Changes Over Time in COVID-19 Incidence, Vaccinations, Serum Spike IgG, and Viral Neutralizing Potential Among Individuals From a North American Gaming Venue

December 2020–August 2021

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Objective: This study aims to evaluate COVID-19 cases and vaccine responses among workers in the gaming/entertainment industry. **Methods:** Participants provided detailed information on occupational risk factors, demographics, COVID-19 history, and vaccination status through questionnaire. Enzyme-linked immunosorbent assays were used to measure serum antiviral antibodies and neutralizing capacity. **Results:** Five hundred-fifty individuals participated with $n = 228$ (41.5%) returning for follow-up. At least 71% of participants were fully vaccinated within 8 months of vaccine availability and COVID-19 rates declined concomitantly. Serum anti–spike IgG levels and neutralizing capacity were significantly ($P < 0.001$) associated COVID-19 history and vaccine type, but not occupational risk factors, and declined (on average 36%) within 5 months. Few vaccine nonresponders ($n = 12$) and "breakthrough" infections ($n = 1$) were noted. **Conclusions:** COVID-19 vaccination was associated with a marked decrease in infections; however, individual humoral responses varied and declined significantly over time.

Keywords: COVID-19, vaccine, Moderna, Pfizer-BioNTech, Johnson and Johnson, mRNA, gaming, entertainment, casino, worker, employee, face, mask, SARS-CoV-2, spike, antigen, neutralization, surrogate, virus, ACE2r, occupational, environmental, immunize, receptor binding domain, RBD, adenovirus, IgG, antibody, infection, gambling, medicine, exposure, serum, job, race, ethnicity, nonresponder, breakthrough, infection

Vaccines against COVID-19 have proven effective in preventing severe disease, hospitalization, and death.[1,2] However, differences in human immune response to different COVID-19 vaccines using different technologies (eg, nanoparticle mRNA, adenovirus) may influence their capacity to prevent infection and severe disease.[3,4] Three vaccines approved or granted emergency use authorization by the US Food and Drug Administration are manufactured by Pfizer-BioNTech, Moderna, and Johnson & Johnson (J&J).[5,6]

The effectiveness of different COVID-19 vaccines in use in the United States has now been well established.[1,2,7] Pfizer-BioNTech, Moderna, and J&J vaccines all protect against severe disease; however, differences in their stimulation of the human immune system and degree of protection have been noted.[3,4,8] Several studies suggest that J&J's vaccine may be less efficacious than mRNA vaccines, potentially related to lower levels of neutralizing antibodies, whereas Moderna's vaccine may stimulate slightly higher levels of protective antibodies than Pfizer-BioNTech's vaccine.[3,4,7–9]

The duration of vaccine-induced protective immunity against severe COVID-19 remains uncertain and is an important consideration in workplace and public health recommendations and policy decisions. One correlate of protective immunity to COVID-19 is the humoral immune response to viral antigens.[10–12] Antibodies that recognize and bind the spike protein are an important indicator of the human immune response to infection and vaccination.[13–15] A subset of anti–spike antibodies may directly block viral entry into host cells and is thought to play an important role in protection from severe disease.[16–18] Antibodies to spike and those capable of blocking viral attachment to host receptor are thus key serological indicators of vaccine responsiveness and likely relate to the duration of immunity.

The present investigation assesses COVID-19 infections and vaccine responses among individuals from a North American gaming and entertainment venue, which faced unique occupational challenges due to SARS-CoV-2. The workers underwent regular COVID-19 screening through their worksite health department with PCR testing as indicated throughout the study period. The investigation spans the 8-month period immediately before and after the availability of COVID-19 vaccines locally (December 2020–August 2021) and documents individual differences in SARS-CoV-2 directed antibody responses and decreases in serum anti–spike IgG over time.

METHODS

Human Subjects

Participants ($n = 550$) were employed volunteers, aged 18 years or older, from a gaming and entertainment venue in North America, and approximately 20% of the available workforce. Invitations to participate were distributed through established workplace communication pathways, including newsletters and onsite signage. All employees were eligible to participate. Upon consenting to participate during the initial visit scheduled in either March, April, or May 2021, participants provided information on occupational risk factors, demographics, COVID-19 history, and vaccination via a secure on-line baseline survey, which captured vaccination date(s), date(s) of PCR positive COVID-19 tests, and for participants who were not vaccinated, whether or not each planned to receive a COVID-19 vaccination. Survey completion by each volunteer was required before blood draw. Three milliliter of blood was obtained by venipuncture using vacutainer tubes (Becton Dickinson, Franklin Lake, NJ); serum was separated and stored frozen at −80°C until tested by enzyme linked immunosorbent assay (ELISA). Invitations to participate in a follow-up visit scheduled in August 2021 were communicated through established channels previously noted and by email to enrolled participants. At follow-up, participants were required to complete a secure on-line follow-up survey to gather updated information on vaccination and COVID-19 diagnoses, including dates for each dose and dates for each

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Ethical Considerations and Disclosure(s): The studies were reviewed, and ethical approval was given by the Yale University Institutional Review Board, protocol number 2000029735. All participants provided informed verbal consent.

Funding Sources: Centers for Disease Control and Prevention Contract (AWD0005821) and the National Institute for Occupational Safety and Health (5T03OH008607).

Conflict of Interest: None Declared.

Supplemental digital contents are available for this article. Direct URL citation appears in the printed text and is provided in the HTML and PDF versions of this article on the journal's Web site (www.joem.org).

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DOI: 10.1097/JOM.0000000000002617

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positive COVID-19 test. New participants at the August 2021 time point completed a modified questionnaire that captured information on both the baseline and follow-up surveys upon enrollment. Participants with positive survey responses for COVID-19 and/or those who exhibited poor antibody response to vaccination were contacted by phone to confirm test results and answer questions about potential health implications. The studies were reviewed, and ethical approval was given by the Yale University Institutional Review Board, under protocol number 2000029735. All participants provided informed verbal consent.

