

Original research article

## SARS CoV 2- specific Serological status of Health Care Workers in a Covid Tertiary Care Hospital in Bangalore, Karnataka.

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### Abstract

**Background:** SARS-CoV-2 (Severe acute respiratory syndrome corona virus 2) seroprevalence studies are being conducted all over the world to identify the level of exposure to the virus, thus giving us a true estimate of the hidden burden of infection in undiagnosed asymptomatic subjects.

**Aim:** To determine the serological status for SARS CoV 2 among unvaccinated frontline health care workers in a Covid designated hospital.

**Materials and methods** - A mono-centric, retrospective, observational, cross sectional study was carried out with the institutional ethical committee approval (CTRI/2021/03/031933). 269 HCW were tested for SARS COV-2 antibodies. Out of these, 250 were frontline HCWs posted in Covid wards. Their data on demographics and symptomology was collected through validated Questionnaires. HCWs with active symptoms and a positive RT-PCR history of less than 14 days at the time of testing were excluded. IgG antibodies were detected by CLIA with 100% sensitivity and 99.8% specificity. A p value of <0.05 was considered as statistical significance.

**Results:** Data regarding RT-PCR and serological status was collected from all the 250 subjects, but only 174 questionnaires which were completed was analyzed statistically. Among the 250 subjects, 43(17.2%, 95%CI: 0.125,0.219) were seropositive and 14(5.6%) were positive for RTPCR. 70%(n=30) of the seropositive HCWs were totally asymptomatic. About 84% of the asymptomatic seropositive subjects were RT-PCR negative, thus reflecting the hidden burden of the infection. Further statistical analysis on the background characteristics was done only for 174 individuals out of the 250 HCWs. 19% of the medical staff were reactive compared to 37.7% of reactivity among the non-medical staff and this was the only statistical significance observed(p=0.008). No significant difference was noted with respect to gender, age, department, BCG vaccination, use of immune boosters or HCQ, no of days of exposure.

**Conclusion:** This analysis helped us generate a pre vaccination baseline information regarding the proportion of asymptomatic / symptomatic frontline workers with respect to serological status. If asymptomatic, they remain undiagnosed, thus playing a significant role in Covid transmission and crippling the healthcare system, and emphasize on educating our non-medical staff on the importance of PPE use, provides an opportunity for generating a database and data

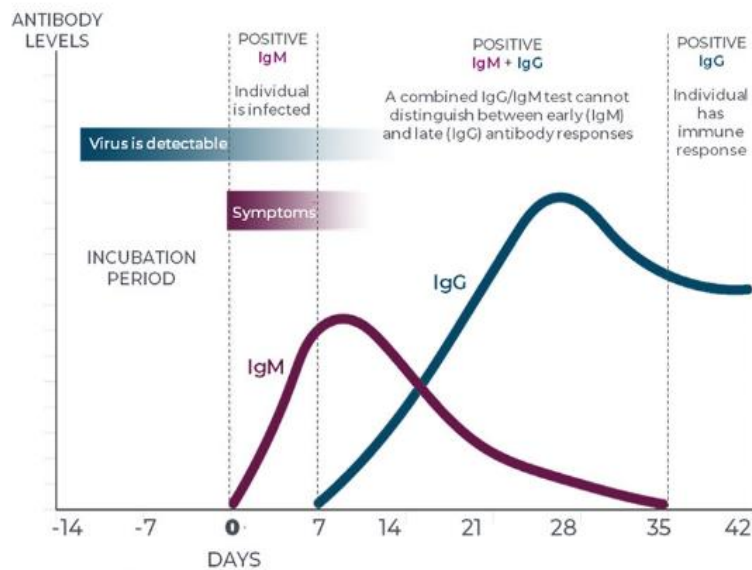
comparison of this newly emerged viral disease, thus helping in predicting and monitoring the future course of the pandemic.

**Keywords:** Covid19, SARS-CoV2 infection, IgG antibody

## Introduction

As the December sun rose, the world witnessed the emergence of coronavirus disease 19 (CoViD 19). The cases clinically resembled viral pneumonia. WHO announced the official name for this disease as covid19 on 11<sup>th</sup> February 2020, also known as SARS-CoV-2 due to its relationship to the virus responsible for SARS outbreak of 2003. On the 11<sup>th</sup> March, it was declared a pandemic<sup>1</sup>. India saw its 1<sup>st</sup> Covid case on 30 January 2020.

