

# EPIDEMIOLOGICAL, CLINICAL AND VIROLOGICAL CHARACTERISTICS OF SARS- COV 2 PATIENTS AT TERTIARY CARE HOSPITAL OF NORTH INDIA

Running title: Clinico-epidemiological profile of SARS- COV 2 patients.

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## Abstract

The pandemic of COVID-19 disease caused by SARS-CoV-2 continues to be the global public health concern. While nations are taking extensive measures to contain the pandemic, several variants have been identified more recently due to mutations of the SARS -CoV 2 which make it highly contagious and well known source of multiple waves. A retrospective and observational study were conducted on 28,897 nasopharyngeal/oropharyngeal swabs of patients consulted or hospitalized at tertiary care hospital. The Epidemiological, demographic & clinical details of all patients were recorded and the diagnosis of SARS- CoV 2 was done by real time reverse transcriptase reaction (RT-PCR). A total 4794(16.6 %) patients were tested positive for SARS CoV-2 & maximum cases (38.57%) were in age group 41-60 years ; males (51.48%) were found to be predominantly infected .In year 2020 maximum positivity of SARS -CoV 2 were reported in the months of August & September (14.72%) , then gradual decline from October to Feb 21 and then second wave lead to abrupt rise, 34.42% cases reported in the month of April- May 2021. More than 75% of positive patients were found to be symptomatic and the most frequent symptoms were fever (82.14%), cough (71.69%), and sore throat (43.5%). Severe acute respiratory illness (SARI) was detected in 38.06 % of positive cases & Influenza like illness (ILI) was reported in 11.28%. Fifty one (51) positive samples tested for whole genome sequencing, detected 56.8% B.1.1.7 lineage (alpha variant), (39.3%) B.1.617.2 lineage (delta variant) and 3.9% delta plus variants (A.Y 4 & A.Y 12). As the pandemic continues to unfold, there is need to analyse the epidemiological & clinical characteristics of the patients infected with SARS CoV 2 and identification of ongoing variants of concern in the presence of new selection pressures such as vaccination.

**Key words:** SARS- CoV 2, Real time RT-PCR, Variants of concern.

## Introduction

Acute Respiratory infections are a group of diseases that are caused by different microorganisms where viral etiologies are responsible for 80% of cases.[1] Most of the viral infections are associated with similar and atypical clinical presentations and infect both upper and lower respiratory tracts, which makes it very difficult for clinicians to distinguish the causative pathogen without a laboratory diagnosis. Rapid detection and identification of viral pathogens causing respiratory tract infections has been demonstrated to significantly decrease the duration of hospitalization and reduce management costs, and to avoid additional laboratory testing and unnecessary antibiotic use; additionally help to guide specific antiviral therapy and appropriate isolation precautions.[2]

Severe acute respiratory illness (SARI) caused by viruses is a major cause of death and morbidity in low and middle income countries, however, the etiologic agents are often undetermined due to the lack of molecular diagnostics in hospitals and clinics. [3] Surveillance for hospitalized patients with severe acute respiratory infections (SARIs) is an important public health tool used to identify aetiologies to understand the disease, track

changes in circulating viruses and as an alert mechanism for potential pandemic viruses. Molecular techniques have found their niches in laboratory diagnosis of viral respiratory tract infections by providing rapid and highly sensitive tools to detect and identify the causal pathogens. The use of molecular assay has notably contributed to identify pathogens possibly involved in severe respiratory disease, thus allowing getting to a diagnosis of viral infection in nearly 80% of study of SARI/ARDS cases. [4]

Some of the emerging viral pathogens had caused pandemics or fatal respiratory infections in the last two decades. SARS-CoV-2 is a novel coronavirus identified as a cause of COVID-19 that began in Wuhan China in late 2019 and spread extensively worldwide and has become a serious global public health crisis. The original SARS-CoV-2 strain from Wuhan (Wild type) was rapidly transmitted to a large number of countries through infected travellers followed by establishment of community transmission and further rapid spread. It has significant person to person transmission and cause acute, severe respiratory illness. Patients with SARS-CoV-2 infection may have mild to asymptomatic illness but some rapidly progress to severe acute respiratory distress syndrome (ARDS), multi organ dysfunction syndrome and death especially in elderly and with underlying comorbidities. [5]