Serum Spike IgG

ELISAs were performed as previously described[19] using 96-well MaxiSorp plates (ThermoFisher, Waltham, MA) coated with 50 μL/well of recombinant SARS-CoV-2 ectodomain (Sino Biologicals, Wayne, PA) at a concentration of 1 μg/mL in $NaCO_3$ buffer pH 9.6 overnight at 4°C. Antigen-coated plates were blocked for 1 hour at room temperature with 200 μL of phosphate-buffered saline (PBS) containing 3% milk powder. One hundred microliters of serum diluted 1:100 in dilution solution (PBS with 0.05% Tween20, 1% milk powder) was added to blocked plates for 1 hour at RT. Plates were washed 3 times with PBS-T (PBS with 0.1% Tween-20), and 50 μL of HRP anti-human IgG antibody (Pharmingen/BD Biosiences, San Jose, CA) was added at 1:2000-fold dilution. After 1 hour of incubation at RT, plates were washed 3 times with PBS-T. Plates were developed with 100 μL of TMB Substrate Reagent Set (BD Biosciences, San Jose, CA), and the reaction was stopped when an internal pooled serum positive control sample reaches an optical densitty of 1.0 at 650 nm, by the addition of 2 N sulfuric acid. Plates were then read at a wavelength of 450 nm with reference wavelength calibration (650 nm).

Serum Viral Neutralizing Capacity

A surrogate assay was used that measures the ability of serum to inhibit viral receptor binding domain (RBD) attachment to host angiotensin converting enzyme 2 receptor (ACE2r). As previously described,[20] 96-well Nunc Maxisorp flat-bottom plates were coated with 50 μL/well of murine gamma immunoglobulin constant region (mFc) dimerized SARS-CoV-2 (2019-nCoV) Spike Protein RBD (mFc-RBD) from Sino Biological (Chesterbrook, PA), catalog number 40592-V05H, at 2 μg/mL in 1 × PBS at 4°C overnight. Plates were blocked with 200 μL of 3% nonfat milk PBS for 60 minutes at room temperature and then preincubated with positive control, negative control, and experimental serum samples titrated 2-fold from 1:10 to 1:1600 in sample dilution buffer (0.05% Tween 20 in 1% milk-PBS). Subsequently, 50 μL of a human ACE2r labeled with biotin was added (40 ng/mL based on linear range in prior titration studies) and incubated at room temperature for 60 minutes on a shaker. After 5 washes with 0.05% Tween 20-PBS, 75 μL of streptavidin-HRP from Thermo Fisher (Waltham, MA), catalog number N100, was added to each well and incubated for 60 minutes at room temperature. After 5 final washes, 100 μL of TMB substrate reagent from BD Biosciences (Franklin Lakes, NJ), catalog number 555214, was added to each well for ~15 minutes at room temperature, and reactions were terminated by addition of 50 μL of 2 N HCl to each well. Plates were read at 450/655 nm. Inhibition of RBD-ACE2r binding was calculated according to the formula 100 × (1 − [OD of sample containing 40 ng/mL ACE2r + serum/OD of sample containing 40 ng/mL ACE2r + no serum]), using values obtained from control wells on each plate. For every individual serum sample, the % inhibition observed at different dilutions was plotted versus the log (serum). The IC_{50} was defined as the serum concentration that reduced ACE2r binding 50% relative to the input ACE2r concentration (40 ng/mL) and was calculated through nonlinear regression with a 3-parameter fit curve using GraphPad Prism v9.1.2 (GraphPad Software, San Diego, CA).

Statistics and software All graphs were generated, and statistical analysis performed using Graph Pad Prism v9.1.2 or SAS v9.4. Significance differences in competitive ELISA data were calculated using the nonparametric Kruskal-Wallis tests and by analysis of variance after log transformation of log-normal distributed IC_{50} values, with Tukey correction for multiple comparisons. IC_{50} values below the LOD limit were assigned a value of 1 for statistical comparisons requiring log transformation. SAS v9.4 was used for descriptive statistics (means and standard deviations for continuous variables and percentages for categorical variables) of both predictor and outcome variables for the entire study population, as well as linear and logistic regression models.

RESULTS

Participants and Workplace

The population evaluated consisted of n = 550 volunteers comprising a distinct group of employees, managers, and owners of a North American gaming and entertainment venue (Table 1). The population was 60% female, with an average age of 50.3 years (range, 19–84 years), and 22.9% older than 60 years. Nearly all (96%) resided locally within in the state where the worksite was located. The population was racially diverse with overrepresentation of Asian and Native Americans. Follow-up data were available for 228 participants (41.5%).

All participants were actively working during the study period and were also employed in the 9 months preceding the investigation (Table 2). The majority reported being a casino worker (56%), gaming and entertainment worker (18%), or part of the workplace management (15%). The n = 306 casino workers listed 16 different job categories with marketing (19%), table games (16%), and food/beverage/culinary (13%) representing roughly half (see Supplemental Digital

TABLE 1. Employed Enrolled Participants

	n (%)
Total	550 (100)
Sex	
Male	220 (40.0)
Female	330 (60.0)
Race	
Native American	57 (10.4)
White	369 (67.1)
Asian	69 (12.5)
Black	22 (4.0)
Other	33 (6.0)
Ethnicity*	
Hispanic	37 (6.7)
Non-Hispanic	495 (78.4)
COVID+	
Yes	108 (19.6)
No	442 (80.4)
Age	
18–29	27 (4.9)
30–39	91 (16.5)
40–49	113 (20.5)
50–59	193 (35.1)
60–69	109 (19.8)
70+	17 (3.1)
Vaccine	
Yes	393 (71.5)
No	157 (28.5)
Vaccine type	
Pfizer	201 (36.6)
Moderna	168 (30.5)
J&J	24 (4.4)

*n = 82 (14.9) did not provide ethnicity.

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TABLE 2. Employment Characteristics

	n (%)
Employment Status	
Employed full time	504 (91.6)
Employed part time	46 (8.4)
Employed participants	
Casino worker	306 (55.6)
Management	81 (14.7)
Gaming and entertainment worker	97 (17.6)
Other	66 (12.0)
Works close proximity others	
Usually/always	254 (46.2)
About half the time	145 (26.4)
Seldom/never	151 (27.5)
Work facemask use	
Usually/always	471 (85.6)
About half the time	45 (8.2)
Seldom/never	34 (6.2)
Non-work facemask use	
Usually/always	440 (80.0)
About half the time	36 (6.5)
Seldom/never	74 (13.5)
Transportation to work	
Personal vehicle	531 (96.5)
Car pool	3 (0.5)
Public transit/rideshare	4 (0.7)
Walk	3 (0.6)
Other	9 (1.6)

Content S1 Table, http://links.lww.com/JOM/B126). The $n = 81$ participants involved in workplace management could be classified into 6 different job duties each with 10 to 16 participants/subgroup (Supplemental Digital Content S2 Table, http://links.lww.com/JOM/B127). The $n = 71$ gaming and entertainment workers held a wide variety of positions that stratified different job duties and locations.

More than 85% of participants reported "usually or always" wearing a mask in the workplace; however, the actual number was likely higher during earlier phases of the study, when local community infections were higher and indoor mask mandates were in place. Almost 70% of those employed reported working "in close proximity" to another employee half the time or greater during their work shift. Only $n = 7$ individuals used public transportation or carpooled to work.