The virus shows human to human transmission which can occur via droplets, contaminated hands or surfaces<sup>3</sup>. The infection begins with flu like symptoms like dry cough, rhinitis, loss of smell or taste, headaches, muscles and joint pains and diarrhoea. A few days later patient may also suffer from fever and respiratory distress<sup>4</sup>. The infection with SARS-CoV-2 usually leads to seroconversion (Development of IgG antibodies in the blood of an individual who previously did not have detectable antibodies), 11 – 14 days after 1<sup>ST</sup> symptoms<sup>4,5,6</sup>. These antibodies are formed against the Spike protein component of the virus, which is critical for its function and causing the infection<sup>7</sup>.



Lab testing has played an essential role in confirming the diagnosis of Covid 19 and these tests are based on quantitative viral RNA or antibody analysis. RT-PCR is the gold standard test for the diagnosis of Covid 19 but with limitations, such as being expensive, time consuming and it requires biological expertise. Inappropriate sample collection leads to false negative reports which could have dramatic consequences, i.e. transmission of virus by an undetected contagious patient. Also, only the symptomatic individuals are usually tested and diagnosed. This also leads to a significant underestimation of the SARS-CoV-2 cases and hence affects the sero-prevalence estimation. Testing should be simple and quick. Hence one should consider rapid diagnostic tests, especially to identify the carriers who can have antibodies developed by the host B cell in response to the Covid antigen. Even if the serological tests are not as effective

as PCR during active infection, they can detect antibodies for a long period after disease recovery<sup>3,5,8</sup>.

WHY health care workers (HCWs) - Frontline HCWs are the personnel in direct contact with patients and body fluids. They include doctors, nurses and staff assisting with patient care such as bathing, eating, cleaning and housekeeping, who are in the forefront in the fight against the present Covid pandemic. Throughout the progression of the pandemic, HCWs have experienced high levels of exposure to SARS-CoV-2 infection, thus representing a large proportion of new source of infection. This has resulted in a great deal of anxiety and distress amongst the providers due to concern for self-infection with Covid 19 and family exposure<sup>9</sup>. Also, they do not have the luxury of work from home and need to commute by common/public transport<sup>10</sup>. This is of concern as the transmission is not only through symptomatic or lab confirmed cases but also from the pre-symptomatic and asymptomatic individuals reporting with non Covid illness. The sero-conversion rate among our frontline providers could provides the following valuable information –

1. Knowledge of immune status of oneself might help to overcome fears of getting infected, thus assist in safe deployment of the staff within the hospital system.
2. Rough estimate of the DARK FIGURE of the unreported infection (prevalence/infection rate).
3. Effectiveness of our PPE strategy and thus reduce nosocomial spread.
4. Planning and execution of infection control measures in a hospital setting and help in an effective national response to this pandemic<sup>4,6,10</sup>.

In the pre vaccination era, when a vaccine for Covid 19 was still being developed and there was no short or sure treatment, rapid and extensive diagnosis could be the way to reach the grass root level of the disease and break widespread chain of the deadliest infection<sup>8</sup>. A systematic “test and trace” program can control the spread of the virus<sup>6</sup>. Since the inception of Covid 19 infection, most of the testing was limited to symptomatic cases and even now not many details have been reported to access the asymptomatic cases<sup>8</sup>. With this point of view, detecting carriers becomes crucial to prevent local spread.

Serosurvey involves collecting and testing of serum specimen from a sample of a define population over a given period to detect the presence of antibodies<sup>11</sup>. Seroprevalence of IgG antibodies against SARS-CoV-2 is an important tool to estimate the true extent of infection in a population. **Seroprevalence studies have been sparse in South East Asia including India**, which, as of now, carries the third largest burden of confirmed cases in the world. The 1<sup>st</sup> national serosurvey, was conducted in May – June 2020 which showed a seroprevalence of 0.73%, which equated to 6.4 million infections, but the caseload through RT-PCR was only 52,592. Again a 2<sup>nd</sup> national survey was conducted in Aug – Sept 2020 in the same districts. The seroprevalence was 6.6% which roughly equates to 85million infections, but the case load reported was only 2.3 million. Also, these studies were conducted in general population using ELISA method<sup>11</sup>.

#### SEROLOGICAL TESTING OF COVID –

Accuracy of a serological test depends on the sensitivity and specificity of the test. A highly sensitive test correctly identifies the true positives - patients that have the disease and a high specific test correctly identifies the true negatives - patients that do not have the disease. For SARS-CoV-2 antibody testing, the CDC suggests use of tests with a specificity  $\geq 99.5\%$  to minimize the potential for false-positive results. False positives might lead somebody to believe that they are immune when they are not<sup>11</sup>.

