgM testing Figure 2 performed on 37 PCR-positive specimens showed positivity in 9 of 17 (53%) IgG-positive cases and, interestingly, in 7 of 20 (35%) IgG-negative samples. IgM positivity occurred at larger proportion for less than 3 days (3/6, 50%) compared to IgG, but at similar rates overall at days 3 to 7 (4/11, 36%), days 8 to 13 (4/11, 36%), and after 2 weeks (4/5, 80%). IgM positivity was low (1/4, 25%) for patients with indeterminate time from symptom onset. SARS-CoV-2 IgG antibody results agreed with the PCR-negative samples for 96 of 97 (99%) of cases, including 55 instances of patients with new or acute-on-chronic symptoms suspicious for COVID-19 and with known time of onset.

Table 2
Autoantibody Interference

Autoantibody	IgG Positivity Rate (%)
Systemic lupus erythematosus (n = 29)[a]	
ANA	0/29 (0%)
Anti-DNA	0/18 (0%)
Anti-U1RNP	0/22 (0%)
Anti-Sm	0/16 (0%)
Anti-Ro52	0/10 (0%)
Anti-La	0/7 (0%)
Rheumatoid arthritis (n = 20)[b]	
Rheumatoid factor (RF)	0/20 (0%)
Anti-CCP	0/17 (0%)

CCP, cyclic citrullinated peptide.
[a]Cases (29 total) were positive for 1 or more of the autoantibodies listed.
[b]All cases were RF positive, with 17/20 (85%) also anti-CCP positive.

Table 3
SARS-CoV-2 IgG Positive Agreement by Days Post Symptom Onset

Time From Symptom Onset, d[a]	IgG Positivity Rate (%)	IgM Positivity Rate (%)
<3	1/15 (7%)	3/6 (50%)
3-7	8/27 (30%)	4/11 (36%)
8-13	5/15 (33%)	4/11(36%)
≥14	5/6 (83%)	4/5 (80%)
Indeterminate	10/13 (77%)	1/4 (25%)
Total	29/76 (38%)	16/37 (43%)

[a]For reverse transcription polymerase chain reaction confirmed SARS-CoV-2 cases.

Disease Severity and IgG and IgM Value

We hypothesized that a more severe disease course was related to an increased immune response, which may result in a higher level of SARS-CoV-2 IgG antibody reactivity. Cytokine storm has been implicated as a potential life-threatening event in SARS-CoV-2 infection, and this would activate many aspects of the immune system including the humoral antibody response. We compared IgG levels from all SARS-CoV-2 PCR-positive patients who had a mild/moderate disease course to those who had severe disease (admitted to the intensive care unit [ICU]), and there was no difference in IgG antibody levels between the 2 groups Figure 3A. Given the impact of time from symptom onset on IgG measurement, IgG indices were additionally plotted relative to day post symptom onset Figure 3B, showing a predominant overlap between mild/moderate and severe groups. Similarly, IgM levels were not much different in mild/moderate and severely affected patients Figure 3C and Figure 3D. It is possible that the course of IgG levels was qualitatively different for severe patients, so data from serially collected IgG samples were plotted against day of symptom onset Figure 4 for a select group with serially available samples. Severely affected patients were tracked longer, because they were hospitalized longer, but a similar early increase in antibody indices was observed in mild/moderately affected patients when compared to severely affected patients. Interestingly, 1 patient was seronegative even on day 28, but this was attributed to immunosuppression to prevent cardiac transplant rejection.

