Prevalence and Longevity of SARS-CoV-2 Antibodies Among Health Care Workers

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Background. Understanding severe acute respiratory syndrome coronavirus 2 antibody prevalence in a spectrum of health care workers (HCWs) may provide benchmarks of susceptibility, help us understand risk stratification, and support enactment of better health policies and procedures.

Methods. Blood serum was sampled at enrollment and 8-week follow-up from HCWs (n = 3458) and from community first responders (n = 226) for immunoglobulin G (IgG) analyses. Demographics, job duties, location, and coronavirus disease 2019–related information were collected.

Results. The observed IgG antibody prevalence was 0.93% and 2.58% at enrollment (May/June) and 8-week follow-up (July/August), respectively, for HCWs, and 5.31% and 4.35% for first responders. For HCWs, significant differences (P < .05) between negative and positive at initial assessment were found for age, race, fever, and loss of smell, and at 8-week follow-up for age, race, and all symptoms. Antibody positivity persisted at least 8 weeks in all positive HCWs.

Conclusions. We found considerably lower antibody prevalence among HCWs compared with other published studies. While rigorous safety process measures instituted in our workplace and heightened awareness at and outside of the workplace among our HCWs may have contributed to our findings, the significant discrepancy from our community prevalence warrants further studies on other contributing factors.

Keywords. antibody; COVID-19; first responders; health care workers; IgG; immunity; prevalence; longevity; SARS-CoV-2.

Since first reported in Wuhan, China, in December 2019, coronavirus disease 2019 (COVID-19) has given rise not only to a tumultuous health care and socioeconomic crisis worldwide, but also to unprecedented psycho-social trauma to the world community, including health care workers (HCWs) in particular, with potentially wide-ranging downstream impact.

COVID-19’s extraordinary infectivity, given its novel nature and presymptomatic transmission, fueled its wide and wild spread across and within countries, with confirmed cumulative cases of >9.4 million in the United States and 47.4 million worldwide as of November 3, 2020. A recent review article reports that ~40%–45% of those infected with severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) can be asymptomatic for an extended period (eg, beyond 14 days) or never develop symptoms, suggesting a much wider spread of the virus than confirmed cases indicate [1]. The Centers for Disease Control and Prevention (CDC) and others estimate that 10-fold or more infections exist for every confirmed case [2–4]. Some subsampled confined cohorts have demonstrated asymptomatic prevalence as high as 96% [1, 5].

With the unprecedented effort that has gone into the development of SARS-CoV-2 vaccines, it is of great importance to better understand the extent of transmission within health facilities and the susceptibility of HCWs to infection, so that optimal early vaccine deployment and prioritized preventive strategies can be developed.

Sero-surveillance studies have been conducted to estimate SARS-CoV-2 antibody prevalence in various countries and settings, including among blood donors [6–8]. Such estimates help better estimate the total numbers of infected individuals to calculate the true infection mortality rate (vs the case fatality rate). True infection prevalence and its change over time would better explain the venues of asymptomatic and pre- or perisymptomatic transmissions, environmental differences, and possibly duration of antibody presence. This is particularly of interest in acute health care settings.
Previously reported results on sero-surveillance have varied greatly due to factors including sample size and geography (eg, high active infection zones vs low), with 57% prevalence in Bergamo, Italy [9], 12.5% in New York State [10], 11.5% more recently in Orange County [11], 4.7% in Los Angeles County [12], and 2.8% in Santa Clara County, California [13], earlier in the pandemic. The reliability of some of the early methodology for measuring antibodies might have also contributed to these varying results [14].

For HCWs, SARS-CoV-2 antibody prevalence has been sparsely reported but also with similar variability with regard to sample size, range of results, timing of sampling, and methodologies. Prevalence rates across different health care settings have ranged widely, with 89.3% in Wuhan, China (n = 424) [15], 35.8% in New York City (n = 285) [16], 13.7% in the greater New York City area (n = 40329) [17], 7.4% in Milan, Italy [18], and 2.67% in Denmark (n = 28792). Higher associations between positivity and job duties, younger age (<30), and self-reported suspicion of prior COVID-19 exposure and prior positive polymerase chain reaction (PCR) testing [19] were found. Determining antibody prevalence in a wide spectrum of randomly sampled HCWs using a validated and accurate serum assay and repeated sampling over time to measure duration of antibody presence may help stratify the workforce for risk, limit transmission across different health care settings, enact better mitigation processes and procedures, and better prioritize future vaccine delivery to front-line workers.