The assays being used for serosurveys are mainly ELISA (enzyme-linked immunosorbent assay), lateral flow immunoassays and CLIA assays<sup>8</sup>.

India used its Made in India - COVID KAVACH ELISA kits to conduct nationwide seroprevalence survey but the kit showed a specificity of 97.9% and sensitivity of 92.37% which lead to high number of false negatives, that means it leads to missing of positive cases and transmission of the Covid virus. Also, there are no details in the public domain about which viral proteins the Kavach assay looks for, ie Nucleocapsid protein or Spike protein which are important components required viral infection and immunity<sup>12,13</sup>.

Most of the private laboratory use lateral flow and CLIA assays.

The rapid antibody testing kits follow the lateral flow chromatographic immunoassay principle which qualitatively assesses the presence of the antibodies. Though the sensitivity and specificity of these kits are claimed to be above 98%, these kits too do not mention which protein is the antibody of interest it is against.

CLIA - Chemiluminescent immunoassay is a quantitative method. These assays detect antibodies to the RBD (Receptor Binding Domain) of the spike protein which is more sensitive than detection of antibodies to the N protein, due to earlier immune response to the S antigen and more specific due to lower cross-reactivity with less conserved regions of spike proteins existing in other coronaviruses<sup>14</sup>.

Majority of confirmed cases of COVID-19 are in low- and middle-income countries, yet there are relatively few studies published on the serosurveillance of SARS-CoV-2 in these countries and these studies focus on prevalence in general population<sup>15</sup>. To our knowledge, only 2 serosurveys have been conducted in HCWs in Bangalore, Karnataka using CLIA<sup>16</sup>. WHO, in its scientific briefing on 2nd April 2021, encouraged the member states to conduct seroepidemiological studies for better understanding of the extent of infection<sup>17</sup>. In this study, we are interested in determining the SARS-CoV-2 IgG antibody status (seroprevalence) among the HCWs using a high sensitive and specific method, thus identifying the burden of hidden infection in an institutional set up.

## OBJECTIVES

1. To estimate the Covid19 IgG antibody levels in unvaccinated frontline HCWs
2. To determine the prevalence of the sero-positive HCWs (thus explore the possibility of subclinical/asymptomatic infection)

## MATERIALS & METHODS

The study was carried out with the approval of ethics review board of RRMCH-IEC/19/2021 and CTRI registration - CTRI/2021/03/031933.

- **Type of study** - A mono-centric, retrospective, observational, cross sectional study
- **Place of study** - Rajarajeswari Medical College and Hospital, Bangalore - RRMCH became a designated Covid hospital in the month of June 2020

- **Participants and setting** –

Data regarding the SARS COV-2 specific IgG antibody status was collected from Department of Biochemistry. 269 HCW of RRMCH had been tested for SARS COV-2 Antibodies. Out of these, 250 HCW were posted in Covid wards (frontline workers as obtained from human resource department of RRMCH). HCW included Doctors, nurses, housekeeping, attendants and lab technicians.

Consent form and validated Questionnaires (google form) were sent to the 250 HCW through WhatsApp/in person or telephonically to collect data on demographics and symptomology.

HCWs who had active symptoms of Covid 19 and history of a positive RT-PCR of less than 14 days history at the time of testing were excluded.

**Testing** – SARS COV-2 specific IgG antibody in the study population was detected using Beckman Coulter Access 2 chemiluminescent qualitative Immunoassay with 100% sensitivity and 99.8% specificity. This test was interpreted as reactive/antibodies present if S/CO >1 or non-reactive/antibodies absent if S/CO < 1<sup>18</sup>.

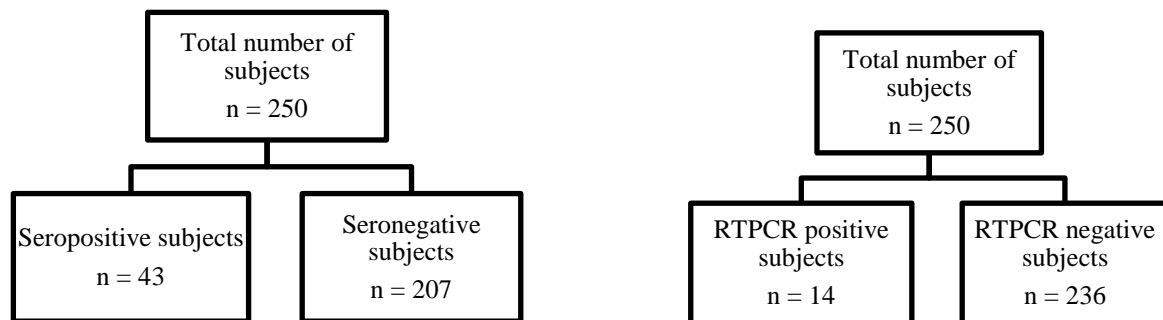
#### STATISTICAL ANALYSIS

Categorical/qualitative variables will be calculated as frequency and percentage, continuous/quantitative variables will be represented as mean ( $\pm$  SD). Chi square test and T test will be used to calculate the statistical differences for categorical variables and continuous variables respectively. A p value of <0.05 will be considered as statistical significance.