Serial Patient Monitoring and Seroconversion

Thirty-eight samples were available from 13 patients with known date of symptom onset and 4 samples from 2 patients with indeterminate date of symptom onset. Within this group, 77% (10/13) became IgG positive, including specifically 0% (0/8) for less than 3 days post symptom onset, 33% (3/9) at 3 to 7 days post symptom onset, 86% (6/7) at 8 to 13 days post symptom onset, and 91% (10/11) at more than 14 days Figure 4. For those where seroconversion was not observed, samples were only available for less than 7 days from symptom onset for 2 cases or the patient was subject to significant

DOI: 10.1093/ajcp/aqaa123

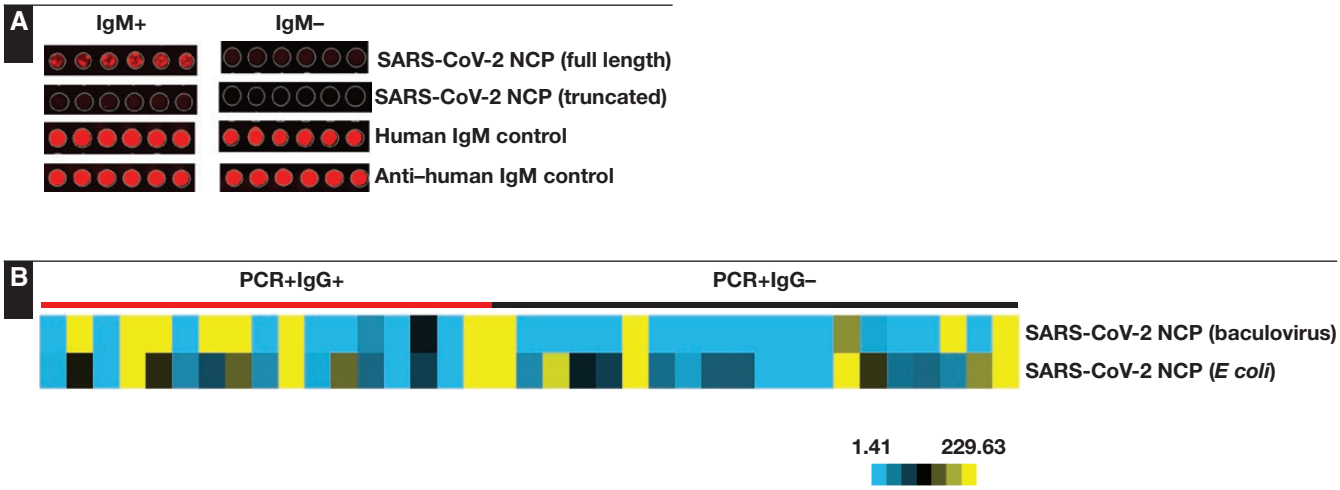

Figure 2 IgM proteome array analysis. Array images of IgM+ and IgM– samples are shown (**A**) as well as a heatmap (**B**) of IgM anti–SARS-CoV-2 nucleocapsid protein (NCP) for IgG+ and IgG– cases of confirmed COVID-19. PCR, polymerase chain reaction.

immunosuppression. For the 2 cases with indeterminate date of symptom onset, 1 demonstrated seroconversion between samples 11 days apart. The second case did not demonstrate seroconversion over 9 days.

Discussion

Here we confirmed the high specificity reported by the manufacturer for a SARS-CoV-2 IgG serologic assay, using comparatively larger groups for certain rheumatologic conditions and infections. Notably, CMV IgG did not cause assay interference despite potential false positivity reported by the manufacturer. Rheumatoid factor is an anti-human antibody (IgM or IgG) that, if complexed with other human immunoglobulins, could falsely increase positivity of an assay. However, we observed no interference by rheumatoid factor in 20 samples. Testing 47 samples with prior endemic coronavirus infection yielded no false positives. Negative agreement between IgG and PCR indicated only 1 case testing IgG positive despite negative PCR testing. This initial PCR result was later determined to be a false negative based on evaluation using an alternative molecular platform. Positive agreement with PCR was lower in the early stages of infection, increasing with time from symptom onset, yet not as quickly compared to the manufacturer's report.

