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Research Article

COVID-19 SERO SURVEILLANCE AMONG LABORATORY PERSONNEL IN COVID TESTING LABORATORY IN A TERTIARY CARE HOSPITAL IN KERALA

Pressy Sankar, Shabina M B, Beena Philomina J and Nimin Hafeez

Department of Microbiology Government Medical College Kozhikode

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ABSTRACT

Introduction: The front-line health care workers are very susceptible to COVID-19 infection. In Kerala, since the diagnosis of first case of COVID 19, the laboratories had to reorganise and start new to implement its testing.Staff working in COVID 19 testing laboratory are exposed to samples of patients suspected of COVID -19. Serological surveys have helped in determining the prevalence of COVID-19 infection in the community. This has helped to understand the infection transmission dynamics in different healthcare settings and helped in designing strategies for prevention of further transmission of infection

Aim: The aim of our study was to determine the seroprevalence of COVID-19 among laboratory personnel working in COVID testing laboratory and to determine the source of infection. The study also aimed to determine the association of several factors affecting seropositivity.

Materials and Methods: A cross-sectional study was performed among HCWs in the COVID testing laboratory in a tertiary care hospital. The participants who volunteered were asked to complete a questionnaire and give written consent to participate in the study. Their blood was collected for analysis of IgG antibodies to SARS-CoV-2 by ELISA.

Results: Of the 120 participants 20 tested positive for COVID 19 with a seroprevalence of 13.3%. The source of infection in majority of symptomatic PCR positive individuals was from the community. Majority of the individuals who were COVID positive (12/20) had antibody response (p value <0.0000001)

Conclusion: The study shows that the people working in COVID 19 testing laboratory are not at increased risk of COVID 19 infection if adherence to infection prevention measures are practised diligently. The seroprevalence among the laboratory personnel is same as that of the seroprevalence in the general population from the same geographical area.

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INTRODUCTION

In December 2019, a mysterious pneumonia started spreading in China, starting in Huanan sea food market Wuhan, Hubei caused by a novel corona virus. It was initially named as 2019 novel corona virus (2019 N CoV) and later renamed as SARS - CoV-2 virus.[1]COVID-19 infection has been reported from every continent and has infected more than464 million people and has caused more than6 million deaths spread across 222 countries world wide.[2] This dreaded virus has high infectivity, increased transmissibility even during pre-symptomatic period and has over time evolved to have variants like alpha, beta, delta and finally Omicron.[3][4]The prevalence of asymptomatic infections along with clinical disease has accelerated the spread of the disease straining the health care system of each and every country across the globe.[5] The main mode of transmission is through respiratory droplets .[6]

Corresponding author:* **Pressy Sankar
Department of Microbiology Government Medical College Kozhikode

The health care workers involved in covid testing are exposed to COVID-19 infection when dealing with samples. The impact, apart from the health hazard it poses, also forces the HCWs to go on isolation along with close contacts, putting a severe strain on the human work force already grappling with increased workload caused by the pandemic. They can be asymptomatic sources of infection who can transmit the disease to the close contacts. [7] Early diagnosis in them is essential to plan the infection control measures to be taken to prevent further spread.[8].Every laboratory must follow strict biosafety guidelines and infection control practices with the sole aim of prevention of spread of infection While handling respiratory samples guidelines recommend wearing filtering facepiece respirator 2 (FFP-2) mask, double pairs of gloves, and a disposable gown. First pre-treatment of samples is done in a microbiological safety station, and further biological inactivation is needed before viral RNA extraction and genome amplification for SARS-CoV-2 detection is carried out.[9,10]According to WHO 80%of COVID-19 cases are

mild or asymptomatic. Hence timely screening of HCWs helps in rapid identification and their isolation to prevent further transmission in the laboratory and the community they are part of.[11]The gold standard for the detection of SARS COV-2 is by detection of viral RNA using real-time reverse transcription PCR (RT-PCR) in properly collected nasopharyngeal and/or oropharyngeal swabs.[12]

Serological surveys have helped in determining the prevalence of COVID-19 infection in various cohorts and communities. They have helped in understanding the transmission dynamics, cumulative prevalence, and the proportion of the remaining susceptible population to COVID-19.[13][14] The asymptomatic, subclinical infections are also identified, and infection control practices can be modified accordingly for a good public health response. They are relatively quicker, simpler and cheaper than the molecular methods but are not sensitive enough. [15]In this study, we aimed to determine the SARS-CoV-2 seroprevalence in faculty and staff working in Covid testing laboratory.

MATERIALS & METHODS

Aim: The aim of our study was to determine the seroprevalence of COVID-19 in laboratory personnel working in COVID testing laboratory.