Our previous study, conducted during a “shelter at home” mandate in May–June 2020, observed an antibody prevalence of 1.06% (adjusted antibody prevalence for test sensitivity and specificity of 1.13%) among our HCWs (n = 2924) at Hoag Memorial Hospital Presbyterian, Orange County, California. This was much lower than those reported by other studies [20]. Therefore, to further analyze the evolving prevalence of antibodies among HCWs, the present study expands our initial study in a larger HCW cohort and also reports on 8-week follow-up to investigate the longevity of antibody presence. The results were compared with those from community first responders (eg, fire fighters, police officers) as well as those calculated from laboratory test results ordered by community physicians.

METHODS

Patient Consent Statement
Institutional review board (IRB) approval was obtained for this study from the Providence St. Joseph Health IRB (IRB# 2020000337). In accordance with the ethical standards of the Helsinki Declaration (1964, amended most recently in 2008) of the World Medical Association, written informed consents were obtained in person originally, then transitioned to electronic consent format starting June 19, 2020.

Subject Recruitment
Study HCW subjects were recruited by direct email notifications to the entire employee workforce (6500+ individuals) and independent medical staff (1600+ physicians), whose work locations include 2 hospital campuses, 9 health centers, 13 urgent care locations, and other clinical and administrative facilities all within a ~20-mile radius. Similarly, study subjects from first responders were recruited from fire and police departments in Orange County, California, by direct email notifications.

Enrollment and Data Collection
Those who were enrolled through in-person consent were surveyed for ethnicity, job duties, location, COVID-19 symptoms, a self-reported PCR test history with test date if available, travel record since January 2020, and existence of household contacts with COVID-19. Those who were enrolled through electronic consent format answered the same survey online at the time of consenting. The COVID-19 symptoms survey included fever, sore throat, cough, runny nose, and loss of sense of smell, with loss of taste added at 8-week follow-up. Using reported job duties and locations, each HCW subject was classified into (a) high (eg, MD, RN, PA, emergency care tech, intensive care unit [ICU] tech), (b) medium (eg, therapist, phlebotomist, medical tech), or (c) low (eg, admin, coder, billing, lab tech/scientist, IT) risk groups to approximate levels of direct exposure to COVID-19 patients. The antibody test results ordered by community physicians for clinical care purposes were aggregated from our laboratory database for comparative purposes. However, no additional data including COVID-19 symptoms were available for this sample.

Blood Sample Collection
The first blood sample (~5 mL) was collected for serum analysis for immunoglobulin G (IgG) antibodies to SARS-CoV-2 at the time of in-person consent, or following electronic consent at 2 main hospital campuses. With the exception of 16 subjects, blood sample collection was within 7 days of electronic consent (M [SD], 1.77 [1.83]). Eight weeks after the first blood sample, the second sample was collected.

IgG Antibodies to SARS-CoV-2 Analysis
Serum analysis for IgG antibodies to SARS-CoV-2 utilized the Ortho Clinical Diagnostics VITROS XT 7600 platform. A 5-mL peripheral draw venous blood sample was collected from each subject into a gold top serum separator vacutainer tube (BD Medical). Samples were centrifuged within 2 hours of collection at 4500 RPM for 5 minutes (RCF 3060). Aliquots were analyzed with calibrated lots of Anti-SARS-CoV-2 IgG Reagent Pack on the VITROS XT 7600 according to the manufacturer’s instructions for use [21]. Positive and negative quality controls were run daily before sample analysis (Ortho Diagnostics Anti-SARS-CoV-2 IgG Control). At the time of writing, this
IgG test is approved only for use under the Food and Drug Administration's Emergency Use Authorization (EUA) and is also used in CDC studies [22].

Manufacturer sensitivity and specificity claims for the Ortho Clinical Diagnostics VITROS Anti-SARS-CoV-2 IgG assay are 100% (407/407) negative agreement (95% CI, 99.1%–100.0%) in 407 presumed SARS-CoV-2 antibody–negative subjects and 87.5% (42/48) positive agreement (95% CI, 74.8%–95.3%) in 48 PCR-positive subjects, with days from positive PCR ranging from 1 day to 22 days and days from onset of symptoms ranging from 12 to 32 days. In-house validation studies were conducted with 35 samples from subjects with a known positive SARS-CoV-2 PCR test a mean of 43 days out from the positive PCR test date (range, 38–48 days), and 50 samples from subjects with a known negative SARS-CoV-2 PCR test. Of 31 PCR samples, 29 were positive for SARS-CoV-2 IgG antibody. All 50 of the PCR-negative samples were SARS-CoV-2 IgG antibody negative. Thus, a sensitivity of 93.6% (95% CI, 78.6%–99.2%) and specificity of 100% (95% CI, 92.9%–100.0%) were calculated for the Ortho Diagnostics VITROS Anti-SARS-CoV-2 IgG assay in our laboratory on the Ortho Clinical Diagnostics VITROS XT 7600 automated instrument platform and were adopted in this study.

