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Prevalence and Epidemiological Profile of COVID-19 Infection Detected Through RT-PCR Among Patients of Various Districts of Punjab, India

Saniya Ohri[1], Kanwardeep Singh[2], Shailpreet K. Sidhu[3], Narinder Singh[4]*

[1]Senior Resident, Department of Microbiology, Govt. Medical College, Amritsar, Punjab, India.
Email: ohrisaniya@gmail.com,
Orchid ID: 0000-0003-2129-5170.
[2]Professor, Department of Microbiology, Government Medical College, Amritsar, Punjab, India.
Email: kdmicrogmcasr@gmail.com,
Orcid ID: 0000-0002-8159-997X.
[3]Associate Professor, Department of Microbiology, Government Medical College, Amritsar, Punjab, India.
Email: shail78@hotmail.com,
Orcid ID: 0000-0002-9302-9675
[4]Associate Professor, Department of Pediatrics, Government Medical College, Amritsar, Punjab, India.
Email: narinder7258@yahoo.co.in,
Orcid ID: 0000-0002-0858-9011

*Corresponding author

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Abstract

Background: The COVID-19 pandemic has led to a dramatic loss of human life worldwide and presents an unprecedented challenge for healthcare systems worldwide. Earlier to SARS-CoV pandemic, coronaviruses were only thought to cause mild, self-limiting upper respiratory tract infections in humans. COVID 19 presents across a spectrum of symptoms. WHO recommends detection of unique sequences of virus RNA by Nucleic Acid Amplification Test (NAAT) such as real-time reverse-transcriptase polymerase chain reaction (rRT-PCR). The aim of this cross sectional study was analysis and confirmation of Nasopharyngeal/oropharyngeal swab specimen by real-time reverse transcription polymerase chain reaction (RT-PCR). **Material & Methods:** This was a cross-sectional retrospective study that reviewed records of samples collected from June 2021 to March 2022. Nasopharyngeal/oropharyngeal swab specimen were collected from suspected COVID-19 subjects of various districts of Punjab and referred to Viral Research Diagnostic Laboratory [VRDL], Government Medical College [GMC], Amritsar for laboratory analysis and confirmation by real-time reverse transcription polymerase chain reaction (RT-PCR). **Results:** During the present study, a total of 11,27,005 samples were analyzed from June 2021 to March 2022 for SARS-CoV-2 detection by ICMR approved COVID-19 RT-PCR kits. Out of total 11,27,005 cases, 24,466 cases (2.17%) were found to be SARS-CoV-2 positive while 11,02,539 cases (97.83%) were SARS-CoV-2 negative. **Conclusions:** Ever since the COVID-19 global pandemic emerged, the developing countries are facing challenges regarding its diagnosis. Isolation of the infected person will eventually decrease the Reproduction number i.e Ro which will further interrupt the transmission cycle leading to decrease in community spread.

Keywords:- RT-PCR, SARS-CoV-2, Coronavirus.

INTRODUCTION

The COVID-19 pandemic has led to a dramatic loss of human life worldwide and presents an unprecedented challenge for healthcare systems worldwide.[1,2] The economic and social disruption caused by the pandemic is devastating. In December 2019, Wuhan in China reported an outbreak due to a novel coronavirus (SARS-CoV-2) causing the disease, later termed COVID-19.[3] SARS-CoV-2 spread rapidly across the globe within two months and was declared as a pandemic in March 2020 by WHO.[4] Coronaviruses (CoVs) are

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enveloped viruses with a single-strand, positive-sense RNA genome approximately 26–32 kilobases in size. The term 'coronavirus' refers to crown like peplomer spikes giving appearance of solar corona.[5]

Earlier to SARS-CoV pandemic, coronaviruses were only thought to cause mild, self-limiting upper respiratory tract infections in humans. COVID 19 presents across a spectrum of symptoms. About 80% of the individuals with SARS CoV 2 infection, either remain asymptomatic or show mild symptoms of flu (e.g., fever, cough, sore throat); which may be managed at home or in isolation centres to assess the spread of transmission.[6] The remaining 10–15% have moderate to severe symptoms, and need institutional care ranging from oxygen therapy, intensive care to ventilator support. The elderly and those with comorbidities (e.g., diabetes mellitus, hypertension, renal diseases, etc.) are at higher risk of and meet adverse outcomes.[6]