Given the alarming levels of spread, severity of disease and a number of affected countries, the World Health Organization (WHO) declared COVID-19 as a pandemic on March 11<sup>th</sup>, 2020.[6] One of the most affected countries during the coronavirus disease (COVID-19) pandemic, caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), is India. COVID-19 was first detected in India on 30<sup>th</sup> January 2020, within few months, the number crossed thousands and then lakhs. Till September 22<sup>nd</sup> 2021, the country reports over 33 billion cases and more than 4 lakhs deaths. [7]

SARS-CoV-2 like all viruses accumulates mutations, changes in its genetic code over time as it replicates. The mutation in coronavirus genetic code is well known for source of multiple waves. Several variants have been identified more recently that appear to increase transmissibility and potential that have an impact on disease severity. With the alarming spike in the second wave of COVID-19 infection in India, the double mutant strain of virus presents a grave concern towards increased infectivity in India and hold the potency to overcome the immunity developed by natural infection or vaccines. [8]

As the pandemic continues to unfold, it is important to analyse the epidemiological & clinical characteristics of the patients infected with SARS-CoV-2 and identification of variants of concern (VOC) particularly in the presence of new selection pressures such as vaccination. Our Study aims to find out the rate of SARS-CoV-2 positivity among the patients consulted or admitted to the tertiary care hospital and to evaluate their epidemiological, demographic and clinical profile.

## Material and Methods

We conducted a retrospective and observational study from March 2020 to June 2021 at Viral Research and Diagnostic Laboratory, Govt. Medical College & Hospital Amritsar. A total of 28,897 Nasal/Nasopharyngeal/Oropharyngeal swabs of patients consulted or hospitalized at tertiary care hospital, were collected in viral transport media by following standardized procedures and general biosafety measures. [9] The Epidemiological, demographic & clinical details of all patients were recorded. Samples were labelled, sealed and while maintaining temperature at 4°C were transported in triple layer packaging to the Viral Research & Diagnostic Laboratory where the samples were handled in biosafety level-2 (BSL-2) facility by following WHO guidelines.[10] Viral RNA was extracted from 200µl of clinical sample by automated viral extraction techniques and the eluted RNA was used for amplification by real-time reverse transcriptase polymerase chain reaction (RT-PCR) assays (approved by ICMR) based on Taqman chemistry was performed on Applied Biosystem QuantStudio™ 5 Real-Time PCR instrument for the detection of SARS-CoV-2 virus. The results were analysed by reading the cycle threshold values and the graphs of amplification for E, N, RdRp & ORF 1b genes of SARS-CoV-2. Internal control will be added to each sample during nucleic acid extraction to validate the process of extraction and PCR technique used in the study. A total of 51 samples positive on RT-PCR with CT value <25 were sent to IGIB/NCDC, New Delhi for whole genome sequencing.

## Results:

A total of 28,897 samples were received at Viral Research & Diagnostic laboratory from March 2020 to June 2021. Among them, 4794(16.6 %) patients were tested positive for SARS-CoV-2 by real time-RT-PCR (Figure 1). Among the positive, maximum cases (38.57%) were in age group 41-60 years followed by 21-40 years (33.74%); males (51.48%) were found to be predominantly infected with Covid-19 as compared to females (Table 1). In year 2020 maximum positivity of SARS-CoV-2 cases were reported in the months of August & September (14.72%), then gradual decline in cases were seen from October to Feb 21 and then second wave led to abrupt rise 34.42% reported in the month of April- May 2021 (Fig.2)

