Original Article

Seroprevalence of SARS-CoV-2 antibodies among healthcare workers in a teaching hospital in Eastern India

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Abstract

Statement of the Problem: Healthcare workers (HCW) are the most vulnerable group for contracting SARS-CoV-2. Assessment of seroprevalence of SARS-CoV-2 antibodies among HCW, thus can provide important data on pathogen exposure, infectivity, and adherence to personal protective equipment (PPE). The present study aimed at assessing SARS-CoV-2 seroprevalence among HCW and exploring associations with demographics, category of exposure to COVID-19 patients, preventive measures taken and relation with COVID-19 symptoms. **Method of Study:** HCWs with a minimum gap 2 weeks from last duty were eligible to participate in the study. The enrolled HCW were categorized into high-risk and low-risk category based on work in COVID-19 areas. HCWs SARS-CoV-2 specific IgG and IgM antibodies were detected using rapid immunochromatography test. **Results:** Out of 821 randomly selected HCWs, either IgM or IgG antibody was detected in 32 HCWs (32/821, 3.9%). Only IgM antibodies were detected in 14 (1.7%), only IgG was detected in 9 (1.0%), and both IgM and IgG antibodies were present in 9 HCWs. Seropositivity was significantly higher in high-risk category (5.7% vs. 2.2.%), HCWs who ever had COVID-19 related symptoms in last 3 months (5.6% vs. 2.8%), and those who had earlier tested positive for SARS-CoV-2 with real-time reverse transcriptase PCR (36.6% vs. 3.5%). Seroprevalence was highest (6.9%) among housekeeping and sanitation staff. **Conclusions:** Overall, low seroprevalence of SARS-CoV-2 antibodies in our HCWs is an indicator of effective infection control practice. HCW posted in dedicated COVID ward need more stringent implementation of infection prevention measures.

Keywords: Antibody, COVID-19, healthcare worker, SARS-CoV-2

Introduction

In December 2019, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), a novel coronavirus was discovered during the investigation of a cluster of pneumonia of unknown origin in Wuhan, China.[1] Considering its high rate of transmission across the globe, World Health Organization (WHO) declared

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coronavirus disease 2019 (COVID-19) as pandemic in March 11.[2] India reported its first case in January 27, 2020 and by early September witnessed over 40 lakhs cases and over 70, 000 deaths and is currently the second worst affected country in the world.[3]

COVID-19 has a wide range of clinical presentation from mild flu like symptoms to pneumonia, acute respiratory distress syndrome leading to death. It has also been reported that approximately four-fifth of COVID-positive cases are asymptomatic.[4-6] Healthcare workers (HCWs) being the frontline work force comprise of a vulnerable cohort for acquiring infection because of frequent and close contact to confirmed and suspected COVID-19 patients. The national real-time reverse transcriptase PCR (RT-PCR) based testing strategy of SARS-CoV-2 in India advises testing of symptomatic HCW and asymptomatic HCW who comes in high-risk contact of laboratory confirmed cases and hence is likely to miss asymptomatic and paucisymptomatic cases.[7] Infected HCW pose risk for the patients, family members, and to the community as well. Antibody-based surveillance during ongoing pandemic can provide data on pathogen exposure.[8] There is uncertainty about the proportion of COVID-19 positivity among the HCWs after exposure to COVID-19 patients, as majority remain asymptomatic or paucisymptomatic. Various countries across the world have reported wide variation in seropositivity in their HCW ranging from 1.6% to 13.7%.[9-11] In the recently published Indian studies, seropositivity in HCW has been shown to be 11% in Mumbai and 11.9% in Kolkata.[12,13]

The aim of this study was to assess SARS-CoV-2 seroprevalence among HCW and to explore associations with demographics, category of exposure to COVID-19 patients, preventive measures taken and COVID-19 symptoms.