WHO recommends detection of unique sequences of virus RNA by Nucleic Acid Amplification Test (NAAT) such as real-time reverse-transcriptase polymerase chain reaction (rRT-PCR).[7] ICMR also recommends use of FDA approved Cartridge-Based Nucleic Acid Amplification Test (CBNAAT) using Cepheid Xpert Xpress SARS-CoV2 for use under an emergency use authorization (EUA) which only detects E and N2 gene.[7]

Aims & Objectives

This was a cross-sectional retrospective study that reviewed records of samples collected from June 2021 to March 2022. Nasopharyngeal/oropharyngeal swab specimen were collected from suspected COVID-19 subjects of various districts of Punjab and referred to Viral Research Diagnostic Laboratory [VRDL], Government Medical College [GMC], Amritsar for laboratory analysis and confirmation by real-time reverse transcription polymerase chain reaction (RT-PCR).

MATERIAL AND METHODS

In the present study, we analyzed the data of Covid-19 samples received from different collection centres across the six districts (Amritsar, Pathankot, Tarn Taran, Gurdaspur, Hoshiarpur and Jalandhar) comprising regions of Punjab state of India over a period of 10 months i.e. from June 2021 to March 2022 in Viral Research Diagnostic Laboratory [VRDL], Government Medical College [GMC], Amritsar. For initial diagnostic testing for current SARS-CoV-2 infections, CDC recommends collecting and testing an upper respiratory specimen i.e. nasopharyngeal and oropharyngeal specimen.[8] Specimens were obtained and transported in viral transport medium under cold chain. The rejection criteria for the samples includes the samples which were not transported in cold chain with proper packaging or were leaked or if the patient's details did not match with the label provided on the samples. SARS-CoV-2 was then detected using ICMR approved COVID 19 RT-PCR kits.[9] It begins with nucleic acid purification from upper respiratory specimens, which is reverse transcribed into cDNA and amplified using a real-time PCR instrument with appropriate filter wavelengths.[9] In the process, probes anneal to two target sequences that are specific to SARS-CoV-2 (ORF1ab and N gene), and one target sequence that is

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specific to RNase P. During the extension phase of the PCR cycle, the 5′ nuclease activity of Taq polymerase degrades the probe. This degradation causes the reporter dye to separate from the quencher dye, which generates a fluorescent signal. With each cycle, additional reporter dye molecules are cleaved from their respective probes, which increases the fluorescence intensity. Fluorescence intensity is monitored at each PCR cycle by the real-time PCR instrument. The data are analyzed and interpreted using the analytical software associated with your real-time PCR instrument.[9]

RESULTS

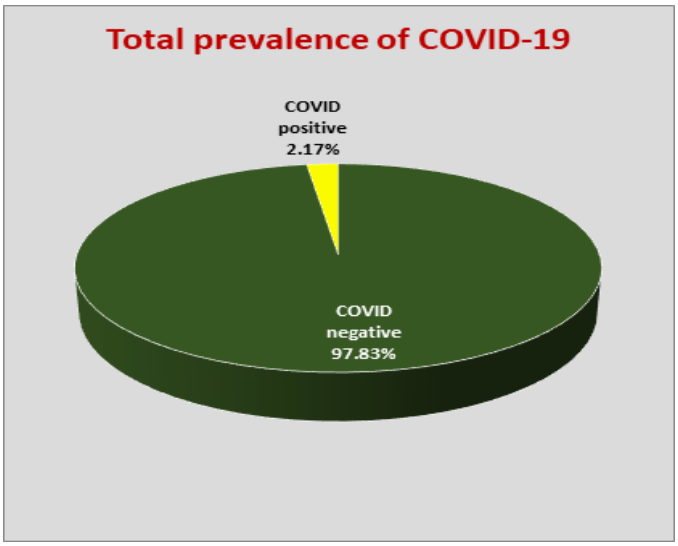

Figure 1: Total prevalence of COVID-19 in the study population.

During the present study, a total of 11,27,005 samples were analyzed from June 2021 to March 2022 for SARS-CoV-2 detection by ICMR approved COVID 19 RT-PCR kits. Out of total 11,27,005 cases which were included in the study, 24,466 cases (2.17%) were found to be SARS-CoV-2 positive while 11,02,539 cases (97.83%) were SARS-CoV-2 negative [Figure 1]. Samples were received from six districts of Punjab and of all the districts, Pathankot was having maximum COVID 19 positivity 7672/2,11,518 (3.63%) followed by other districts [Table 1].