SARS-CoV-2 Infection Rates and Changes Over Time

The number of new SARS-CoV-2 infections per month among the enrolled participants of the present investigation and the local state population are shown in Figure 1.[21] Infection rates among both groups began rising in the "second wave" in October of 2020 and reached peak levels in December, before vaccines became available. COVID-19 cases among the participants enrolled in the present investigation declined during January and February 2021, concomitant with reduction in local state case numbers.[21] Substantial reductions in COVID-19 occurred across the workforce and were not significantly related to occupational risk factors (job title, mask use, or social distance at work). COVID-19 cases continued to decline steadily among the enrolled participants through the end of the study (August 2021) despite ongoing spikes in new infections in the local state population in March 2021 and again in August 2021 as beta (B1.351) and delta (B1.617.2) variants became dominate.[8,9,21] The last reported COVID-19 case among the enrolled participants occurred June 16, 2021, the single vaccine "breakthrough" infection identified at the follow-up time point.

COVID-19 Vaccination

COVID-19 vaccination among the enrolled participants began at the end of December 2020, and at least 71% of the study population was fully vaccinated by August 2021 compared with <65% of the eligible local state adult population during the same period.[21] Of the unvaccinated participants, the bulk ($n = 136$, 86.6%) were evaluated only once in March or early April 2021, and 37.6% reported that they

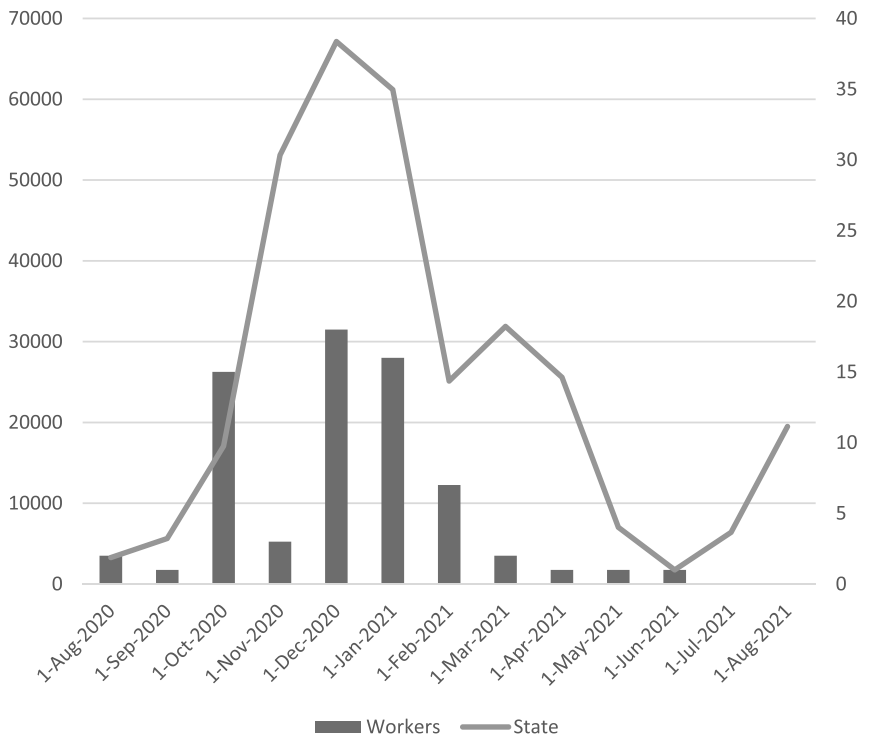

FIGURE 1. Changes in COVID-19 incidence over time. The number of COVID-19 cases among the enrolled participants (bars, right *y* axis) and the local state population (line, left *y* axis) are shown per month for the period covering August 2020 through 2021.

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"definitely," and an additional 10.2% reported they "probably," were going to get vaccinated. Of note, unvaccinated participants who reported prior COVID-19 were 2.5-fold less likely to get vaccinated than those without prior SARS-CoV-2 infection (odds ratio, 0.39; 95% confidence interval, 0.25–0.60).

Antibody Responses to Vaccination

ELISA was used to measure serum levels of IgG against the SARS-CoV-2 spike protein, the basis for COVID-19 vaccines and major antigen in natural infection. As expected, vaccination resulted in marked increases in spike IgG in almost all (96.9%) subjects. Individuals with prior COVID-19 responded to vaccination with significantly higher spike IgG levels than those without prior SARS-CoV-2 infection, whereas vaccinated individuals without prior COVID-19 had significantly higher spike IgG than those with prior infection but no vaccination (Fig. 2). Among vaccinated individuals, spike IgG levels were higher in those that received Moderna > Pfizer > J&J vaccine (Fig. 3). Among those with natural SARS-CoV-2 infection before vaccination, the level of spike IgG was significantly higher in those who reported symptoms versus those who reported asymptomatic infection (Supplemental Digital Content S1 Fig, http://links.lww.com/JOM/B128).

Subjects That Did Not Mount Detectable Antibody Responses to Vaccination

A minor subset of all vaccinated individuals (n = 12, 3.1%) failed to develop a detectable IgG response to spike antigen (Supplemental Digital Content, S3 Table, http://links.lww.com/JOM/B129). More than half of those (n = 7, 58%) also reported 1 or more COVID-19 comorbidity risk factors, including 3 with autoimmunity, 3 with hypertension, 2 with asthma, hypertension, cancer, or obesity, and 1 with heart disease (not mutually exclusive). Notably, n = 8 (67%) of the subjects that did not develop spike IgG received J&J vaccine (vs 6.4% of all vaccinated), including 4 of the 5 subjects without comorbidities. Of the total 12 subjects without detectable antibody response to vaccination, 11 (91.7%) were age 50 or older. Although only 3.1% had no detectable IgG response to vaccination, these individuals represent an important subgroup of employees at potentially higher risk of more severe COVID-19 infection despite vaccination.

Duration of Serum Levels of Spike IgG Post Vaccination

To begin assessing duration of vaccine-induced humoral responses, we evaluated the association of spike serum IgG levels with time after vaccination. As shown Figure 4, mean serum spike IgG levels were significantly lower over time (post peak response) for Moderna, Pfizer BioNTech, and J&J recipients, with sustained differences in magnitude.

The duration of vaccine-induced responses was evaluated more directly by comparing spike IgG levels in individual participants' paired serum collected at 2 different time points >3 months apart, with the second time point >5 months after full vaccination. Figure 5 shows data for the subset of vaccinated subject that met the criteria (n = 63). On average, individual spike IgG serum levels decreased 36% between the 2 different time points (Fig. 5).