Overall, our results largely corroborate and add to the findings by Bryan et al[9] who evaluated the same platform. That study showed high specificity in testing 1,020 specimens submitted for herpes simplex virus Western blot serology from before the COVID-19 pandemic. As such, specificity and cross-reactivity were not specifically addressed in the setting of underlying rheumatologic disease or previous endemic coronavirus. A possible difference between our findings was sensitivity after 14 days of symptoms. In our study, a single negative case attributed to a patient with marked immunosuppression resulted in reduced sensitivity beyond 14 days. An unknown factor in similar studies is the number of cases included with severe underlying immunosuppression. For instance, a recent publication by Long et al[8] also indicated 100% IgG positivity at 17 to 19 days. This latter study utilized a different assay and focused on a population in China and was thus not as comparable. However, the same question persists regarding the makeup of comorbidities in the test population and highlights that discrepancies may arise in antibody response when comparing serology in unequal groups. Nonetheless, within our serial testing group, given the higher number of cases beyond 14 days, we did encounter sensitivity of 91% (10/11), which was closer to previous findings.[8,9] As with Bryan et al,[9] we have noticed alternative cutoff values for IgG level could be used with potentially beneficial diagnostic effects. As an example, lowering the cutoff by half (to 0.7) would capture an additional 4 cases with midrange days (5-11 days) from symptom onset without any loss in specificity based on the PCR result.

Long et al[8] had reported a counterintuitive peak of IgM positivity (20-22 days) later than for IgG positivity (17-19 days). IgM responses usually peak within the first week after infection and before IgG class switching. When early in infection, IgG may not yet be positive. When we tested samples for IgM reactivity, 7 IgG negative cases