Objectives

Primary

1. To detect SARS COV2 antibodies in health care workers dedicated to COVID testing laboratoryand to determine their source of infection.

Secondary

1. To estimate the proportion of asymptomatic individuals among the seropositives
2. To determine the association between symptoms & immune response in COVID positive individuals

Study design*:* The study is a cross sectional study of COVID-19 Sero surveillance among Laboratory personnel working in the COVID testing Laboratory (Regional Virus Research and Diagnostic Laboratory RVRDL) under the department of Microbiology, Govt Medical College, Kozhikode.

Study population: The laboratory personnel include Medical Officers, Research Scientists, Lab technicians, Data entry operators, Lab Assistants & the cleaning staff working in the COVID testing laboratory.

The purpose of the study was explained to all the individuals recruited into the study and samples were collected from the participants after obtaining their informed consent.

Data collection*:* Data was collected from each participant recruited into the study to complete a proforma which covers demographic and exposure information.

METHODS

Specimen collection*:* About 2-3 ml of venous blood was collected from each participant by staff trained in Infection prevention and control measures like safe handling practices and spill decontamination procedures. All specimens were properly labelled and transported to the laboratory immediately after collection. The serum was separated from whole blood by centrifuging at 4500 rpm for 5 minutes and stored at -80 c till processing.

Serological testing*:* The presence of SARS-CoV2 antibody was determined using EUROIMMUNE Anti SARS CoV -2 IgG ELISA. The assay provides a semiquantitative invitro determination of human antibodies of IgG class against SARS CoV-2in serum. The reagent wells of the ELISA are coated with S1 domain of the spike protein of SARS CoV-2 expressed recombinantly in the human cell line HEK293.

The sensitivity and specificity of the test kit have been reported to be 90% & 100% respectively with 95% CI. The results were evaluated by calculating the ratio of the optical density of the control or patient sample over OD of the calibrator. A ratio of < 0.8 was taken as negative, > 0.8 – 1.1 as border line and > 1.1 as positive.

COVID testing was done by either RT PCR or Antigen testing. The RT PCR kits used were(1) Seegene (manufacturer)& (2) SD Biosensor.

The genes detected in the Seegene kit were E gene, RdRp& N gene. The cut off value was 40. The genes detected in SD Biosensor were E gene & ORF1ab gene. The cut off value was 36.

The antigen kit used in the study was Standard Q COVID-19 Ag test kit (SD Biosensor). Mouse monoclonal anyi-SARS-CoV-2 antibody conjugated with color particles are used as detectors for SARS-CoV-2 antigen device. The sensitivity & specificity of the Standard Q COVID-19 Ag test kit were 76.6% & 99.3% respectively with 95% CI.

RESULTS

Of 124 Healthcare workers in the COVID testing laboratory during the study period, 120 consented for the study. Among them 16 (13.3%) tested positive for COVID-19 IgG antibodies.

Table No1 SARS COV-2 seropositivity

COVID-19 Ig G Antibody	TOTALn=120(%)
POSITIVE (1.1)	16 (13.3)
NEGATIVE (<0.8)	104 (84.2)
TOTAL	120 (100)

Table No 2 Occupational status

Category	Study population	COVID-19 Ig G Antibody Positive n (%) =16
Medical Officers	26 (21.7)	2 (12.5%)
Research Assistants	2 ((1.7)	0
Lab Technicians	46 (38.3)	9 (56.25%)
JLA	25 (20.8)	3 (18.75%)
Data Entry Operators	16 (13.3)	1 (6.25%)
Cleaning Staff	5 (4.2)	1 (6.25%)
Total	120 (100)	16 (100)

The demographic data were as follows:

Table No 3 Gender wise distribution

Gender	SARS CoV-2 IgG LEVELS			p- value
	Positive	Negative	Total	
Males	4 (12.9)	27 (87.1)	31 (100)	
Females	12 (13.5)	77 (86.5)	89 (100)	0.99
Total	16 (13.3)	104 (86.7)	120 (100)	

*Fisher Exact Test

Out of the 120 samples tested, 31 (25.8%) were males and 89 (74.1%) were females.

Age: The median age of the study population was 34.9 (SD ± 10.4). In the study population, 74% (89) were in the age group of 20-40 years and 26% (31) was above 40 years indicating majority of the laboratory personnel were in the younger age group.

Table No.4 COVID-19 infection & seropositivity

SARS COV-2 IgG LEVELS	Positive	Negative	Total (n)	OR (95% CI)	p-Value
Diagnosed with COVID-19	12 (60%)	8 (40%)	20 (100)	36 (9.409-137.7)	<0.0000001
Not Diagnosed with COVID-19	4 (4%)	96 (96%)	100 (100)		
Total	16 (13.3%)	104 (86.7%)	120 (100)		

*Fisher Exact Test

Among the 120 study subjects, 20 were positive for COVID-19 infection by either RT-PCR or Antigen testing of whom 12 (60%) showed seropositivity, while 8 (40%) did not develop antibodies post infection.