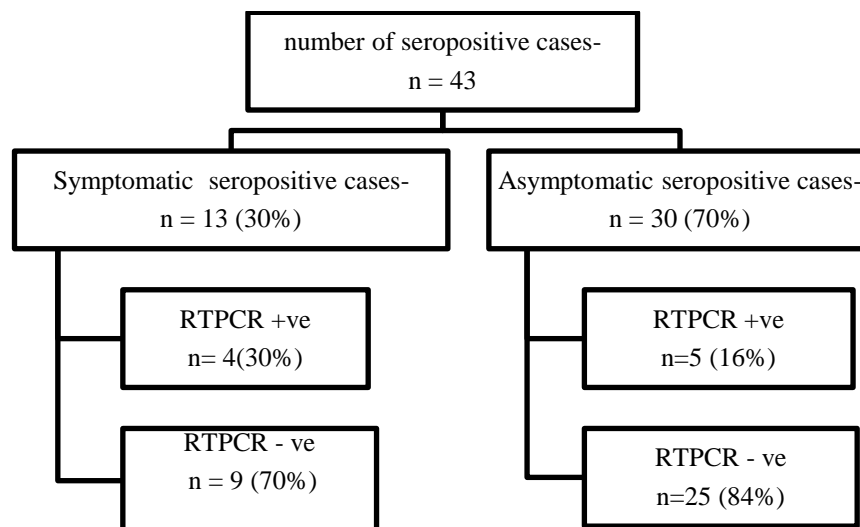
#### RESULTS

Data regarding RT-PCR and serological status was collected from all the 250 subjects in this retrospective study, but only 174 questionnaires were complete, which were finally included and analysed statistically.

Among the 250 subjects, 43 (17.2%, 95%CI: 0.125,0.219) were seropositive and 14 (5.6%) were positive for RTPCR. (Figure 1)



**Figure 2- Prevalence of RT-PCR status and symptoms in seropositive HCWs**



Out of the 43 seropositive subjects, 70% (n=30) of the HCWs did not give any history of symptoms such as fever, sore throat, runny nose and headache. About 84% of the asymptomatic seropositive subjects were RT-PCR negative, thus reflecting the hidden burden of the infection. Based on the completed questionnaire, further statistical analysis on the background characteristics was done only for 174 individuals out of the 250 HCWs (Table 1).

**Table 1: Background characteristics and SARS CoV 2 antibody positivity of the 174 HCWs**

s.no	Characteristics	Number of HCW n=174 (%)	SARS CoV 2 Antibody positive	P value
			N = 43 (%)	
<b>1</b>	<b>Gender</b>			
	Male	69 (40)	16 (23.5)	0.77
	Female	106(60)	27 (25.5)	
<b>2</b>	<b>Age in years</b>			
	≤ 25 years	17 (9.7)	4 (23.53)	0.43
	26-30 years	85 (48.8)	16(18.8)	
	31-40 years	46 (26.4)	14 (30.4)	
	41-50 years	17 (9.7)	6 (35.3)	
	51-60 years	9 (5.1)	3 (33.3)	
<b>3</b>	<b>Job title</b>			
	Medical	121(69.5)	23 (19.0)	<b>0.008</b>
	Non Medical	53 (30.5)	20 (37.7)	
<b>4</b>	<b>Department</b>			
	Medicine	8(4.6)	1 (12.5)	0.71
	Anaesthesia	21(12)	7 (33.3)	
	OBG	15(8.6)	5 (33.3)	
	Pulmonology	2(1.1)	0	
	ENT	19(10.9)	4 (21.1)	
	Others	109(62.6)	26 (23.8)	
<b>5</b>	<b>BCG vaccination status</b>			
	Yes	155(89.1)	35 (22.6)	0.72
	No	6(3.4)	1 (16.6)	
	Unknown	13(7.5)	7 (53.8)	
<b>6</b>	<b>History of chronic diseases</b>			
	Yes	13(7.5)	4 (30.8)	0.5
	No	161(92.5)	39 (24.2)	
<b>7</b>	<b>Duration of exposure in Covid Ward</b>			
	Ward visit for few minutes/hours for inspection/ consultation/meeting a Covid patient	33(18.9)	5 (15.1)	0.35
	Have worked for 9 days or less	10(5.7)	3(30)	