A total of 3722( 77.63%) positive patients were found to be symptomatic and the most frequent symptoms were fever (82.14%), cough (71.69%), sore throat(43.5%), myalgia/arthralgia(43.5%), breathlessness(29.2%), Diarrhoea(6.74%), and anosmia(5.68%) .Patients hospitalized due to severe acute respiratory illness (SARI) was reported in 38.06 % cases & Influenza like illness (ILI) was detected in 11.28%. Among Health care workers, SARS CoV 2 was detected in 28.45% of cases. COVID - 19 positivity was reported 5.53 % of pregnant women and 0.18 % of international travellers. (Table 2). Fifty one (51) samples tested for whole genome sequencing at IGIB/NCDC New Delhi and detected 29(56.8%) B.1.1.7 lineage (alpha variant) and 20(39.3%) B.1.617.2 lineage (delta variant) & 02 (3.9%) delta plus variants(A.Y 4 & A.Y 12) reported.(Fig 3)

### Discussion:

We conducted a retrospective and observation study on 28,897 cases presented / admitted at tertiary care hospital, out of them, 4794(16.58%) cases were found be positive for SARS-CoV 2 by real time RT- PCR at Viral Research & diagnostic laboratory. The real time RT-PCR test is a delicate and valuable tool to identify novel coronaviruses and is considered as gold standard for detecting respiratory viruses. In present study, maximum cases (38. 57%) were reported in age group 41-60 years (mean age- 49.1 years) and 51.48 % among positive cases were males. Tambe et al screened total of 2154 subjects in their study, out of which 9.1% were confirmed as COVID 19 positive and maximum cases were in age group 31-60 years with mean age 45.8+17.3 years and also reported male preponderance with male: female ratio 1:1.2 .[11] Study on COVID 19 by Aggarwal et al reported medium age 54.5 years and 9.3% were males, with similar findings reported by Gupta et al , mean age 40 years with male predominance (58%)among SARS- CoV 2 cases. .[12] [13] The higher incidence in males reported in various studies could be more outside exposure, activities, travel and partly by the higher concentration of angiotensin converting enzyme 2 in males than in females. A series of 5700 COVID 19 cases from USA also revealed slight male preponderance and similar findings were also observed by Guan and colleagues. [14] .[15] Data from Chinese national reporting system, the median age of the confirmed cases was 51 years of which 77.8% were 30-69 years and total of 51.1% were males.[16]

Our findings showed, among the SARS CoV 2 positive,77.63% patients were found to be symptomatic and the most frequent symptoms were fever (82.14%), cough (71.69%), sore throat (43.5%) followed by myalgia/arthralgia (43.5%), breathlessness(29.2%),and Diarrhoea (6.74%). Another study found 73.5% of COVID 19 patients symptomatic with fever being the most common symptom (54.5%) followed by cough (45.5%), sore throat (33%) and myalgia (27%) and almost half of the patients(42%) had underlying comorbid conditions. .[13] Study done by Zayet et al reported more than 50 % cases symptomatic with most frequent symptom were fatigue, cough & fever followed by headache, myalgia & arthralgia . [17] However Tambe et al found most common symptom breathlessness (46.7 %), cough (43.5%)and then fever (42.6 %) <sup>11</sup>. Soni et al studied 42.1% patients symptomatic on admission and the common presenting complaints were fever (77.1%) and cough (54.2%).[18] In our study group, among the SARS CoV 2 positive cases, 38.06% were hospitalized with severe acute respiratory illness (SARI) & Influenza like illness(ILI) was detected in 11.28% of cases. Study done by Aggarwal et al on SARI, 39% patients were confirmed to be SARS- CoV 2 positive. <sup>11</sup>. Another study by ICMR team reported a total of 104 of the 5911 SARI patients were tested positive for COVID -19. [19]