Methodology

Study design and setting

This cross-sectional study was conducted from August 1 to 8, 2020 at All India Institute of Medical Sciences (AIIMS), Bhubaneswar which is a 960 bedded teaching hospital located in capital city Bhubaneswar in Odisha, an eastern state of India. Symptomatic screening and testing for SARS-CoV-2 through RT-PCR in the hospital was started on 16th March 2020. The first case of COVID-19 was admitted in our hospital on 19th March 2020. Since then, both COVID-19 and non-COVID-19 patients are regularly admitted to the hospital. COVID-19 patients were admitted in the dedicated blocks within the hospital. Both outpatients and inpatients were tested as per the prevalent national guidelines.[7] For HCW, all symptomatic HCWs and all asymptomatic HCWs who were high-risk contacts of COVID-positive patients were screened by RT-PCR as per ICMR guidelines.[7] Universal screening for COVID-19 of all the newly admitted patients to the hospital started in middle of June 2020. From July 8th onwards, hospital admission was restricted to only COVID-19 patients and patients requiring emergency intervention, whereas routine outpatient consultations has been discontinued since then because of sudden surge in COVID-19 cases in community and hospital. As of July 20th, 266 COVID-19 patients were admitted in this hospital and 55 HCWs tested positive for SARS-CoV-2. The study was approved by Institute ethics committee . Approval No. T/IM-NF/Micro/20/85. Dated 28 July,, 2020.

Study procedures

Ethical approval was obtained from the Institutional Ethics Committee of AIIMS Bhubaneswar. List of all categories of HCWs (i.e., doctors, nurses, other paramedical staff) and supporting staff like sanitary, housekeeping, and security personnel working in hospital was obtained from hospital administration. Sample size of 810 was calculated assuming the seroprevalence of 5%, allowable error of 1.5% at a significance level of 95%. Considering a non-response rate of 20%, we randomly selected 1,000 HCWs without any sampling weights and invited them to participate in the study. Since HCWs were randomly selected from the list of all staff, it also included HCWs who had already tested positive for SARS-CoV-2 using RT-PCR method. Apart from them who were randomly selected, we also invited all other HCWs who had earlier tested positive for COVID-19 to participate in the study as positive control. However, for estimation of seroprevalence, we used data of only those previous positive HCWs who were randomly selected, to avoid overestimation of seropositivity.

All HCWs and other supporting staff were categorized into high-risk and low-risk category based on work in COVID-19 areas. HCWs who had worked for at least 15 days in designated COVID wards or COVID Intensive Care Units (ICUs) and were directly involved in patient care were categorized as high risk. All other HCWs, whether involved in direct patient care or not, who were not high risk were categorized as low risk. Other risk factors were categorized as dichotomous variables.

HCWs were requested to report to a designated place for sample collection. Measures were taken to maintain physical distancing, compliance to wearing of mask, and hand hygiene measures. Demographic, work, exposure related information, and history of symptoms suggestive of COVID-19 in the past was elicited and a blood sample was collected by venipuncture by trained phlebotomists after obtaining written informed consent. Samples were then transferred to Microbiology laboratory where serum was separated by centrifugation.

SARS -CoV-2 specific IgG and IgM antibodies were detected using rapid immunochromatography test (Standard Q COVID-19 IgM/IgG Duo test, M/s S.D. Biosensor, Inc South Korea).[14] This test detects IgG and IgM antibodies against recombinant SARS-CoV-2 nucleocapsid protein. The sensitivity and specificity of the assay for combined IgM and IgG has been reported by the manufacturer as 99.10% and 95.09%, respectively, as compared to PCR after 14 days of symptom onset.[14] The test was carried out following the manufacturers' instruction.[14] The result was read after 15 min. and was considered valid when control line appeared. In case of non-appearance of control line sample was

retested using another kit strip. Each test was read independently by two observers. Participants were considered seropositive when either IgM or IgG or both IgM/IgG were detected.

Statistical Analysis was done using SPSS Version 20.0 (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.). Appropriate test of significance, that is, Chi-square test or Fischer's exact test was applied to determine the association of seropositivity with various factors. *P* value of 0.05 was considered significant.

Results

Out of randomly selected 1,000 HCWs, 821 participated in the study (non-response rate of 17.9%). The mean age of participants was 32 years (SD ± 7.70) and 65.2% were male. Study population comprised of all the type of HCWs and supporting staff; 32.9% were nurses, 23.1% were doctors, 22.6% were housekeeping and sanitation personnel, rest were security staff and other paramedical staff. HCWs were equally distributed in high-risk (50.8%) and low -risk (49.2%) category. COVID-19 related symptoms in last 3 months was reported by 38.9%, whereas 40.4% of HCWs were never tested for SARS-CoV-2 either for symptoms or after being a direct contact with a COVID-19 positive patient. Out of them, 11 had tested positive for SARS-CoV-2 before the sero-survey. Descriptive characteristics is presented in Table 1.