DOI: 10.1093/ajcp/aqaa123

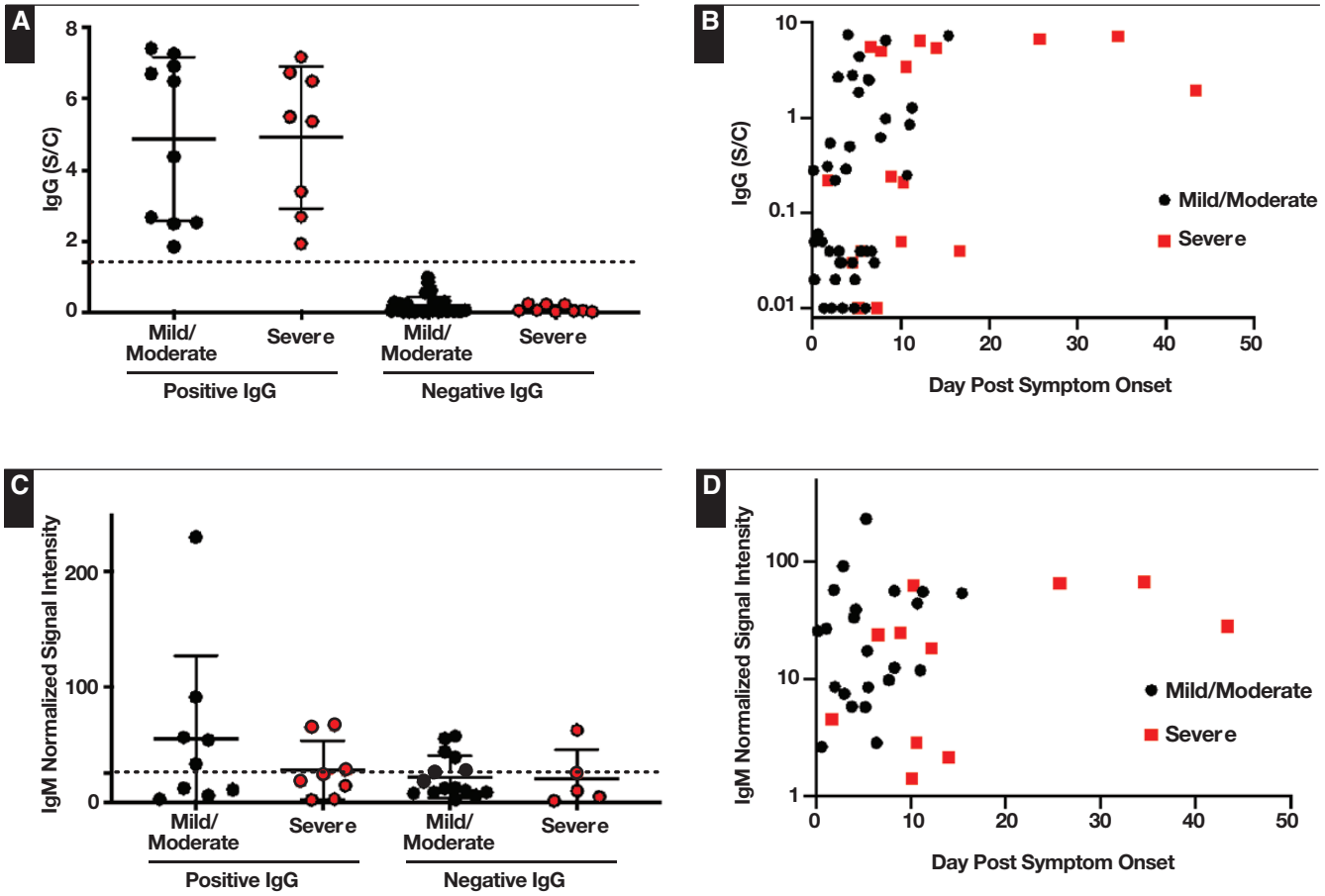

Figure 3 Antibody levels by disease severity for polymerase chain reaction–positive subjects. **A**, SARS-CoV-2–specific IgG antibody results that were positive or negative were divided by disease severity and plotted against number of days from symptom onset (**B**). **C**, SARS-CoV-2 nucleocapsid–specific IgM antibody results were divided by IgG positivity to demonstrate when a sample was IgM+ but IgG–. **D**, IgM antibody levels were plotted against number of days from symptom onset. Middle line is the mean; bars represent standard deviation. The dotted lines represent negative cutoff levels. S/C, specimen/calibrator.

were positive for IgM. These samples were positive for IgM earlier than IgG, ranging from 0 to 11 days from symptom onset. This increased the sensitivity by 35% within the IgG-negative samples tested (7/20) and improved diagnostic utility by 9% overall (7/76). These findings emphasize that results will differ between assays focused on IgG alone compared to those that separately measure multiple immunoglobulin classes or test for total antibodies (eg, Roche).

As described, we segregated our IgG and IgM results based on severity (ICU care vs no ICU care). Long et al[8] indicated that a severe disease course resulted in a high IgG level during the second week of disease that becomes indistinguishable from milder cases after 14 days. We did not observe such a difference using a different CMIA method. This could be due to fewer patient samples, but the significance of their finding was very strong, which indicates it should have replicated were it a real phenomenon. A limitation in our study, however, is that, although the S/C values are retrievable for the assay, the package insert only describes qualitative reporting based on the cutoff value. Furthermore, it should be noted that antibody levels may correlate with other factors such as how long disease lingers before final resolution, long-term complications, or period of communicability. IgM levels in our study based separately on the proteome microarray also showed no significant difference when analyzed by disease severity. Thus, antibody levels themselves do not appear to reflect disease severity, although serologic reactions not assessed here could potentially do so.

A major hurdle to validation was access to patients after a sufficient period of infection because most patients presented before 14 days from symptom onset. Limited resources and self-quarantine measures have impaired repeated testing for serial testing at a later date.

DOI: 10.1093/ajcp/aqaa123

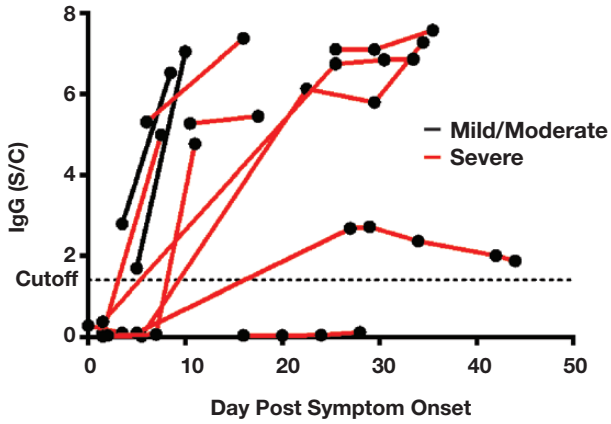

Figure 4 Serial IgG measurements. For patients with multiple samples taken, the IgG level was plotted against time from symptom onset. Dots represent the IgG level at a specific time. Samples from the same patient are connected by either red (severe cases, based on intensive care unit admission) or black (mild/moderate cases) lines. The dotted line indicates the threshold for a sample being positive. S/C, specimen/calibrator.