Serum Viral Neutralizing Capacity

We further characterized serum SARS-CoV-2 neutralizing capacity based on its ability to inhibit RBD engagement of ACE2r in a competitive ELISA (eg, surrogate neutralization). Analysis was performed on a subset of samples obtained during the period of peak vaccine response (7–14 days post second dose of mRNA, or 14–30 days

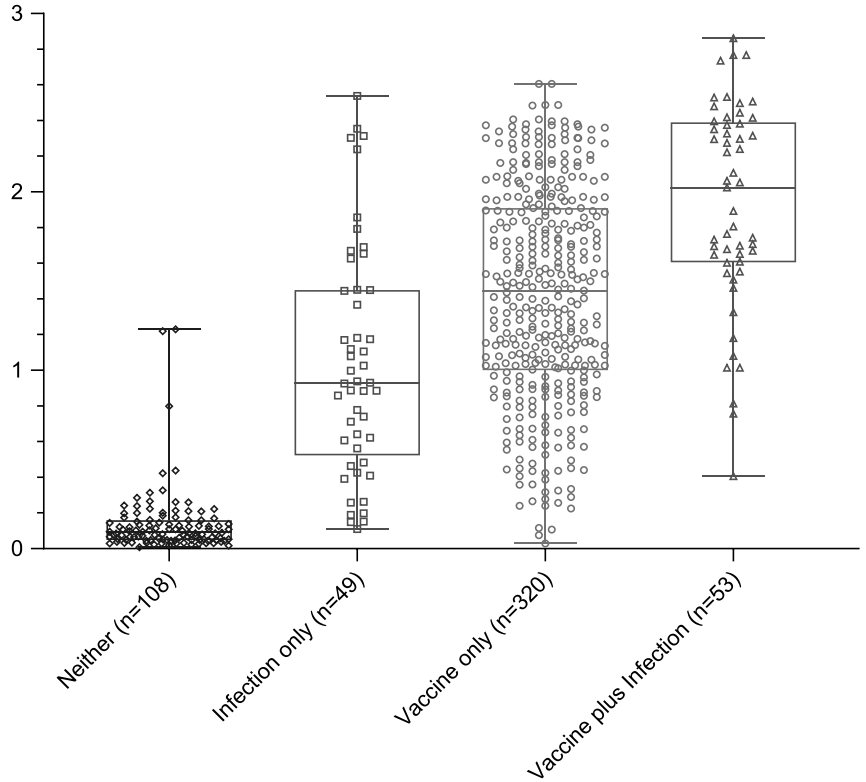

FIGURE 2. Serum spike IgG levels depend upon vaccination and prior COVID-19 history. Box plot shows the median, interquartile range, and min/max ELISA OD values (y axis) for unvaccinated individuals without prior COVID-19 (n = 108, neither) unvaccinated individuals with prior COVID-19 infection (n = 49, infection only), fully vaccinated individuals without prior COVID 19 (n = 320, vaccine only), and fully vaccinated individuals with prior COVID-19 (n = 53, vaccine plus infection). Each symbol represents an individual participant.

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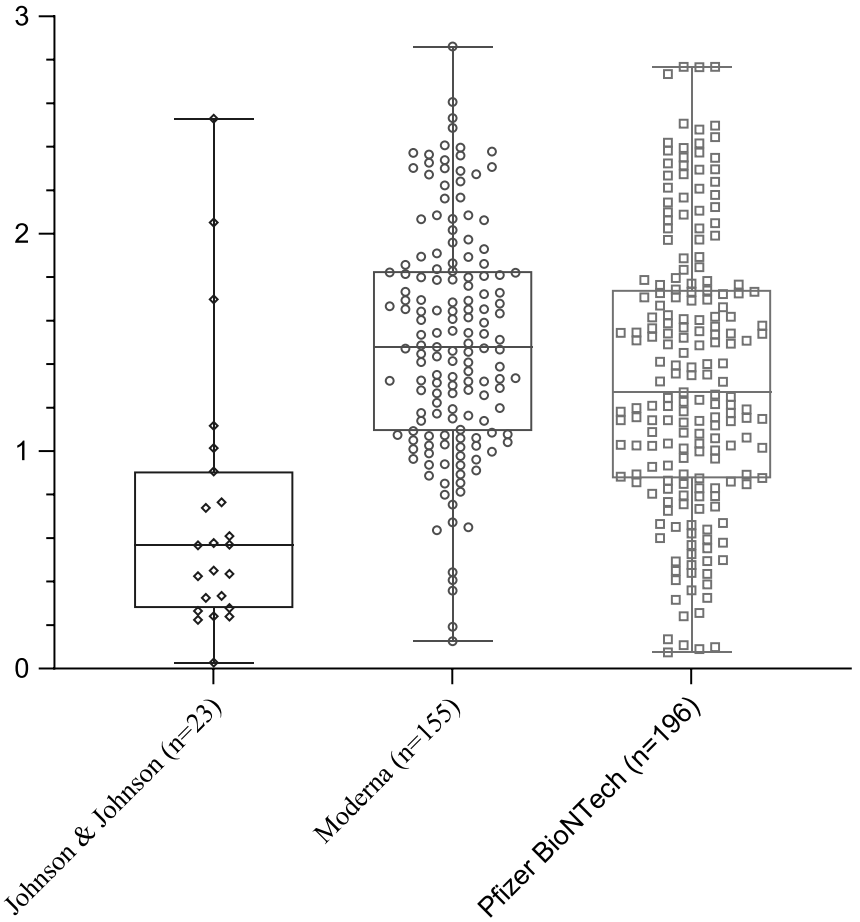

FIGURE 3. Spike IgG response to vaccination depending upon type of COVID-19 vaccine received. Box plot shows the median, interquartile range, and min/max ELISA OD values (*y* axis) for individuals that received Johnson & Johnson (*n* = 23), Pfizer-BioNTech (*n* = 196), or Moderna (*n* = 155) COVID-19 vaccines. All individuals were >7 days second dose of mRNA vaccine or 14 days post J&J vaccine. Each symbol represents an individual participant.