Sero-prevalence of SARS-CoV-2 antibodies was estimated to be 3.9% (95% CI, 2.6--5.1%) among randomly selected 821 HCWs. Either IgM or IgG was detected in 32 HCWs. Only IgM antibodies were detected in 14 (1.7%), only IgG was detected in 9 (1.0%) and both IgM and IgG antibodies were present in 9 HCWs [Table 2]. Sero-prevalence among HCWs with different job profile ranged from zero among doctors to 6.99% among housekeeping and sanitation staff [Table 3].

Of the 31 HCWs who were recruited as positive control *(i.e., those who had tested positive for SARS-CoV-2 through RT-PCR before sero-survey and which also included 11 HCWs who were enrolled through random selection)*, either IgM or IgG antibody was detected in 17 HCWs (54.8%). Among them, 8 HCW mounted both IgG/Ig M response, 7 HCW mounted only Ig G response, and 2 mounted only Ig M response.

Seropositivity was significantly higher in high-risk category, that is, HCWs who had worked in COVID areas for at least 15 days (5.7% vs. 2.2.%, OR = 2.74; 95% C.I. 1.25--5.99), HCWs who ever had COVID-19 related symptoms in last 3 months (5.6% vs. 2.8%, OR = 2.74; 95% C.I., 1.25--5.99) and those who had earlier tested positive for SARS-CoV-2 with RT-PCR (36.6% vs. 3.5%, OR = 15.96; 95% C.I 3.28--37.66). No significant association of seropositivity was found with hydroxy chloroquine (HCQS) prophylaxis, history of seasonal influenza and BCG vaccine [Table 4].

Table 1: Baseline characteristics of HCWs included in the study

Variable	Number	%
Sex		
Male	535	65.2
Female	286	34.8
Age (in years)		
Up to 20	7	0.8
21-30	423	51.5
31-40	289	35.2
41-50	71	8.6
51 and above	31	3.8
Mean age (±S.D.)	32.00±7.70	
Job Profile		
Nurses	270	32.9
Resident doctors	121	14.7
Faculty	69	8.4
Other Paramedical Staff	88	10.8
Housekeeping and Sanitation staff	186	22.6
Security	87	10.6
Risk Category during work (i.e worked in COVID areas for at least 15 days)		
Low	417	50.8
High	404	49.2
Direct contact with any COVID-19 patient at home or workplace	531	64.7
Had any COVID-19 related symptoms in last three months	319	38.9
Ever tested by RT PCR for SARS-COV-2 infection	332	40.4
Tested positive on RT PCR for SARS-CoV-2 before sero survey	11	1.33
Locality declared as containment zone during COVID-19 pandemic	172	21.0
Ever taken HCQS prophylaxis during COVID-19 pandemic	97	11.8
Ever taken any vaccine for seasonal influenza	55	6.7
History of BCG vaccination during childhood	570	69.4

Table 2: Seroprevalence among Health Care Workers (HCWs)

Outcome	Number (*n*=821)	Percentage (95% CI)
Seropositive (Either IgM or IgG)	32	3.9 (2.6-5.1)
Only IgM Positive	14	1.7
Only IgG Positive	9	1.0
Both IgM and IgG Positive	9	1.0

Table 3: Seroprevalence among Health Care Workers with respect to Job profile

Designation	Total	Participated in study	Seroprevalence (%)
Nursing Officer	830	270	10 (3.70)
Resident doctors	419	121	4 (3.31)
Faculty	205	69	0
Other paramedical staff	224	88	2 (2.25)
Housekeeping and sanitation staff	560	186	13 (6.99)
Security personnel	233	87	3 (3.45)

Discussion

The overall seropositivity of 3.9% among HCWs in the present single center study is in line with majority of the published studies across the continents which range from less than 2% to up to 13%.[9,10,15-20] Seropositivity in general population was estimated to be around 1.9% in survey done in last week of July 2020.[19] Thus, HCWs had slightly higher seropositive but this was not significant. The implementation of standard operating procedures on infection control measures, adequate supply of PPE, screening of symptomatic staff by RT-PCR along with contact tracing and quarantine might have attributed to the low seroprevalence among HCWs.