Of the 100 HCWs who were not previously diagnosed with COVID-19, 4 persons developed antibodies.

Table No 5 COVID symptoms & seropositivity

SARS COV-2 IgG status	Positive	Negative	Total	p-Value
Symptomatic	10 (71.4%)	4 (28.6%)	14 (100)	
Asymptomatic	2 (33.3%)	4 (66.7%)	6 (100)	0.2745
Total	12 (60%)	8 (40%)	20 (100)	

*Fisher Exact Test

Among the 14 who were symptomatic, 10 (71.4%) showed seropositivity.

Among the 6 asymptomatic HCWs, only two persons developed antibodies post infection.

Table No 6 Common Symptoms

*Symptoms (n=20)	
Fever	12 (86%)
Headache	9 (64%)
Tiredness	8 (57%)
Myalgia	6 (43%)
Shortness of breath	6 (43%)
Loss of taste & smell	5 (36%)
Sore throat	4 (29%)
Coryza	3 (21%)
Cough	3 (21%)
Loose stools	1 (7%)

*multiple response

Statistical Analysis

Statistical analysis was done using PASW version 25.0 software. All qualitative variables were expressed as frequencies and percentages. Chi square test (or Fisher's Exact Test was used when the cell count ≤ 5) was used to determine the statistical significance. p- value < 0.05 was considered as statistically significant.

DISCUSSION

Ever since the news of SARS COV2 outbreak in China in 2019 [16] all the nations including India were on high alert and the first known case of COVID 19 was documented in three medical students from Wuhan arriving in Kerala as early as January 2020.[17] But because of effective disease containment and preventive strategies at community and state level the peak of the disease spread could be delayed, until August 2020 after which active caseloads showed a sharp rise to hit the peak by October and November and began to flatten thereafter. By the first week of January 2021 the active case load had risen to a cumulative total of 8,84,242 with 3587 deaths in the state[18]

The number of RT PCR tests for the detection of SARS COV-2 virus done in the COVID testing lab was around 700 in March 2020 which increased to 1700 by June 2020. The sample load at the time of study was around 1500 samples/day.Of the 124 HCW in the study population, 16 laboratory personnel were tested positive for COVID-19 IgG antibody with a seroprevalence of 13.3%.

A study conducted by Indian Council Of Medical Research, one month before the present study from Kerala showed a sero-prevalence of 11.6% among the general populationand a national prevalence of 21% [19]. The same survey reported the sero prevalence of SARS CoV2 antibodies among HCW at 25.7% at the national level.[20] Meanwhile the prevalence of SARS CoV antibodies across the global stage among HCW is reported as 8.7% according to a large meta-analysis.[21] The seroprevalence studies among the HCW show wide variation and is reflective of the disease burden in the community. In a study conducted in AIIMS, New Delhi in July 2020 the seroprevalence among the HCW was 13% which is in line with the present study [22].Seroprevalence studies focussing on laboratory staff are comparatively less. In a study in France evaluating the seroprevalence among laboratory staff, the seroprevalence was observed to be 2.3%.[23]In a study conducted by Jessy *et al* at Govt medical College, Thiruvananthapuram, SARS C0V-2 IgG antibodies was detected in 19.1% of the HCW.[24]

To our knowledge this is the only study focussing on seropositivity in HCWs working in COVID testing laboratory in Kerala. In the present study, the Lab technicians had a higher seropositivity compared to doctors and other staff. In a study by Rafi *et al* from Kerala, the seroprevalence among HCWs was found to be 8.5% and the seropositivity of the lab technicians was 7.1%.[25]Though antibody positivity was higher in Lab technicians, most of them got the infection from the community. None of them acquired the infection from the workplace. But three of them got infection due to room sharing at hostel as infection control practices including physical distancing & universal masking were not strictly followed despite advice on strict adherence to infection control practices.

In the study population, 12.9% males and 13.4% females were positive for Anti SARS CoV2 antibodies. The median age of the study population was 34.9 (SD ± 10.4). In the study population, 74% (n =89) were in the age group of 20-40 years and 26% (n =31) of the population were above 40 years . No association was seen between age & sex and COVID-19 seropositivity. A large study in HCWs in a tertiary care hospital in Poland showed no association between age, sex & IgG antibody to SARS COV2[26]. In another study conducted among HCW in a frontline hospital in Tokyo there was no significant difference in seropositivityin association with age and sex.[27]

Among the 120 study subjects, 20 (16.66%) HCWs had documented COVID-19 infection by either RT-PCR (12) or Antigen tests.(8)Of the 20 COVID positive patients, only 12 (60%) showed seropositivity. COVID-19 screening was done among the Laboratory personnel at regular intervals or when

symptomatic as per the infection control protocols of the Institution.