Data Analysis
Data were examined for HCWs and first responders at first and second blood draw results, each comparing antibody negativity vs positivity. Nonparametric tests for group differences were performed for demographics and 5 symptoms of COVID-19 at the first blood draw, with an additional 1 symptom at the second blood draw. The effect of occupational risk was also evaluated for HCWs. Mann-Whitney U tests were used for assessing group difference in age, and a series of 1-sided Fisher exact tests were used for the remaining categorical factors; for group differences in race (a 7 × 2 table) and occupational risk (a 3 × 2 table), the Mehta-Patel algorithm was applied [23]. Logistic regressions were used in assessment of continuous and categorical factors for HCWs at baseline and 8-week follow-up, applying the King-Zeng correction for rare events [24]. A P value <.05 was used for statistical significance. For all analyses, the Stata statistical software package, edition 15, was used [25].

RESULTS
After excluding subjects for missing data, the final analyses included 3458 subjects from the first blood draw and the subset of those who returned for the second draw (n = 2754; 79.6% return rate) for HCWs, and 226 subjects from the first blood draw and the subset of those who returned for the second draw (n = 92; 40.7% return rate) for first responders. At baseline measurement, demographic comparison indicated no significant difference of age for HCWs (M [SD], 42.33 [12.13]) and first responders (M [SD], 42.04 [8.61]) but significant differences for both gender for HCWs (73% female) and first responders (9% female; P < .001) and race for HCWs (eg, 50% White) and first responders (78% White; P < .001).

Among HCWs’ initial blood draw, 32 antibody-positive cases (3426 negative) were identified, with an observed prevalence of 0.93% (exact binomial 95% CI, 0.63%–1.30%). Accounting for test sensitivity (93.6%) and specificity (100%), an adjusted prevalence of 0.98% (exact binomial 95% CI, 0.68%–1.37%) was calculated, indicating 34 positive cases (3424 negative) after adjustment. At their 8-week follow-up blood draw (n = 2754), 71 antibody-positive cases (2683 negative) were identified, with an observed prevalence of 2.58% (exact binomial 95% CI, 2.02%–3.24%). Of the original 32 positive subjects, 28 remained positive (4 did not return for the second blood draw) with an additional 43 new cases during an 8-week period (Table 1A). An adjusted prevalence of 2.76% (exact binomial 95% CI, 2.18%–3.44%) was calculated, indicating 76 positive cases (2678 negative) after adjustment. Table 2 summarizes HCW sample characteristics and group differences.

Nonparametric tests for group differences were performed for demographics and 6 symptoms of COVID-19. Significant differences between observed negative vs positive cases at initial assessment were found for age (z = 2.396), race, fever, and loss of smell. At 8-week follow-up, significant differences were found for age (z = 4.718), race, and all symptoms (all P < .05). Occupational risk did not contribute significantly to negative vs positive group differences at either blood draw time point. Logistic regression analysis at initial assessment identified loss of smell as the sole significant factor (P < .05), with a 66.066

| Table 1. Sample Size Summary |
|--------------------------------|
| (a) Health Care Workers        |
| 1st Draw Antibody Results  | 2nd Draw Antibody Results |  |
| Negative  | 3426               | 2683          |
| Positive  | 32                 | 4            |
| Total  | 3458               | 2754          |
| (b) First Responders          |
| 1st Draw Antibody Results  | 2nd Draw Antibody Results |  |
| Negative  | 214                | 88*           |
| Positive  | 12                 | 1            |
| Total  | 226                | 92            |

*One subject at second draw was missing at first draw.
times increase in relative risk (95% CI, 13.964–238.448). At 8-week follow-up, logistic regression significant factors were age (relative risk [RR], 0.955; 95% CI, 0.931–0.982), fever (RR, 5.112; 95% CI, 2.391–11.519), sore throat (RR, 0.204; 95% CI, 0.069–0.568), loss of smell (RR, 9.204; 95% CI, 3.246–23.934), and loss of taste (RR, 3.259; 95% CI, 1.145–8.771).

