Seroprevalence of COVID-19 and associated factors in a medical institution in Pakistan

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Objective: This study investigates the association of preventive measures with coronavirus disease (COVID-19) seropositivity.

Methods: This cross-sectional study was conducted at the Combined Military Hospital Kharian Medical College, Pakistan, in September 2020. A total of 442 participants from three different strata (faculty, students, and administration/technical staff) were enrolled using a convenient sampling technique. A rapid antibody testing method was used to detect antibodies. The Ichroma COVID Ab test is an in vitro diagnostic device that helps in the rapid identification of COVID-19 by measuring the levels of IgG and IgM antibodies against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in the blood. An automated fluorescent immunoassay system (AFIAS-6), with a clinical sensitivity of 95.8% and specificity of 96.7%, was used for qualitative analysis. A self-administered questionnaire was used to collect data, and data analysis was performed using SPSS version 25.

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**Abstract**

Aim: To investigate the association of preventive measures with coronavirus disease (COVID-19) seropositivity.

Methods: This cross-sectional study was conducted at the Combined Military Hospital Kharian Medical College, Pakistan, in September 2020. A total of 442 participants from three different strata (faculty, students, and administration/technical staff) were enrolled using a convenient sampling technique. A rapid antibody testing method was used to detect antibodies. The Ichroma COVID Ab test is an in vitro diagnostic device that helps in the rapid identification of COVID-19 by measuring the levels of IgG and IgM antibodies against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in the blood. An automated fluorescent immunoassay system (AFIAS-6), with a clinical sensitivity of 95.8% and specificity of 96.7%, was used for qualitative analysis. A self-administered questionnaire was used to collect data, and data analysis was performed using SPSS version 25.
Introduction

As of November 2020, the novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has infected over 50 million people worldwide. The resulting yet poorly understood disease, coronavirus disease (COVID-19), has claimed 1.25 million lives worldwide. In Pakistan, a total of 343,000 cases and approximately 7,000 deaths have been reported since March 2020. Although effective in curbing the spread of the infection, the almost 6-month-long “smart lockdown” imposed by the government of Pakistan dealt a heavy blow to the country’s economic, commerce, and education sectors and did not provide a practical and permanent solution for safe resumption of pre-lockdown activities.

Without a clear therapeutic strategy in place and an effective and widely available vaccine still in the preliminary stages of development, the importance of rapid, ubiquitous, cost-effective, and reliable diagnostic and screening tests is evident. There are two main approaches for testing patients for COVID-19: molecular tests that detect viral RNA and serological tests that assess the host response to the virus. Real-time reverse-transcriptase PCR (RT-PCR), despite being the most extensively used molecular method of diagnosing COVID-19, is not free of limitations, such as inconstant diagnostic precision as the disease progresses, expensive and scarcely available testing kits and equipment, and inability to provide information regarding previous SARS-CoV-2 infection.

As the country begins to tentatively lift restrictions on commercial establishments and educational institutions, determining the actual number of individuals infected in the past few months is vital for disease surveillance, contact tracing, assessing the possibility of herd immunity, and being better equipped to control further spread of the disease. It is presumed that a significant population in Pakistan that has been infected with SARS-CoV-2 has recovered and currently possesses some degree of immunity.

Seroprevalence studies offer insights into how well containment strategies, such as wearing masks, hand washing, and social distancing, are working to control transmission. This study aims to estimate the seroprevalence of COVID-19 and the preventive measures taken.

Materials and Methods

A cross-sectional study was carried out at the CMH Kharian Medical College. The study was conducted after the reopening of the Medical Institute in September 2020. Samples were collected using a convenient sampling technique. An a priori sample size estimation method was employed to calculate the sample size; 442 participants from three different strata (faculty, students, and administration/technical staff) were enrolled in the study. A rapid antibody testing method was used to detect the level of antibodies. The Ichroma™ COVID-19 Ab test was used as an in vitro diagnostic medical device. It detects IgG and IgM antibody levels in patients with early/mild, asymptomatic, or acute SARS-CoV-2 infection with a clinical sensitivity and specificity of 95.8% and 97.0%, respectively. An IgG level ≥1.1 was considered a positive result, and an IgG level <0.9 was considered a negative result. The procedure was performed by trained lab personnel from a local company dealing with medical equipment, while considering biosafety measures.