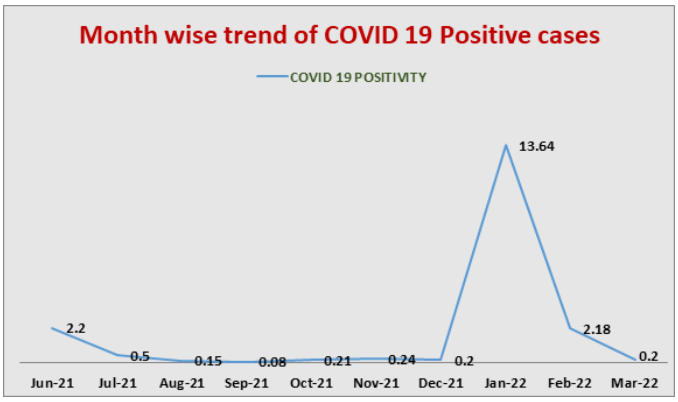

Figure 2: Month Wise Distribution Of Covid 19 Samples.

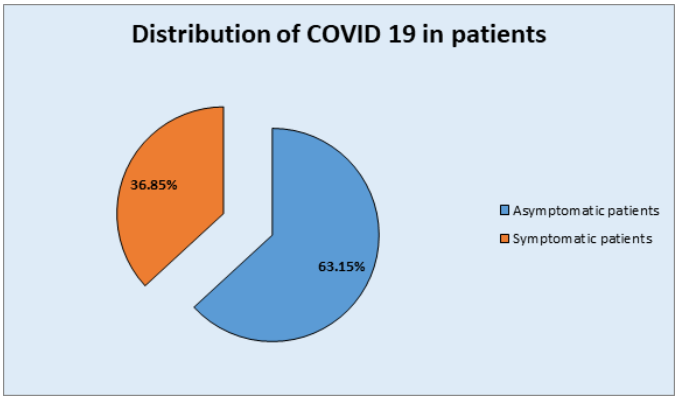

Figure 3: Distribution of COVID 19 in patients

SARS-CoV 2 positivity was seen mostly from the elderly group (>61 years) 3,095/56,348 (5.50%) followed by 21-40 years 12,679/5,07,159 (2.50%) as shown in Table 2. Out of 24,466 SARS-CoV 2 RT-PCR positive, 15,453 were males while 9,013 were females as in [Table 2].

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January 2022 month experienced maximum number of COVID 19 positive cases 17149/125713 (13.64%) and minimum cases were observed during the month of September 2021, 108/126591 (0.08%) [Figure 2].

The prevalence of COVID 19 positivity in symptomatic patients was found to be 9016/24,466 (36.85%) whereas in asymptomatic patients it is 15450/24466 (63.15%) [Figure 3].

Table 1: District wise distributution of COVID-19 patients.

Districts	COVID-19 Positive	COVID-19 Negative	Total Samples
Amritsar	9,349 (2.57%)	3,54,826 (97.43%)	3,64,175
Gurdaspur	5,870 (1.80%)	3,11,953 (98.20%)	3,17,823
Pathankot	7,672 (3.63%)	2,03,846 (96.37%)	2,11,518
Hoshiarpur	402 (0.65%)	61,724 (99.35%)	62,126
Tarntaran	1,169 (0.68%)	1,69,870 (99.32%)	1,71,039
Jalandhar	4 (1.23%)	320 (98.77%)	324
Total	24,466	11,02,539	11,27,005

Table 2: Age & Gender Wise Distribution Of Covid 19 Patients.

Variable	COVID-19 Positive	COVID-19 Negative	Total
Age			
0-20 years	5,871 (2%)	2,75,878 (98%)	2,81,749
21-40 years	12,679 (2.50%)	4,94,480 (97.50%)	5,07,159
41-60 years	2,821 (1%)	2,78,928 (99%)	2,81,749
≥61 years	3,095 (5.50%)	53,253 (94.50%)	56,348
TOTAL	24,466	11,02,539	11,27,005
Gender			
Male	15,453 (2%)	7,40,410 (98%)	7,55,863
Female	9,013 (2.43%)	3,62,129 (97.57%)	3,71,142
Total	24,466	11,02,539	11,27,005

DISCUSSION

The ongoing outbreak of SARS-CoV-2 infection has emphasized the significance of quick and precise laboratory diagnosis in order to limit the spread as well as help patients to prevent the illness progression. Diagnostic testing for COVID-19 is critical for understanding epidemiology, contract-tracing, case management, and to repress the transmission of the SARS-CoV-27. Currently, the Nucleic Acid Amplification Test (NAAT)-based RT-PCR technique is a gold standard test used for routine diagnosis of COVID-198 infection under Biosafety 2 level (BSL-2) conditions and with appropriate biosafety precautions.