Among the eight persons who were seronegative post COVID 19infectionsin our study four were asymptomatic (50%). In a study in Bangladesh, it was observed that in 171 COVID positive individuals, the serological response in asymptomatic individuals were significantly lower than that of the mildly symptomatic group[28].

The symptomatic seronegative individuals had mild to moderate symptoms. The reason for seronegativestatuscould be due to mild infections not eliciting a robust immune response sufficient to cause a detectable antibody response or waning antibody levels over time[28,29].Serial screening for IgG levels at regular intervals could have unravelled phenomena like non development of antibodies, waning antibody levels and delayed seroconversion after a diagnosis of COVID 19 by RT PCR.[30]. In this study a repeat serological testing was not done in all the COVID positive seronegative subjects to look for a delayed immune response. Another reason for delayed humoral immune response is immunosuppressed state which was not the case in our study where all the participants are healthy and active.[31]

Of the 100 HCWs who were not previously diagnosed with COVID-19, four were seropositive(4%)**.** Two of them never had any symptom suggestive of COVID-19, signifying asymptomatic past infection. The other two had symptoms suggestive of COVID-19, but was not tested in time.

Of the 20 COVID positive patients, 14 were symptomatic. Among the symptomatics, 71.4% were seropositive while only 33.3% of the asymptomatics developed anti SARS CoV-2 antibodies. The p-value (<0.0000001) was significant showing that symptomatic disease occurrence is associated with a better antibody response.

Majority of the symptomatic patients presented with fever (90%) and headache (70%) Other symptoms were loss of taste & smell, tiredness, myalgia, sore throat, coryza, shortness of breath, cough and palpitations. One person developed loose stools.

Source of infection: The source of infection could be traced among 17 HCW in our study as from the community, spouse/family members as they were primary contacts of confirmed COVID 19 cases. In the remaining three persons the source of infection was their roommates in hostel facility who had documented COVID 19 infection. Outside the workplace universal masking and social distancing practices were not followed in all these cases. None of them acquired the infection from the workplace as strict infection control measures were followed in the lab like proper hand hygiene techniques, universal masking, physical distancing, use of appropriate PPEs. Their primary contacts in the laboratory were all tested negative for COVID 19 subsequently. Proper training of the staff was given regarding safe handling of the specimen, infection control practices, routine decontamination of the lab area and appropriate decontamination and disinfection strategies for Biomedical waste management along with regular screening of the employees at regular intervals or whenever symptomatic.

It is thought that HCWs are at increased risk of carrying and spreading COVID 19 infection because of the risks involved as in exposure to specimens from suspected COVID 19 patients. [32] In our study the prevalence of IgG Antibodies to SARS CoV2 is comparable to that of general population in the same geographical area but significantly less when compared to the national level seroprevalence in India. All those who tested positive for COVID 19 infection could trace the source as from community and not from the workplace. This shows that effective implementation of universal masking and hand hygiene practices would have helped in preventing the spread into HCWs.

The study shows that a vast majority of HCW are susceptible (87%) warranting urgent need for vaccination. The study was completed just before the vaccination drive for HCW at the national level. The data from this study will be a valuable tool to understand the baseline serological status to accurately assess the vaccine responsiveness post vaccination.

This is the first study looking at the seroprevalence of SARS CoV2 antibodies particularly among laboratory personnel dealing with COVID 19 samples. The study shows that the people working in COVID testing laboratories are not at increased risk of COVID 19 infection if adherence to infection prevention measures are practised diligently.

Limitations: The limitations to the study include that this is a single centre study with a limited sample size. The study was restricted to the faculty & staff working in COVID testing lab. The participants were all healthy and had no significant co morbidities. Also, in the study a repeat serological testing was not done in all the COVID positive seronegative subjects to look for a delayed immune response.

CONCLUSION

This study, the first of its kind among HCWs working in a COVID testing laboratory in Kerala shows that the laboratory personnelexposed to samples from COVID suspected patients are not at an increased risk of COVID 19 infection if adherence to infection prevention measures are practised diligently. The serological testing helped us to identify the asymptomatic past infections. The seroprevalence among the laboratory personnel is same as that of the seroprevalence in the general population from the same geographical area. None of the HCWs acquired the infection from the workplace, those who were seropositive had acquired the infection from the community. Our institution had implemented stringent workforce education on hand hygiene, social distancing and appropriate PPE usage, emphasizing the importance of infection control measures to prevent these infections. The findings of this study conducted on a one time basis support the need for active vaccination strategies prioritising HCWs.

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