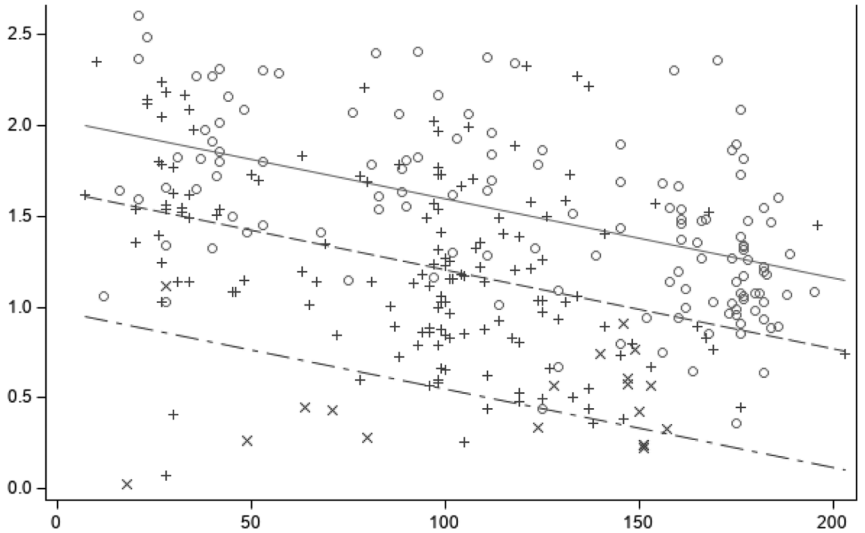

FIGURE 4. Association of spike IgG with time post vaccination. The amount of serum spike IgG measured by ELISA (OD values on the *y* axis) is shown for *n* = 301 individual samples (each symbol represents a different participant, o = Moderna, + = Pfizer, x = J&J) obtained at varying days (*x* axis) after the point of maximal vaccine responses (7–14 days after second mRNA vaccine dose or 14–30 days after Johnson & Johnson vaccine). Trend lines are shown for individuals receiving different COVID-19 vaccines (solid line = Moderna, dashed line = Pfizer, dash/dot line = J&J). Individuals with autoimmune conditions were excluded from analysis.

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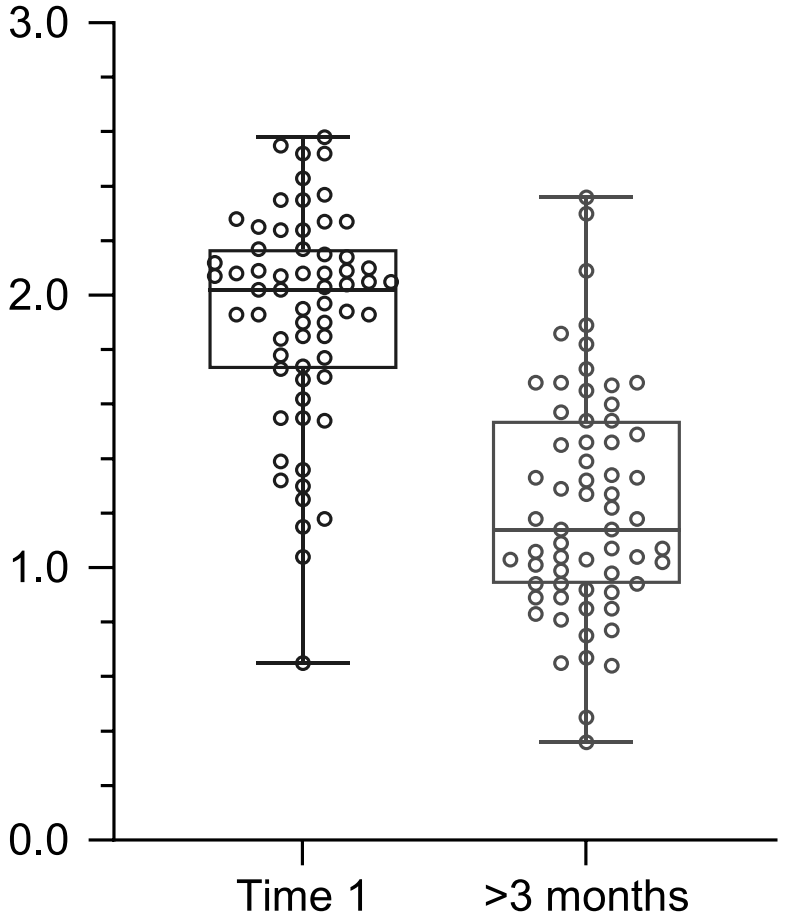

FIGURE 5. Decrease in vaccine-induced spike IgG over time. Serum spike IgG measured by ELISA (OD on the *y* axis) from paired serum samples obtained at baseline visit in 2021 and at follow-up visit greater than 3 months later (>5 months post vaccination). Each symbol ($n = 63$ at each time point) represents an individual participant. Box plot shows the median, interquartile range, and min/max ELISA OD values; $P < 0.001$ between time points. Individuals with autoimmune conditions were excluded from analysis.

post single dose J&J vaccine).[7,8,19,20] Significant difference in surrogate neutralization were observed depending upon prior history of COVID-19 and type of vaccine received (Fig. 6). The strongest neutralizing capacity (IC_{50} values) was observed among those with prior COVID-19 infection and vaccination > vaccination only > infection only. Significant differences in neutralizing capacity were associated with the type of vaccine received, with highest IC_{50} values for Moderna, followed by Pfizer, and lowest for J&J, consistent with recent reports.[3,4] In general, there was a good correlation between spike IgG levels and neutralizing capacity; however, not all individuals with high spike IgG demonstrated high titer neutralizing capacity (Supplemental Digital Content S2 Fig, http://links.lww.com/JOM/B130).

Rapid Decrease in Surrogate Neutralizing Activity

Surrogate neutralizing activity was again measured in the subset of available follow-up samples obtained >3 months after vaccination. As shown in Figure 7, neutralizing activity was significantly lower in all follow-up samples tested and below detection limits in roughly one third of individuals. Serum neutralizing potential was sustained at a relatively higher levels among vaccinated individuals with prior COVID-19 (vs those without prior COVID-19) consistent with higher levels during peak vaccine responses as shown in Figure 6 previously. Notably, only 1/11 J&J vaccine recipients that developed neutralizing antibodies during the peak vaccine response period had detectable neutralizing capacity when retested >3 months later (data not shown).

Negative Association of Vaccine-Induced Serum Spike IgG and Age

A significant ($P < 0.05$) negative correlation was observed between spike IgG and age among those fully vaccinated and no self-reported medical conditions ($n = 114$). Predicted spike OD value by vaccine and age at 14 day and day 180 post full dose are shown in Figure 8, modeled with time post vaccination as an interacting factor. Albeit limited in magnitude, the negative correlation of spike IgG with age was observed regardless of which type of COVID-19 vaccine was received.