A total of 11,27,005 samples were received in the lab from June 2021 till March 2022, out of which 24,466 (2.17%) were SARS CoV 2 positive 11,02,539 cases (97.83%) were SARS-CoV-2 negative by real-time PCR. Samples were received from six districts of Punjab and among all of the districts Pathankot was

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having maximum COVID 19 positivity 7672/2,11,518 (3.63%).

Age wise percent positivity in our study was 2% in age group of ≤20, 2.5% in 21-40 yrs, 1% in 41-60 yrs and 5.50% in ≥60 yrs. This is in concordance with the data obtained from CDC in 2021 as well as with the study conducted by Davies NG et al in 2020.[10,11] The age gradient in recent reported cases which depicted elderly people at a higher risk of COVID-19 infection is due to their decreased immunity as well as multiple associated co-morbidities. Children have decreased susceptibility to COVID infection which could be due to immune cross-protection from other coronaviruses or from non-specific protection resulting from recent infection by other respiratory viruses. The younger group individuals are also infected with COVID-19 which maybe because they are actively working and outdoor engagements were rampant, with little adherence to safety protocols, resulting in increased cases.[12]

In our study, SARS-CoV-2 positivity was seen more in males as compared to females. This finding is well supported by a study conducted by Chen et al in 2020 in Wuhan, China,[13] as well as by a study conducted by Tuli AK et al in 2021.[14] The rationale for this gender disparity can be attributed to number of factors such as social factors, genetic, immunological factors as well as hormonal difference. According to a recent study, the SARS-CoV-2 positivity variation among the gender could be due to mast cells from females which are able to initiate a more active immune response, which may help females fight off infectious diseases better than men. Recent evidence also indicates that mast cells are activated by SARS-CoV-2 which causes COVID-19.[15] Also, testosterone generally inhibits immune functions which is a possible explanation for men higher susceptibility to infections.[16] Males may be at increased risk for severe disease, because in general, they tend to smoke and drink more, wash their hands less frequently and often delay seeking medical attention.[15]

The maximum number of COVID 19 positive cases were reported in the month January, 2022 with minimum number in September, 2021 which is shown in [Figure 2]. The results were in concordance with the data received by WHO from national authorities.[17] Sudden upsurge in cases from December to March is mainly because of emergence of a new variant called 'Omicron'. Due to its high transmissibility, immune escape ability or both, it is becoming the dominant variant. Omicron (B.1.1.529), named by the WHO after the 15th letter of the Greek alphabet, has over 30 mutations on its spike protein, continues to fuel the third wave of Covid-19 pandemic in India. On Jan 20th, the country witnessed a whopping 3.47 lakh cases, the highest in the last eight months. Globally, India is the second worst-hit country after the US.[18] Indian Council of Medical Research (ICMR) has approved a made-in-India testing kit, named Omisure, for detecting the Omicron variant of the SARS-CoV-2 coronavirus which uses S-Gene Target Failure (SGTF) strategy.[19]

Out of the total 24,466 positive samples, 9,016 were symptomatic (36.85%) and 15,450 (63.15%) of patients were asymptomatic. In our study, asymptomatic individuals majorly comprises of the pandemic. This is in concordance with a study conducted by Shabir A. et al,[20] in 2022 in which it states that

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individuals tend to have mild disease in this wave as compared to previous variants which can be attributed to lower intrinsic virulence.[21] The Omicron variant of SARS-CoV-2 escapes immunity generated by vaccines and previous infections. Hence, infection with Omicron is considerably milder as compared to previous variants.[22]

CONCLUSIONS

Ever since the COVID-19 global pandemic emerged, the developing countries are facing challenges regarding its diagnosis. Early screening and diagnosis of SARS CoV-2 is the most important step ultimately leading to isolating the infected person which is necessary to prevent its further transmission. Isolation of the infected person will eventually decrease the Reproduction number i.e Ro which will further interrupt the transmission cycle leading to decrease in community spread. Also, with increase in diagnostic capacity of COVID-19, detecting more positive patients in the community will lead to the reduction of secondary cases.

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