A self-administered questionnaire was used to collect data after obtaining informed consent from the participants. The confidentiality of the participants was ensured. The statistical software SPSS version 25 (Statistical package for social sciences by IBM) was used for data analysis. Frequency and percentage were calculated for qualitative variables. The chi-square test was used, and a p-value ≤0.05 was considered significant.

Results

In total, 442 participants were included in the study: 40 (9%) faculty members, 299 (67%) students, and 103 (23.3%) administration/technical staff members (Figure 1).

![Figure 1: Distribution of participants.](image-url)
With respect to factors related to COVID-19, 14.9% of the participants were symptomatic, 32.4% always used a mask, and 14% never wore a mask. A total of 69.7% of participants washed their hands frequently for 20 s, and 75.6% had awareness regarding social distancing (Table 1). The seroprevalence trend of COVID-19-related antibodies showed 16.96% positivity for IgG antibodies (Table 2).

Most of the administration/technical staff who tested positive for IgG were asymptomatic (68.42%) (Table 3). Seven faculty members tested positive for IgG, and they were all symptomatic, tallying the percentage of symptomatic faculty members to 100%. Similarly, 49 students tested positive for IgG, and they were all symptomatic, tallying the percentage of symptomatic students to 100%. In contrast, only 31.57% of the administration/technical staff members were symptomatic (Table 3). A significant association \(p < 0.001\) was found between adhering to safety measures (wearing masks, handwashing, social distancing) and the occurrence of COVID-19 (Table 4).

**Discussion**

The COVID-19 pandemic has led to a dramatic loss of human life worldwide, and the most drastic effects initially reported are linked to respiratory distress. Responding swiftly to the pandemic by enforcing preventive protocols and estimating the extent of severity of infection are the only measures that can be adopted until safe and efficient vaccines are made available.

Kharian, a tehsil located in Gujrat District, has the highest number of people constantly moving to and from European countries, such as Spain, Italy, Norway, England, and Middle Eastern countries. Many Pakistanis living overseas and Pakistanis of the above-mentioned nationalities have been linked to the increased spread of COVID-19 in the periphery of Gujrat District in the time period from February to May 2020. We know from the literature that seroprevalence studies have been conducted in hotspot areas to identify the trend of asymptomatic population counteracting the disease and to get an overview of the extent of the spread of infectious disease, particularly in the presence of asymptomatic patients.

In our study, we aimed to determine the seroprevalence of COVID-19 and study associated factors at the end of the first wave of the pandemic after the institute reopened. Among the 442 participants, 16.96% tested positive for IgG (Table 2), and , 14.9% were symptomatic (Table 1). Our study showed a slightly high seroprevalence compared to a nationwide study conducted in Spain in April by Pollán et al., which showed an overall seroprevalence of 5%, with a 2% prevalence of IgG antibodies against SARS-CoV-2 in asymptomatic individuals; the seroprevalence was

| Table 1: Factors related to COVID-19 (N = 442). |
|----------------|-------------------|------------------|
| Factors                  | Percentage (N)    |
| Symptomatic              |                   |
| Yes                      | 14.9% (66)        |
| No                       | 85.1% (376)       |
| Family members had symptoms in the past 6 months |                   |
| Yes                      | 16.0% (70)        |
| No                       | 84% (367)         |
| Wearing a mask           |                   |
| Always                   | 32.4% (143)       |
| Most of the time         | 29.9% (132)       |
| Sometimes                | 23.8% (105)       |
| Never                    | 14.0% (62)        |
| Using sanitiser          |                   |
| Always                   | 28.3% (125)       |
| Most of the time         | 34.8% (154)       |
| Sometimes                | 23.1% (102)       |
| Never                    | 13.8% (61)        |
| Hand washing             |                   |
| Yes                      | 69.7% (308)       |
| No                       | 30.3 (134)        |
| Maintaining social distance       | |
| Yes                      | 75.6% (334)       |
| No                       | 24.4% (108)       |