We reported a total of 4794 SARS-CoV positive cases over the period of 16 months (March 2020 to June 2021), first case reported positive in March was international traveller. In 2020, Maximum positivity of SARS- CoV 2 cases were reported in the month of August & September (n=706, 14.72 %), then gradual decline were seen from October 2020 to February 2021 followed by abrupt rise from March 21 to May 2021 (n=2941, 18.2%). Scientists around the world have proposed numerous theories to explain unprecedented fall in India but there is no definitive and conclusive data to explain that. Cases have been consistently decreasing since October 2020 which may be due to the controlled transmission of SARS CoV 2 virus among the people because of their awareness of the safety measures, hygiene practices and following social distancing. According to The other reason for the downward trend can be attributed to localised herd immunity playing its role against the virus. Herd immunity develops when sufficient number of people have been infected with the virus within the community . [20] Data from India showed peak in the month of September 2020, 97,860 cases were recorded. Subsequently, the number of cases reduced considerably with lowest cases in the months of January and February. The second wave started from the middle of March 2021 with highest number of cases (414,188) recorded on 6th May 2021. Currently, India is experiencing significant drop in the number of active infections to 41,831 as on 31st July 2021.[21] India the first and second wave are separated by about 5 months. On April 15, 2021, the number of new cases was about 0.2 million which is already more than double of the first peak value. Prior to March 2021, less than 0.7% of the Indian population was infected with COVID-19. This current second wave took only 2 months to infect an additional ~0.36% of the population, and India is now recording over 0.4 million new cases per day (as of 23 April 2021). The true number is probably even higher, with some estimates putting the number of daily new cases at over 1 million, more than five times the officially recorded number.

[22] The sudden surge in the number of cases after a relatively long cooling time may be attributed to highly infectious double mutant strain of the virus, to negligent behaviour of the population and to the relaxation of interventions. [23]

A new lineage of the SARS CoV 2 virus linked with high infectivity and immune escape mutation has been identified in India and is believed to with the alarming spike in the second wave of COVID 19 infection, the double mutant strain of the virus presents a grave concern towards increased infectivity in India. The variant B.1.617 has been called double mutant

as it evolved through mutations in two separate sequences of the spike protein, namely E484K and L452R, both of which are located essentially on the antibody-recognizable site of the virus that might be responsible for reinfections with a breakthrough 16% confirmed cases of COVID-19 vaccinated health workers . [24] [25] The World Health Organization (WHO) has already recognized this exceptional transmissibility of this variant and declared it as a variant of global concern, which has already pervaded more than 60 countries. [25]

Among our study cases, 51 random positive samples (with Ct value <25) tested by whole genome sequencing, 29 (56.8%) showed B.1.1.7 lineage (alpha variant) and 20(39.3%) showed B.1.617.2 (delta variant) & 02 (3.9%) delta plus variants(A.Y 4 & A.Y 12) reported(Fig 3). Variants of concern in terms of higher transmissibility, virulence and impact of vaccines currently in use were identified in countries from different continents SARS Cov 2 lineage B.1.1.7 (alpha variant) was first detected in United Kingdom in Nov 2020. Subsequently other variants of concern (VOC) from South Africa (B.1.351- beta), Brazil (P.1& P.2.2 , gamma) and India (B.1.617- delta) is significantly more contagious than the wild type SARS CoV 2. [19]<sup>9</sup>

Many countries have seen a two wave pattern in reported cases of coronavirus disease and the empirical data show that the characteristics of the effects of the virus do vary between the two periods. Mutated strains are significantly more contagious than the variants in early 2020 however there is no clear evidence on the severity of new strains. [27] [27] . Mutations in the coronavirus genetic code is the well-known source of multiple waves as the virus gets time to mutate and find ways to evade immune system if vaccination is done slowly. Experts predict 3<sup>rd</sup> covid wave as every RNA virus has tendency to mutate and these mutations are inevitable.

More than one year since COVID 19 was declared a pandemic by world health organization the deadly SARS CoV 2 virus continues to disrupt public life across the world and the social life is still far from normal. Several countries are experiencing resurgence leading to second/third waves of the disease. Multiple vaccines have been approved in multiple countries however production and distribution at massive scale remain a formidable challenge. The global effect of the latest crisis on public health, social well-being and economic prosperity is still unknown. So the need of the hour is consistent monitoring of virus epidemiology and surveillance of variants of concern and the currently available vaccines efficacy against the new mutant strains.