Among first responders’ initial blood draw, 12 antibody-positive cases (214 negative) were identified, with an observed prevalence of 5.31% (exact binomial 95% CI, 2.77%–9.09%). Accounting for test sensitivity and specificity, an adjusted prevalence of 5.75% (exact binomial 95% CI, 3.10%–9.64%) was calculated, indicating 13 positive cases (213 negative) after adjustment. Significant differences were found for the symptoms of fever, cough, and loss of smell (all $P < .05$). At their 8-week follow-up blood draw (n = 92), 4 antibody-positive cases (88 negative) were identified, with an observed prevalence of 4.35% (exact binomial 95% CI, 1.20%–10.76%)—an original 1 case remained antibody positive (11 did not return for the second blood draw) with an additional 3 new cases during an 8-week period (Table 1B). Adjusted prevalence was equal to observed prevalence. See Table 3 for first responder sample characteristics and group differences.

Given our observed 8-week antibody persistence in HCWs, we also conducted an extrapolated prevalence calculation for the 8-week follow-up to include those with antibody positives at the first blood draw who did not return for the second draw (Table 1). For HCWs, adding these 4 cases (total positive n = 75) resulted in a prevalence of 2.72% (exact binomial 95% CI, 2.14%–3.40%). Similarly, adding 11 cases in the first responders (positive n = 15) resulted in a prevalence of 14.56% (exact binomial 95% CI, 8.39%–22.88%). Table 4 summarizes observed, adjusted, and extrapolated prevalence.

Among HCWs with a previous PCR-confirmed diagnosis of COVID-19 (n = 75), 40 (53.3%) were antibody positive and 35 negative (46.7%) at 8-week follow-up. Those 35 were also antibody negative at enrollment and had no history of hospitalization or severe illness. They also reported no or 1–2 COVID-19 symptoms, except 2 cases with 4 and 5 symptoms, respectively, indicating possible rapid antibody decay in those with no to mild COVID-19 symptoms [26]. While gender, race, and occupational risk did not significantly contribute to group differences between antibody negatives vs positives, age and frequency of all symptoms were significantly different (all $P < .05$), with positives being significantly younger and presenting more symptoms than negatives. Among those with available PCR test date, the time between PCR and antibody test ranged from 16 to 94 days (M [SD], 41.33 [23.27] days) for the negatives (n = 9) and from 12 to 151 days (M [SD], 59.69 [41.90] days) for the positives (n = 35), with no significant difference ($t(42) = –1.26; P = .215$) (Table 5 summarizes group differences).

**DISCUSSION**

The present study found a considerably lower adjusted antibody prevalence (0.98%) on initial sampling (95.9% blood drawn during a “shelter at home” mandate in May–June 2020) among HCWs, confirming our initial findings [20] in
a larger cohort. The community prevalence during this early period was relatively low (3.64%), as reflected by those tested by physician order in our community, and also reflected in adjusted prevalence among the first responders tested in this study (5.75%), though both these prevalence results were considerably higher than that of our HCWs. While selection bias likely affected our estimate of community prevalence in serum drawn from physician orders, the early antibody prevalence studies in other locales of Southern California support our estimate [12, 13]. The second period (100% blood drawn in July–August 2020) reflected wider community transmission after partial state re-opening, as evidenced by a spike of hospitalization in Orange County, yet the higher adjusted prevalence rate in our HCWs (2.76%) was still well below the considerably higher prevalence reported by a recent seroprevalence study in our community (11.5 %) [11] and a estimate from laboratory testing results ordered by community physicians for patient care purposes (22.47%).

Several factors for the relatively low sero-prevalence in our HCWs may explain current findings. Upon caring for the first COVID-19 patient in California (the third in the United States), our organization reacted immediately. We established an internal weekly COVID-19 task force meeting and opened regular communication with the Orange County Healthcare Agency as well as the CDC to stay current with the rapidly changing guidelines from county, state, and federal agencies. The task force oversaw a rigorous approach to preparedness, including resource allocation (eg, personal protective equipment, cohorted emergency room and hospital beds as well as ICU beds, dedicated staff and hospital triage and process protocols,