DISCUSSION

This investigation characterizes SARS-CoV-2 infection and vaccination in a unique workforce population with potential for exposure during indoor face-to-face contact with customers and coworkers. We found a high vaccination rate concomitant with a significant reduction in new COVID-19 cases, which exceeded that of the local population. Only 1 "breakthrough" infection was reported among nearly 400 fully vaccinated individuals (0.06%/month) during the last 2 months of the investigation (ending August 2021). Although COVID-19 vaccination was efficacious, significant differences in humoral responses (serum spike IgG and neutralizing capacity) were observed among individuals depending upon prior SARS-CoV-2 infection and type of vaccine received. The clinical significance of

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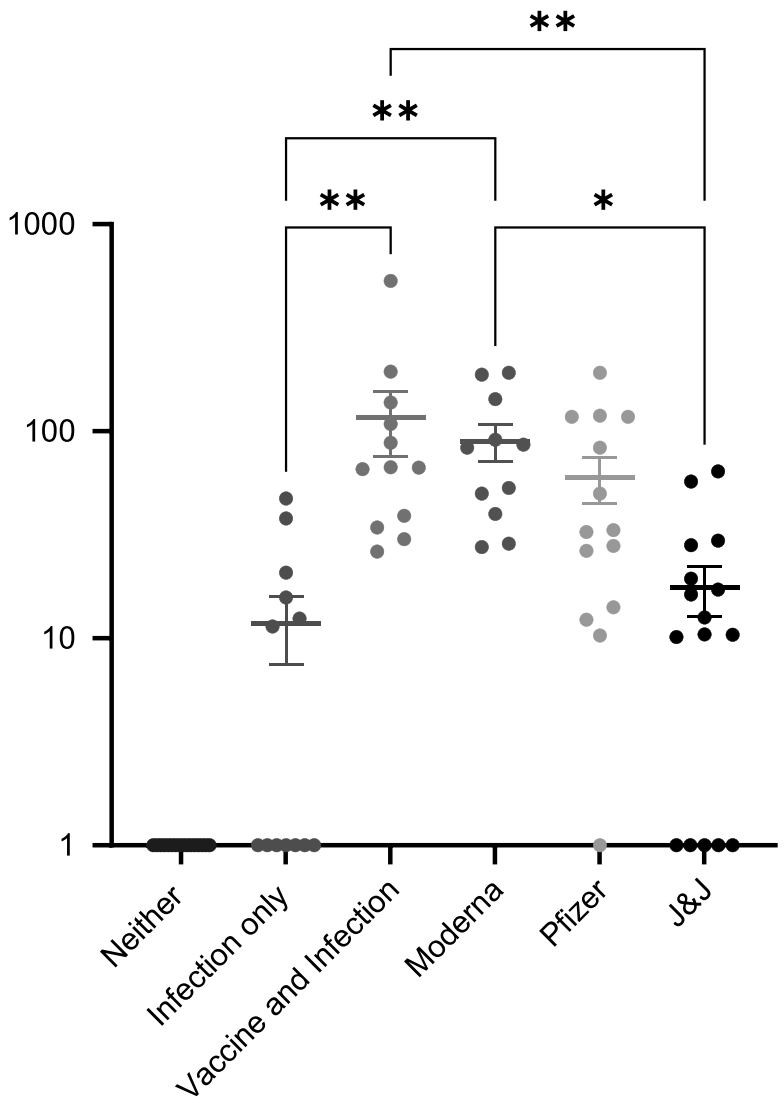

FIGURE 6. Differences in serum neutralizing capacity depending upon type of vaccine and prior COVID-19 history. The IC_{50} values (*y* axis) are shown for $n = 16$ individuals with neither vaccine nor infection (neither), $n = 13$ COVID-19 infected only (infection only), $n = 12$ vaccinated individuals with prior COVID-19 (vaccine and infection), and individuals without prior COVID-19 vaccinated with Moderna ($n = 11$), Pfizer ($n = 14$), or J&J ($n = 16$) as labeled. Each symbol represents a different participant; the mean and 95% confidence interval values are depicted; *$P < 0.05$, **$P < 0.005$ between groups as labeled. Individuals with autoimmune conditions were excluded from analysis.

differential humoral responses to COVID-19 vaccination remains to be determined but is consistent with recent reports on differential vaccine efficacy.[3,4] Significant decreases in serum anti–spike IgG levels and neutralizing capacity occurred >5 months post full vaccination, consistent with current recommendations for, and timing of, booster shots.

The present data found no significant associations of COVID-19 with known risk factors related to occupational duties in the gaming and entertainment industry. The data suggest that stringent occupational hygiene (including mask use and social distancing) has been effective in preventing occupation-related SARS-CoV-2 infection in the present workplace. In contrast, the increase and decrease in case numbers largely paralleled that of the local population, suggesting environmental (including vaccine availability) rather than occupational risk factors as the major driver of infection, but cannot rule out the possibility of similarly high exposures across different jobs. The sustained decrease in COVID-19 cases among the working participants, despite spikes in the local state population, may be attributable to higher vaccination rates among those enrolled in the present investigation combined with regular mask use and social distancing.

Although 96.9% of enrolled participants responded to COVID-19 vaccination with measurable anti–SARS-CoV-2 spike IgG, 3.1% or $n = 12$ employees failed to mount detectable responses. The majority of vaccine "nonresponders" reported 1 or more COVID-19 "comorbidity factors," of which autoimmunity was most prevalent. Johnson & Johnson vaccine recipients accounted for 67% of "vaccine nonresponders," despite representing <7% of all vaccine recipients consistent with recent reports of limited humoral responses to J&J versus other COVID-19 vaccines.[4] The lack of detectable anti–spike antibodies in these employees is concerning given that vaccine-induced antibodies generally reflect "helper" T cell responses with capacity for long-lived "memory."[22]

The differences in humoral response to different vaccines among the employees of the present study are similar to those recently

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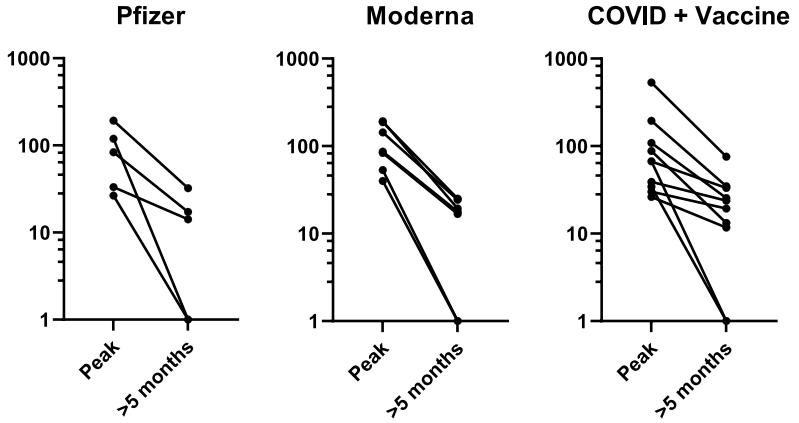