## References

1. Amad MAA, Mahaqri AAA, Serouri AAA, Khader YS. Severe Acute Respiratory Infections with Influenza and Noninfluenza Respiratory viruses: Yemen, 2011- 2016.*The Journal of Health Care Organization, Provision and Financing*. 2019; 56:1-7.
2. Shu Zhang, Wenhong Zhang, Yi-Wei Tang. Molecular diagnosis of viral respiratory Infections. *Curr Infect Dis Res*.2010;13:149-158
3. Le YH, Nguyen KC, Coleman KK, Nguyen TT, Than ST, *et al*. Virus detections among patients with severe acute respiratory illness, Northern Vietnam. *Plos One*. 2020;15(5):1-10
4. Pariani E, Martinelli M, Canuti M, Farsani SMJ, Munnink BBO, *et al*. Influenza and other Respiratory Viruses involved in Severe Acute Respiratory Disease in Northern Italy during the Pandemic and Postpandemic Period (2009-2011). *Biomed Research International*. 2014; 1-5.
5. TaNU singhalA Review of Coronavirus Disease-2019 (COVID-19). *Indian J pediatr* .2020; 87(4) :281-286
6. World Health Organization. WHO Director- General's opening remarks at the media briefing on COVID-19. (2020). Available at: <https://www.who.int/director-general/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020> (Accessed: 21 September 2021)
7. WHO Coronavirus disease (COVID 19 pandemic. Available from <https://www.who.int/emergencies/diseases/novel-coronavirus-2019>. (Assessed on 22 September 2021)
8. Zeyaulah M, AlShahrani AM,Muzammil K, Ahmad I, Alam S,Khan WH and Ahmad R (2021)COVID-19 and SARS-CoV-2 Variants:Current Challenges and Health Concern. *Front. Genet.* 12:693916.doi:

- 10.3389/fgene.2021.693916.
9. Ruce J, Garcia K, Tran T, Papadakis G, Birch C. Evaluation of Swabs, Transport Media, and Specimen Transport Conditions for Optimal Detection of Viruses by PCR. *J Clin Microbiol.* 2012 Mar 1; 50(3):1064–5.
  10. Chan PKS, To W-K, Ng K-C, Lam RKY, Ng T-K, Chan RCW, et al. Laboratory Diagnosis of SARS. *Emerg Infect Dis.* 2004 May;10(5):825–31.
  11. Tambe MP, Parande MA, Tapare VS, Borle PS, Ladke NJ et al. An epidemiological study of laboratory confirmed of covid 19 cases admitted in a tertiary care hospital of Pune, Maharashtra, India. *Indian Journal public health* 2020; 64:S183-7.
  12. Aggarwal A, Shrivastava A, Kumar A, Ali A. Clinical and epidemiological features of SARS CoV 2 patients in SARI ward of a tertiary care centre in New Delhi. *Journal of the association of physicians of India* 2020 ;68:19-26.
  13. Gupta N, Ish P, Kumar R, Dev N, Yadav SR et al. Evaluation of the clinical profile, laboratory parameters and outcome of 200 Covid 19 patients from a tertiary care centre in India. *Monaldi archives for chest disease* 2020 ;90:1507.
  14. Richardson S, Hires JS, Narasimhan M et al. Presenting characteristics, comorbidities and outcomes among 5700 patients hospitalised with covid 19 in the New York city area. *JAMA* 2020;323:2052-9
  15. Guan WJ, NiZY, Hu Y et al. Clinical characteristics of coronavirus disease 2019 in China, *N Eng J Med.* 20;382:1908-2020
  16. Su Eun Park. Epidemiology, virology and clinical features of SARS CoV 2; coronavirus disease 19. *CEP* 2020;63(4)119-124.
  17. Zayet S, Kadiane- Oussou NJ, Leppiler Q, Zahra H, Royer PY et al. Clinical features of covid 19 and influenza: a comparative study on Nord Franche Comte cluster. *Microbes and infection* . <https://doi.org/10.1016/j.micinf.2020.05.016>.
  18. Soni SL, Kajal K, Yaddanapuddi LN, Malhotra P, Puri GD et al. Demographic & clinical profile of patients with covid 19 at a tertiary care hospital in North India. *Ind J Med Res* Nov 2020;IP251,143,47.5.
  19. Gupta N, Prahraj I, Bhatnagar T, Thangaraj JWB, Giri S, Chauhan H et al. Severe acute respiratory illness surveillance for coronavirus disease 2019, India, *Ind J Med Res* 151(2): 236040.
  20. Omer SB, Yildirim I, Forman HP. Herd Immunity and Implications for SARS-CoV-2 Control. *JAMA.* 2020 Nov 24;324 (20):2095.
  21. Shrivastava s, Mahaske ST, Modak MS, Virkar RG, Pisal SS et al. Emergence of Two Distinct Variants of SARS-CoV-2 and Explosive Second Wave of COVID-19: An Experience From A Tertiary Care Hospital, Pune, India. *Archives of virology* DOI: [tps://doi.org/10.21203/rs.3.rs-824960/v1](https://doi.org/10.21203/rs.3.rs-824960/v1).
  22. Singh J, Rahman SA, Ehtesham NZ, Hira S, Hasnain SE. SARS CoV 2 variants of concern are emerging in india. 2021. *Nature Medicine* 27: 1131-33.
  23. Xu S, Li Y (2020) Beware of the second wave of covid-19. *The Lancet* 395(10233):1321{1322.
  24. Thiagarajan K. Why is India having a COVID-19 surge? *BMJ* 2021;373:n1124. doi:10.1136/bmj.n1124.
  25. Dyer O. COVID-19: India sees record deaths as "black fungus" spreads fear. *BMJ* 2021; 373:n1238. doi: 10.1136/bmj.n1238.
  26. Chen J, Wang R, Wang M, Wei GW (2020) Mutations strengthened sars-cov-2 infectivity. *Journal of molecular biology* 432(19):5212-226.
  27. Davies NG, Abbott S, Barnard RC, Jarvis CI, Kucharski AJ, Munday JD, Pearson CA, Russell TW, Tully DC, Washburne AD, et al (2021) Estimated transmissibility and impact of sars-cov-2 lineage b. 1.1. 7 in England. *Science* 372:6538

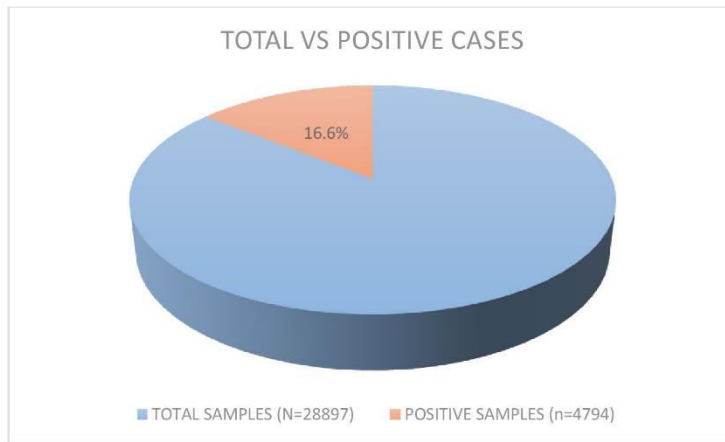
**Table 1. Age & Gender distribution of SARS-CoV-2 positive Cases (n=4794)**

Variable	Number	Percentage (%)
<b>AGE RANGE ( IN YEARS)</b>		
<20	204	4.25
21-40	1603	33.44
41-60	1849	38.57
>60	1138	23.74