	Have worked for more than 10 days	131(75.3)	35 (26.7)	
8	History of Hydroxychloroquine (HCQ) prophylaxis			
	Yes	39 (22.4)	10 (25.6)	0.87
	No	135 (77.6)	33 (24.4)	
9	History of taking immune booster/medications			
	No	78 (44.8)	15 (19.2)	0.1
	Yes	96 (55.2)	28 (29.1)	

The only statistically significant difference seen between the Medical and non-medical staff ( $p=0.008$ ) was that 19% of the medical staff (professors, associate professors, assistant professor, residents, interns and nurses) were reactive, whereas 37.7% of the non-medical staff (housekeeping, attendants and lab technicians) were reactive.

No significant difference was noted with respect to gender, age, department, BCG vaccination, use of immune boosters or HCQ, no of days of exposure.

### Discussion

This is a special population serosurveillance study involving frontline HCWs. By virtue of their proximity and long duration of exposure, they are at a higher risk of contracting Covid infection which may or may not be symptomatic.

This study is aimed to assess the serological status among the unvaccinated frontline HCWs (post 1<sup>st</sup> wave) with an objective to estimate the burden of disease among the frontline HCW both diagnosed and undiagnosed. Also, to emphasize the significance of usage of PPEs even in asymptomatic personnel.

This retrospective analysis done in a Covid designated hospital has demonstrated 17% seropositivity for SARS CoV 2 among frontline HCWs but only 5.6% were RT-PCR positive. The data analysis has shown a gross mismatch between seropositivity and RT-PCR positivity. We also observed 27% of the asymptomatic cases were seropositive. A study done by Chandrasingh S et al<sup>19</sup>, in a Bangalore based hospital in 2 phases has showed that the seropositivity was minimal among RT-PCR Negative HCW during the initial phase of the pandemic which increased to 9.2% during the 2<sup>nd</sup> phase. Another study by Madhura N S et al<sup>20</sup>, done in Institute of Nephrourology, Bangalore, showed a seropositivity of 6.3% among RT-PCR negative individuals. Studies from other countries among HCW showed a seroprevalence as low as 0% in Malaysia to as high as 26% in UK<sup>19</sup>. These variations can be attributed to many factors such as subject selection (HCW from Covid wards vs HCWs from ICU), different techniques used for SARS CoV 2 antibody estimation (ELISA vs CLIA), socio cultural differences, and influence by genetic, ethnic and climatic factors<sup>21</sup>.

The serological status gives valuable insight regarding the disease burden of both reported and unreported infection and the effectiveness of our PPE strategy. Asymptomatic infection has been more commonly reported in Indian serosurveys, exceeding 90% in some, in contrast to high-income countries, where about one-third of infections are asymptomatic<sup>22</sup>. Hence, policies should be implemented for regular serosurveillance which will determine the extent of community transmission through undocumented, unreported asymptomatic cases, in other words influence herd immunity.

This study showed that only 19% of the medical staff were seropositive as compared to the 37.7% among the non-medical staff. This was in line with a study by Mahto M et al <sup>21</sup>, which stated that occupation emerged as a significant influencer of IgG seropositivity. This could be due to a possibility in breach of personal protective equipment usage while discharging duties. The breach could be attributed to lack of awareness/knowledge and the non-medical staff tend to handle patients more closely (assisting in changing clothes and bathing the patients etc). Use of hydroxychloroquine did not show any correlation in the serological status of the HCWs. Also, other demographic and background characteristics did not influence the serological status in our study.

## CONCLUSION

This study gives an insight regarding the proportion of asymptomatic / symptomatic frontline workers with respect to Covid specific antibody status. If asymptomatic, they remain undiagnosed and thus play a significant role in the spread of Covid, crippling the healthcare system and a threat to the vulnerable group of patients and community.

This analysis has helped us generate a pre vaccination baseline information on our HCWs and the need to educate our non-medical staff on the importance of PPE use. This study also provides an opportunity for generating a database and data comparison of this newly emerged viral disease, thus helping in predicting and monitoring the future course of the pandemic.

## LIMITATIONS

1. Single centred study
2. Small sample size

## ACKNOWLEDGEMENT

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Source of the image : Partners in Health Guide Covid-19

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