Consequently, less data on mild and moderate patients existed compared to patients admitted to the ICU. We do, however, have the advantage of reviewing medical charts to find examples of false negative by PCR and false negative by serology. These examples indicate that molecular and serologic testing have complementary roles in tracking exposure to SARS-CoV-2. Our data do not provide information on how long IgG stays positive in the long term or whether it specifically confers immunity.

Conclusions

As communities continue to grapple with the COVID-19 pandemic, reliable measures of previous exposure and immunity are essential. Several platforms are now coming into broader clinical use, providing a window into the SARS-CoV-2 antibody response. Widespread efforts to track SARS-CoV-2 patients for antibody development will clarify expectations for when testing should return positive, situations in which seroconversion may fail, and what the antibody response can tell us in patients with active infections.

Corresponding author: Alagarraju Muthukumar, PhD, DABCC; Alagarraju.Muthukumar@UTSouthwestern.edu.

**First authors.*

Acknowledgments: We thank Clements University Hospital core laboratory staff, especially Brittany Diaz, Sylynn Garza, and Charles Alexis, for serology testing and archiving of all the samples related to this study. We also acknowledge Carter Blood Care (Bedford, TX) for their archived patient samples for specificity studies, the human leukocyte antigen laboratory at the University of Texas Southwestern for their help in retrieving banked serum samples for cross-reactivity studies, and Ashley Young, MD, for aid in chart review.

References

1. Zhou P, Yang X-L, Wang X-G, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*. 2020;579:270-273.
2. Coronaviridae Study Group of the International Committee on Taxonomy of Viruses. 2020. The species severe acute respiratory syndrome-related coronavirus: classifying 2019-nCoV and naming it SARS-CoV-2. *Nat Microbiol*. 2020;5:536-544.
3. Centers for Disease Control and Prevention. CDC 2019-novel coronavirus (2019-nCoV) real-time RT-PCR diagnostic panel for emergency use only: instructions for use. https://www.fda.gov/media/134922/download. Accessed June 12, 2020.
4. Corman VM, Landt O, Kaiser M, et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. *Euro Surveill*. 2020. doi: 10.2807/1560-7917.ES.2020.25.3.2000045.
5. World Health Organization. Coronavirus disease (COVID-19) technical guidance: Laboratory testing for 2019-nCoV in humans. https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/laboratory-guidance. Accessed March 31, 2020.
6. Chan JF-W, Yip CC-Y, To KK-W, et al. Improved molecular diagnosis of COVID-19 by the novel, highly sensitive and specific COVID-19-RdRp/Hel real-time reverse transcription-PCR assay validated in vitro and with clinical specimens. *J Clin Microbiol*. 2020;58:e00310-00320.
7. Chu DKW, Pan Y, Cheng SMS, et al. Molecular diagnosis of a novel coronavirus (2019-nCoV) causing an outbreak of pneumonia. *Clin Chem*. 2020;66:549-555.
8. Long Q-X, Liu B-Z, Deng H-J, et al. Antibody responses to SARS-CoV-2 in patients with COVID-19. *Nat Med*. 2020;26:845-848.
9. Bryan A, Pepper G, Wener MH, et al. Performance characteristics of the Abbott Architect SARS-CoV-2 IgG assay and seroprevalence in Boise, Idaho. *J Clin Microbiol*. 2020. doi: 10.1128/JCM.00941-20.
10. Zhen QL, Xie C, Wu T, et al. Identification of autoantibody clusters that best predict lupus disease activity using glomerular proteome arrays. *J Clin Invest*. 2005;115:3428-3439.

DOI: 10.1093/ajcp/aqaa123