<b>GENDER</b>		
<b>MALE</b>	2468	51.48
<b>FEMALE</b>	2326	48.52

**Table 2. Category wise distribution of SARS - CoV 2 positive cases ( n=4794)**

<b>Categories</b>	<b>Number</b>	<b>Percentage (%)</b>
Hospitalized SARI (Severe Acute Respiratory Illness ) cases	1825	38.07
Symptomatic Influenza like illness( ILI ) cases	541	11.28
Symptomatic Healthcare / Frontline workers	1323	27.59
Asymptomatic Healthcare / Frontline workers	41	0.86
Symptomatic direct & high risk Contact of lab confirmed case	33	0.69
Asymptomatic direct and high risk contact of lab confirmed case/ high risk individuals	123	2.56
International Travellers	09	0.18
Pregnant women	265	5.53
Others	634	13.22



**Figure 1. Prevalence of SARS CoV 2 Cases**

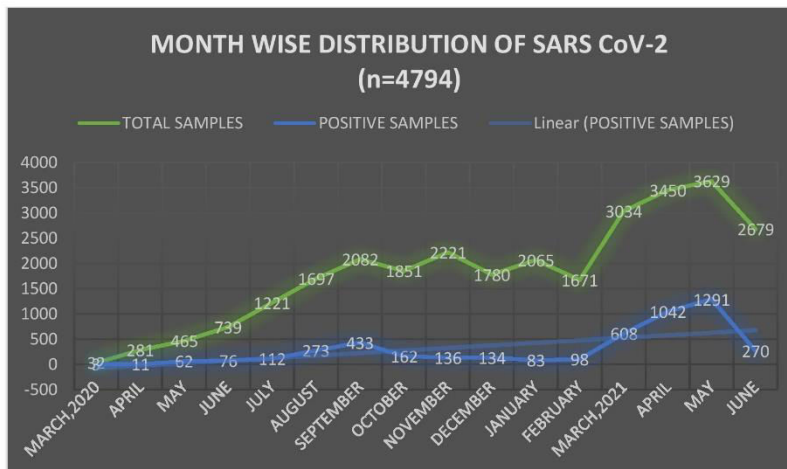
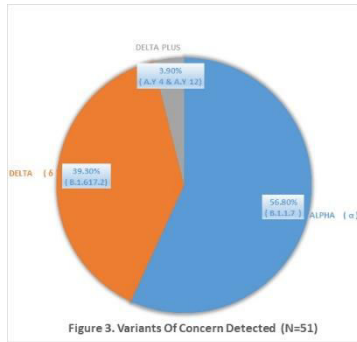


Figure 2. Month wise distribution of Cases





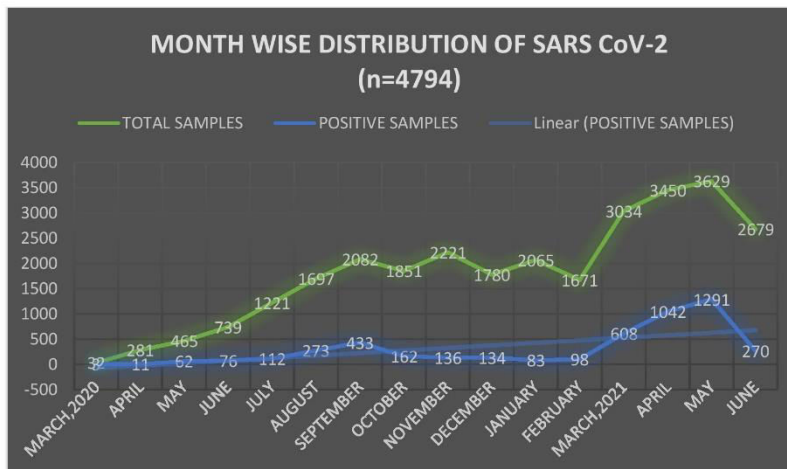


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