FIGURE 7. Decrease in serum neutralizing capacity over time. Serum neutralizing activity (IC_{50} *y* axis) from paired serum samples obtained at time of peak vaccine response and at a follow-up visit greater than 3 months later. Each symbol/line represents an individual participant vaccinated with Pfizer or Moderna vaccines (and no prior COVID-19) or vaccinated individuals with prior COVID-19 as labeled. Individuals with autoimmune conditions were excluded from analysis.

described in a recent Centers for Disease Control and Prevention report comparing the effectiveness of Moderna, Pfizer-BioNTech, and J&J vaccines in preventing COVID-19 hospitalizations among adults without immunocompromising conditions.[4] The data are also consistent with COVID-19 vaccine "mixing and matching" studies, which describe similar differences upon subsequent booster doses.[23] Given that antibodies are recognized correlates of protective immunity, these findings along with information on side/adverse effects provide important data in making personal health choices, as well as and public and workplace health policy decisions.

Potent vaccine responses among individuals with prior COVID-19 (significantly higher on average than vaccinated SARS-CoV-2–naive individuals) have been a consistent finding in our laboratory and published reports.[24,25] Such findings follow classical immunologic dogma that "recalls" responses are faster and amplified versus primary responses.[26–28] Together, the data highlight the potential that exists for

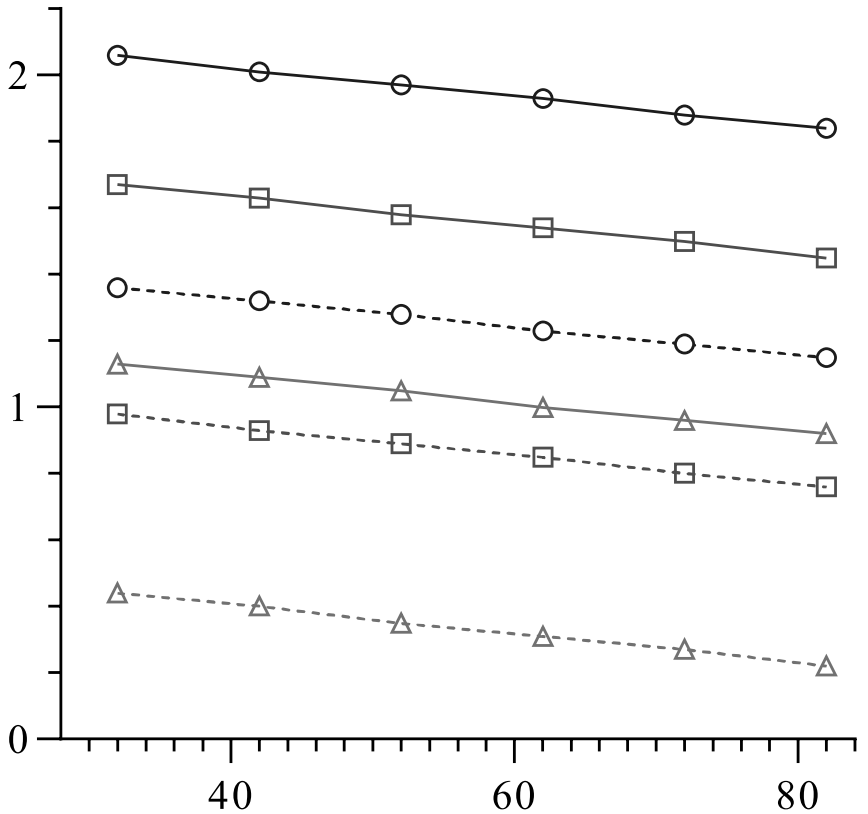

FIGURE 8. Association of vaccine-induced spike IgG levels with age. Serum spike IgG was measured by ELISA (OD *y* axis) for individuals across an age range of 32 to 82 years. Predicted OD values for individuals at day 14 (solid lines) and day 180 (dashed lines) across the age range receiving different vaccine types (circle = Moderna, square = Pfizer, triangle = J&J) are modeled with time post vaccination as an interacting factor.

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"boosting" adaptive SARS-CoV-2 immunity, particularly for those without prior COVID-19, as well as those with prior COVID-19.

This investigation's strengths and weaknesses should be recognized in considering the findings' significance. The strengths include the large number of participants from a gaming and entertainment workforce, which faces unique challenges due to COVID-19, and which rapidly provided employees, on a voluntary basis, with vaccination upon availability. The investigation includes a high proportion of minority workers (Asian and Native American) and a subset of employees with follow-up samples at distant time points (>5 months) post full vaccination allowing assessment of changes over time. Measurements of viral neutralizing capacity were assessed in addition to total spike levels based on RBD-ACE2r competitive inhibition as recently described.[20,29–31]

The major weakness of the investigation is its timing, before booster doses and the emergence of omicron and BA.2, and extension of the findings to these (and future) variants, and "boosted" individuals remains uncertain. Other weaknesses included the voluntary participant enrollment, which may not reflect the entire workforce, and limited sample sizes of some studies (serum neutralizing capacity). Finally, the investigation focused on working age adults, which may not be reflective of the general population, or responses in children receiving lower vaccine doses.

CONCLUSIONS

Workers in the gaming and entertainment industry faced unique occupational risks for SARS-CoV-2 infection, used extensive industrial hygiene controls, and were rapidly vaccinated (71%) within 8 months of COVID-19 vaccine availability. The number of COVID-19 cases among the enrolled participants concomitantly decreased, with no new cases identified during the last 2 months of the investigation (July and August 2021), despite continued spikes in cases among the local population. Antibody responses to vaccination differed significantly depending upon type of vaccine received, with levels of anti–spike IgG and neutralizing capacity that parallel relative differences in vaccine efficacy recently described.[3,4,8] Data demonstrating lower serum antibody levels (total spike IgG and neutralizing) in vaccinated individuals without prior COVID-19 (vs vaccinated individuals with prior disease) suggest that potential exists to further augment immunity, potentially through boosters. Data demonstrating declines over time in vaccine-induced viral neutralizing activity and anti–spike IgG further support the likely benefit of booster shots approximately 5 months after completing initial vaccination regimes (2 dose mRNA, 1 dose adenovirus), as currently recommended. As SARS-CoV-2 becomes endemic and new variants emerge, continued monitoring of viral immunity among this at-risk workforce should help evaluate the ongoing risk from infection and inform workplace policies aimed at disease intervention.