Present study found the seropositivity (Ig G and or Ig M) was 55% among HCW who were earlier positive on RT-PCR. This is relatively lower than the previous studies from Iceland, China, New York City region, where the 91--99% seropositivity was demonstrated in prior RT PCR positive individuals.[21-23] In the large-scale study from Denmark, the seropositivity was 64.17%. The possible factors of relatively lower seropositivity in different studies could be dependent upon the coated target antigen. The kit used in the present study detects IgG/IgM against the recombinant COVID-19 nucleocapsid protein, whereas in the study from Iceland multi-antigen, multi-isotype antibody surveillance was used. Though a median of 19 days' time gap is considered adequate for seroconversion, the study by Wajnberg *et al.*[23] has demonstrated 99% of the patients to have seroconverted within 50 days of RT-PCR. We had recruited participants who had at least 4 weeks gap from RT-PCR. Perhaps, this could have attributed to the relatively lower seropositivity and hence they need to be followed up.

HCWs are considered a vulnerable group for acquiring infection by virtue of their role in patient care. To find out the association of direct patient care among the HCW we stratified them in to high-risk and low-risk groups based on their level of exposure to COVID-19 patients. Our study found significantly higher seropositivity against SARS-CoV-2 in high-risk group as compared to low-risk group which was categorized based on their duties in designated COVID ward/ICU and non-COVID wards, respectively. Several studies have reported similar finding of higher seropositivity among the HCW posted in COVID area as compared to HCW posted in non-COVID area of the hospital, among the HCWs who were directly involved in COVID-19 patient care, and who spent more time in suspected or confirmed COVID-19 patients' room and exposure to more number of such patients.[24,25] In the study from Mumbai by Kumar *et al.*, significantly higher seropositivity was noted in ancillary workers [18.5%, than doctors (7%) and nurses (6.8%)].[12] Iversen *et al.*[18] in their study found significantly higher seroprevalence in HCW participants who were posted in the dedicated COVID-19 wards (95/1321; 7.19%) as compared to the HCW who were frontline HCW but posted in other areas of the hospital (95/696; 4.35%).[18] However, several authors also have reported similar or lower seropositivity in high-risk HCWs as compared to HCW who belonged to low-risk category.[9,17,26]

Surprisingly it has been observed in many non-COVID-19 hospitals have higher seroprevalence among HCW than COVID hospitals across the globe. It may be a common believe and assumption that HCW in COVID care set up have a higher chance to get COVID-19 infections. But the frequency of SARS-CoV-2 antibodies among HCW in the non-COVID-19 hospital shown to be 32.7% when compared to 28.2% in COVID hospital ($P = 0.129$).[9]

The present study showed highest seroprevalence among housekeeping and sanitation staff and lowest among teaching medical faculty. It is probably linked to teaching doctors having better knowledge of transmission of infection and they are overcautious while working in a COVID-19 care facility, and have better use of PPE, and proper adherence to standard practice for donning and doffing. The same is also observed among the doctors and nurses in the high-risk group were more cautious and better protected as compared to the lower to intermediate risk group (1.2% vs. 5.4%), with an odds ratio of 0.22 (95% CI: 0.04-- 1.35); ($P = 0.13$).[9]

The present study observed high seropositivity among housekeeping and sanitation personnel which is possibly because of non-strict adherence of hygiene and infection prevention control measures and their enhanced risk of contracting infection in the community. Among those who are involved in direct patient care, it was higher among nurses and resident doctors who are the frontline staff and have maximum exposure to the patient as compared to faculty. Barett *et al.*[25] have reported high seropositivity in nurses as compared to other categories of HCW which has been attributed because of more time spend with COVID-19 positive patients,[26] whereas Iversen *et al.*[18] found high positivity in medical students which was explained due to a social gathering for the medical students.[18]

Table 4: Factors associated with sero-positivity among HCWs (*n*=821)