### Table 4. COVID-19 Prevalence Summary

| Sample                  | First Blood Draw<sup>a</sup> | 8-Week Follow-up<sup>b</sup> | P    |
|-------------------------|-----------------------------|-------------------------------|------|
| HCW                     | Observed (95% CI)           | 0.93 (0.63–1.30)              | 2.58 (2.02–3.24) | <.001 |
|                        | Adjusted (95% CI)            | 0.98 (0.68–1.37)              | 2.76 (2.18–3.44) | <.001 |
| First responders        | Observed (95% CI)            | 0.93 (0.63–1.30)              | 2.72 (2.14–3.40) | <.001 |
|                        | Adjusted (95% CI)            | 5.31 (2.77–9.09)              | 4.35 (1.20–10.76) | .486 |
|                        | Extrapolated (95% CI)        | 5.75 (3.10–9.64)              | 4.35 (1.20–10.76) | .423 |
|                        | Extrapolated (95% CI)        | 5.31 (2.77–9.09)              | 14.56 (8.39–22.88) | .006 |
| Community               | Observed                     | 3.64                          | 22.47 | <.001 |

Note:
- Exact binomial 95% CI was calculated. Group difference testing was performed with the Fisher exact test.
- Abbreviation: COVID-19, coronavirus disease 2019.
- <sup>a</sup>95.9% were drawn in May/June 2020.
- <sup>b</sup>100% were drawn in July/August 2020.
- <sup>c</sup>Estimated from antibody tests orders by community physicians.
environmental cleansing and dietary rigor, and strict visitation policies), all to amplify patients’ and workforce safety and infection prevention measures. Mandatory employee education and training on safety measures and prevention were implemented, heightening awareness among employees not only at work but also, and more importantly, outside of their work place. All those efforts likely contributed to this lower prevalence.

A relatively low regional estimated overall prevalence of infections in Orange County (total population of 3.2 million) also likely contributed to this low prevalence, although this is only the case for the earlier period of our study (May and June 2020). This geographical effect can be seen in high antibody prevalence in HCWs in New York City, Wuhan, China, and Bergamo, Italy, where much higher community prevalence was reported. When our data were compared, using the economic re-opening in our county as a cutoff, between May/June vs July/August, the lower observed prevalence for both our HCWs (0.93% vs 2.58%) and those tested by physician orders (3.6% vs 22.5%) was reinforced. Incidentally, this trend was not observed for first responders, possibly due to a smaller sample size and a large percentage of nonreturning subjects at 8-week follow-up (although our extrapolated prevalence calculation did show this trend—5.31% vs 14.56%). Therefore, regional consideration must be given when considering antibody prevalence in HCWs.

Given the recent report of considerably higher antibody prevalence in our county (11.5%) [11], our findings of significantly lower prevalence in our HCWs may not be explained entirely by a community effect or workforce education. Other possible explanations for lower susceptibility to infection among our HCWs include biological heterogeneity and the preexisting presence of innate immunity [27] in HCWs acquired through T-cell-mediated [28] cross-reactivity to more common coronavirus species [29–31]. This hypothesis postulates that greater exposure to such predecessors is experienced more commonly in hospital settings than in the community at large. Recent studies document up to a 35% presence of such innate immunity in noninfected family members of those with confirmed infection and in donor blood sampled before the epidemic [6, 31]. Such innate immunity may also help explain the relatively low rate of infection susceptibility in younger children, given the common exposure to everyday viral infections in preschool and grade school [33–35].

Among HCWs with self-reported PCR-confirmed COVID-19, 46.7% were antibody negative (Table 5), which cannot be fully explained by antibody test sensitivity or specificity itself. Recent studies found a rapid decay of IgG antibodies within the possible span of 2–3 months in patients with milder COVID-19 symptoms [27]. This may support our findings of the negative cases with significantly fewer symptoms compared with the positives. It should be noted that the loss of antibody positivity is not equivalent to loss of immunity [27].

**CONCLUSIONS**

Our findings suggest that strict preparedness in the health care setting, rigorous processes for safety, triage, and the availability of personal protective equipment are effective in reducing the
risk to HCWs and raising confidence in those who need hospital care for urgent conditions to not delay seeking it. Also, this significantly lower prevalence in our HCWs, compared with that estimated for our community [11], warrants further studies on other contributing factors. Finally, the fact that all of our seropositive HCWs have maintained antibody positivity for at least 8 weeks, with no reported re-infection, is encouraging, given the earlier reports of antibody evanescence [26, 36].

Acknowledgments

We acknowledge our health care workers and first responders who participated in this study.

Financial support.

This study was supported by the Orange County Healthcare Agency and Hoag Hospital Foundation.

Disclaimer.

The funders did not have any role in the study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Potential conflicts of interest.

M.Z. and C.C. are employees of the Orange County Healthcare Agency, which provided the funding for this study, and have no financial or other conflicts of interest related to this study. All authors and J.R.B. and J.H. are employees of Medical Care Corporation and have no financial or other conflicts of interest related to this study. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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