ACKNOWLEDGMENTS

We are indebted to all the individuals that participated in the investigation and the Centers for Disease Control and Prevention and National Institute for Occupational Safety and Health for providing financial support.

REFERENCES

1. Lopez Bernal J, Andrews N, Gower C, et al. Effectiveness of Covid-19 vaccines against the B.1.617.2 (Delta) variant. *N Engl J Med*. 2021;385:585–594.
2. Pilishvili T, Gierke R, Fleming-Dutra KE, et al. Effectiveness of mRNA Covid-19 vaccine among U.S. Health Care Personnel. *N Engl J Med*. 2021;385:e90.
3. Tada T, Zhou H, Samanovic MI, et al. Neutralization of SARS-CoV-2 variants by mRNA and adenoviral vector vaccine-elicited antibodies. *Front Immunol*. 2022;13:797589.
4. Self WH, Tenforde MW, Rhoads JP, et al. Comparative effectiveness of Moderna, Pfizer-BioNTech, and Janssen (Johnson & Johnson) vaccines in preventing COVID-19 hospitalizations among adults without immunocompromising conditions—United States, March–August 2021. *MMWR Morb Mortal Wkly Rep*. 2021;70:1337–1343.
5. Banerji A, Wickner PG, Saff R, et al. mRNA vaccines to prevent COVID-19 disease and reported allergic reactions: current evidence and suggested approach. *J Allergy Clin Immunol Pract*. 2021;9:1423–1437.
6. Shay DK, Gee J, Su JR, et al. Safety monitoring of the Janssen (Johnson & Johnson) COVID-19 vaccine—United States, March–April 2021. *MMWR Morb Mortal Wkly Rep*. 2021;70:680–684.
7. Sacks HS. The single-dose J&J vaccine had 67% efficacy against moderate to severe-critical COVID-19 at ≥14 d. *Ann Intern Med*. 2021;174:JC75.
8. Lin DY, Gu Y, Wheeler B, et al. Effectiveness of Covid-19 vaccines over a 9-month period in North Carolina. *N Engl J Med*. 2022;386:933–941.
9. Centers for Disease Control and Prevention. *COVID-19 Science Briefs*. Atlanta, GA: Centers for Disease Control and Prevention (US); 2020. March 4, 2022. Available at: https://www.ncbi.nlm.nih.gov/books/NBK570439/. Accessed May 25, 2022.
10. Feng S, Phillips DJ, White T, et al. Correlates of protection against symptomatic and asymptomatic SARS-CoV-2 infection. *Nat Med*. 2021;27:2032–2040.
11. Poonia B, Kottilil S. Immune correlates of COVID-19 control. *Front Immunol*. 2020;11:569611.
12. Krammer F. A correlate of protection for SARS-CoV-2 vaccines is urgently needed. *Nat Med*. 2021;27:1147–1148.
13. Wei J, Stoesser N, Matthews PC, et al. Antibody responses to SARS-CoV-2 vaccines in 45,965 adults From the general population of the United Kingdom. *Nat Microbiol*. 2021;6:1140–1149.
14. Blain H, Tuaillon E, Gamon L, et al. Spike antibody levels of nursing home residents with or without prior COVID-19 3 weeks after a single BNT162b2 vaccine dose. *JAMA*. 2021;325:1898–1899.
15. Wei J, Matthews PC, Stoesser N, et al. Anti–spike antibody response to natural SARS-CoV-2 infection in the general population. *Nat Commun*. 2021;12:6250.
16. Li D, Sempowski GD, Saunders KO, Acharya P, Haynes BF. SARS-CoV-2 neutralizing antibodies for COVID-19 prevention and treatment. *Annu Rev Med*. 2022;73:1–16.
17. Dispinseri S, Secchi M, Pirillo MF, et al. Neutralizing antibody responses to SARS-CoV-2 in symptomatic COVID-19 is persistent and critical for survival. *Nat Commun*. 2021;12:2670.
18. Min L, Sun Q. Antibodies and vaccines target RBD of SARS-CoV-2. *Front Mol Biosci*. 2021;8:671633.
19. Wisnewski AV, Campillo Luna J, Redlich CA. Human IgG and IgA responses to COVID-19 mRNA vaccines. *PLoS One*. 2021;16:e0249499.
20. Wisnewski AV, Liu J, Lucas C, et al. Development and utilization of a surrogate SARS-CoV-2 viral neutralization assay to assess mRNA vaccine responses. *PLoS One*. 2022;17:e0262657.
21. Connecticut COVID-19 Response. Available at: https://portal.ct.gov/coronavirus/covid-19-data-tracker. Accessed November 21, 2021.
22. Dorner T, Radbruch A. Antibodies and B cell memory in viral immunity. *Immunity*. 2007;27:384–392.
23. Atmar RL, Lyke KE, Deming ME, et al. Heterologous SARS-CoV-2 booster vaccinations—preliminary report. *medRxiv*. 2021:2021.2010.2010.21264827.
24. Callaway E. COVID super-immunity: one of the pandemic's great puzzles. *Nature*. 2021;598:393–394.
25. Wisnewski AV, Redlich CA, Liu J, et al. Immunogenic amino acid motifs and linear epitopes of COVID-19 mRNA vaccines. *PLoS One*. 2021;16:e0252849.
26. Palm AE, Henry C. Remembrance of things past: long-term B cell memory After infection and vaccination. *Front Immunol*. 2019;10:1787.
27. Akkaya M, Kwak K, Pierce SK. B cell memory: building two walls of protection against pathogens. *Nat Rev Immunol*. 2020;20:229–238.
28. Ibarrondo FJ, Hofmann C, Fulcher JA, et al. Primary, recall, and decay kinetics of SARS-CoV-2 vaccine antibody responses. *ACS Nano*. 2021;15:11180–11191.
29. Abe KT, Li Z, Samson R, et al. A simple protein-based surrogate neutralization assay for SARS-CoV-2. *JCI Insight*. 2020;5:e142362.
30. Tan CW, Chia WN, Qin X, et al. A SARS-CoV-2 surrogate virus neutralization test based on antibody-mediated blockage of ACE2-spike protein-protein interaction. *Nat Biotechnol*. 2020;38:1073–1078.
31. Valcourt EJ, Manguiat K, Robinson A, et al. Evaluation of a commercially-available surrogate virus neutralization test for severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2). *Diagn Microbiol Infect Dis*. 2021;99:115294.

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