Variable	Sero-negative *n*=789 (%)	Sero-positive *n*=32 (%)	*P*	Odds Ratio (95% CI)
Gender				
Male	516 (96.4)	19 (3.6)	0.483	Ref
Female	273 (95.5)	13 (4.5)		1.29 (0.63-2.66)
Risk Category during work (i.e. worked in COVID areas for at least 15 days)				
Low-risk	408 (97.8)	9 (2.2)	0.009	Ref
High-Risk	381 (94.3)	23 (5.7)		2.74 (1.25-5.99)
Direct contact with any COVID-19 patient at home or workplace				
No	279 (96.2)	11 (3.8)	0.909	
Yes	510 (96.0)	21 (4.0)		1.04 (0.49-2.20)
Had any COVID 19 related symptoms in last 3 months				
No	488 (97.2)	14 (2.8)	0.039	Ref
Yes	301 (94.4)	18 (5.6)		2.08 (1.02-4.25)
Ever tested by RT PCR for SARS-COV-2 infection				
No	457 (96.2)	18 (3.8)	0.851	Ref
Yes	332 (96.0)	14 (4.0)		1.07 (0.52-2.18)
Tested positive on RT PCR for SARS-CoV-2 before sero survey				
No	782 (96.5)	28 (3.5)	0.001	Ref
Yes	7 (63.6)	4 (36.6)		15.97 (3.28-37.66)
Locality declared as containment zone during COVID 19 pandemic				
No	622 (95.8)	27 (4.2)	0.450	Ref
Yes	167 (97.1)	5 (2.9)		0.69 (0.262-1.818)
Ever taken HCQS prophylaxis during COVID-19 pandemic				
Yes	91 (93.8)	6 (6.2)	0.215	Ref
No	698 (96.4)	26 (3.6)		0.56 (0.22-1.40)
Ever taken any vaccine for seasonal influenza				
Yes	54 (98.2)	1 (1.8)	0.409	Ref
No	735 (96.0)	31 (4.0)		2.27 (0.30-16.56)
History of BCG vaccination during childhood				
Yes	552 (96.8)	18 (3.2)	0.099	Ref
No	237 (94.4)	14 (5.6)		1.81 (0.88-3.70)

The knowledge of seroprevalence of SARS-CoV-2 antibodies among HCWs is useful for assessing the level of exposure among hospital personnel which further gives an idea of the adequacy of infection control measures taken. More importantly, this information would aid in work stratification of HCWs and thus better healthcare resource planning during COVID-19 pandemic in primary care settings. Our study had various strengths and limitations. We adopted random sampling and all cadre of HCWs were selected in proportion to their size, thus eliminating the change of over and underestimation of seropositivity. HCWs who were either tested earlier for SARS-CoV-2 or not had almost equal seropositivity which showed that HCWs having higher perceived risk were not overrepresented in the sample. Antibody testing was done on the same day within 3--4 h of sample collection. Study laboratory is ICMR approved laboratory for conducting COVID-19 RT-PCR testing and has quality control mechanisms in place. Study was conducted in a hospital which catered to both COVID-19 and non-COVID-19 patients and thus was a usual setting for most of the hospitals. However, it was a single center study and seropositivity among HCWs would be correlated to background seroprevalence in the community. Thus, seropositivity estimated in this study may not be generalizable to other areas in different stages of pandemic.

Our study noted higher seropositivity of some of the HCW posted in COVID areas as compared to HCW posted in non-COVID area of the hospital. Many HCW who were seropositive were not tested by RT-PCR before as they were not symptomatic.

Rigorous practice of COVID appropriate behavior and PPE by primary care physician providing direct care of all kind patients can efficiently prevents COVID-19 transmission.

In conclusion, the overall low seroprevalence of SARS-CoV-2 antibodies in our HCWs may be due to effective infection control practice. However, the higher seroprevalence among the HCW posted in dedicated COVID ward indicates the need of more stringent implementation of infection prevention and control measures and strict monitoring of symptoms to take adequate measures for prevention of virus transmission between the patients and HCWs.

Declaration of patient consent

The authors certify that they have obtained all appropriate participant consent forms. In the form the participants have given their consent for their images and other clinical information to be reported in the journal. The participants understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Conflicts of interest

There are no conflicts of interest.

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