| Table 2: IgG results in different groups. |
|-----------------------------------------|
| IgG status   | Faculty | Student | Administration/technical staff | Total |
|--------------|---------|---------|--------------------------------|-------|
| Positive     | 7       | 49      | 19 (18.4%)                     | 75    |
| (17.5%)      | (16.3%) | (16.96%)|
| Negative     | 33      | 250     | 84 (81.5%)                     | 367   |
| (82.5%)      | (83.6%) | (83.03%)|
| Total        | 40      | 299     | 103                            | 442   |

| Table 3: IgG positivity in symptomatic and asymptomatic COVID-19 patients. |
|----------------|-------------------|------------------|
| IgG- positive | Symptomatic | Asymptomatic | Total number of IgG-positive patients |
| Faculty       | 7          | 7 (100%)     | 0 (0%)            | 7   |
| Student       | 49         | 49 (100%)    | 0 (0%)            | 49  |
| Administration/technical staff | 19        | 6 (31.57%)   | 13 (68.42%)      | 19  |
reported to be 5% higher in Madrid, a hotspot area compared to the coastal areas of Spain.16

In a study conducted in a county in Los Angeles, the seroprevalence of anti-SARS-CoV-2 antibodies was 4.34%, 17 and in May, the seroprevalence in university students was 4%.18 This is in line with the previously reported results by a study conducted in Santa Clara County and Los Angeles County, where the mean estimated prevalence was 4.65%. The high seroprevalence (16.96%) (Table 2) in our study might be associated with the overlapping of the end of the first wave of COVID-19 and the start of the second wave.19

In addition, blood sample analysis in 3,300 people in Santa Clara County in early April showed a seroprevalence of 2.8%.20 Low prevalence in both the student population and general population of Santa Clara County revealed that SARS-CoV-2 was not circulating in the student population before the closure of the campus in mid-March 2020; this shows that the general population has been adhering to preventive strategies and is not being exposed to the virus unnecessarily.

Serological surveys are the best means for estimating the prevalence of an infectious disease, particularly in the presence of asymptomatic patients.21 Our study reported an increased number of individuals who tested positive for IgG antibodies against SARS-CoV-2 and were asymptomatic. Compared to that in the above studies, the high prevalence in our study can be attributed to the timing of testing, as the tests were conducted in September, when the frequency of testing had increased relative to that during the start of the pandemic.

In addition, the high seroprevalence in our region is similar to the increased infection rate among ethnic minorities living in overcrowded, high-density areas. One major difference between our study and those conducted in Europe and the USA might be the population density, i.e., the number of people living in a stipulated space. Overcrowding in the subcontinent region might have led to a high number of seropositive individuals, despite adequate compliance to preventive measures.

Limitations

The results of seroprevalence studies depict the exposure to a specific disease but do not reflect the true incidence of that disease.22

Conclusion

This study showed a slightly higher seroprevalence than other studies as it was conducted at the end of the first wave of the COVID-19 pandemic; however, we are still far from achieving herd immunity. Furthermore, strict adherence to preventive measures is the only way to control infection spread until the development of vaccines.

Recommendations

It is recommended that patients be followed-up biannually to monitor antibody levels and to check for reinfection.23 Adherence to preventive protocols should also be monitored.

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Conflict of interest

The authors have no conflict of interest to declare.

Ethical approval

This study was approved by the Ethical Approval Committee of CMH Kharian Medical College (Approval no: 2000/19/CKMC/123, dated: 17.09.2020).

Authors contributions

IN conceptualised the study, analysed the data, and formulated and proofread the initial draft; AFA collected the data, contributed to the introduction, and proofread the draft; AMK conceptualised the study, contributed to the discussion and conclusion, and proofread the draft; IN contributed to the methodology, formulated the graphics, and proofread the draft; MSA contributed to data collection, methodology, and proofreading; MZA contributed to the collection of data and proofreading. All authors critically reviewed and approved the final draft and are responsible for the content and similarity index of the manuscript.

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