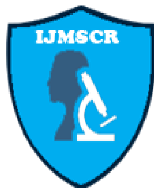


(International Print/Online Journal)

SJIF IMPACT FACTOR: 5.565
PUBMED-National Library of Medicine ID-101739732

ISSN (Print): 2209-2870
ISSN (Online): 2209-2862

International Journal of Medical Science and Current Research (IJMSCR)
Available online at: www.ijmscr.com
Volume 5, Issue 1, Page No: 863-867
January-February 2022

IJMSCR

Comparative Evaluation of Truenat Screening And Confirmatory Assays With RT-PCR For Diagnosis of SARS CoV2 infections

[1]Dr. Kala Yadhav ML, [2]Dr. Sandhya Narayanan, [3]Dr. Sunitha BU, [4]Dr. Chetana GS, [5]Dr. Deepa MC, [6]Dr. Ranjani R
MBBS, [1]MD, Professor & HOD, [2]MD, Senior Resident, [3]MD, Associate Professor, [4]MD, Assistant Professor, [5]MBBS PG Student, [6]D Bac, Tutor
[1,2,3,4,6]SABVMCRI, Bengaluru
[5]BMCMCRI, Bengaluru

***Corresponding Author:**
Dr. Sandhya Narayanan
MD, Senior Resident, SABVMCRI, Bengaluru

Type of Publication: Original Research Paper
Conflicts of Interest: Nil

Abstract

ICMR recommends Truenat testing as a comprehensive assay for screening and confirmation of SARSCoV-2 cases. This study was done to do comparative evaluation of Truenat screening and confirmatory assays with RTPCR testing for the diagnosis of SARSCoV-2 infections. Accurate diagnosis of SARSCoV-2 is very important for early identification, isolation and for tracing the patient contacts. This comparative study of Truenat screening & confirmatory assays with RTPCR was conducted from June 2021 to November 2021. Out of 1026 samples, 221 (21.5%) of them tested positiveOverall, the E gene was found to be positive in 21.5% (n=221) and 21.7% (n=223) using both Truenat and RTPCR tests. Rdrp gene was found to be positive in 21.5% (n=221) using both Truenat and RTPCR tests. Sensitivity and specificity of Truenat Beta Cov (E gene detection) testing was found to be 100% and 99.7% respectively. Whereas Truenat SARS-Cov-2 (Rdrp gene) testing was found to be 100% sensitive and specific. RTPCR is the gold standard method used for the detection of SARS COV 2 infection. Truenat screening test and confirmatory tests are highly recommended testing methods during the pandemic owing to rapid detection the results being available in a very short period.

Keywords: SARS COV-2, Rdrp gene, Truenat, RTPCR, Comparative study

Introduction

COVID-19 is the disease caused by a new coronavirus called SARS-CoV-2. The first case was reported in December 2019, following a report of a cluster of 'viral pneumonia' cases in Wuhan, People's Republic of China. These Coronaviruses belong to a subfamily of Orthocoronavirinae of the family of Coronaviridae. They are enveloped viruses with positive- stranded RNA.1

A combination of different tests and testing platforms has been used to augment capacity to 1·2 million trials per day, as of Sept 25, 2020.Indigenous portable Truelab (Molbio Diagnostics, India) workstations, previously used and recommended by WHO for tuberculosis and also deployed for detection of Nipah virus disease (unpublished) and leptospirosis (unpublished), are now being used for detection of SARS-CoV-2 for faster resuts.2

The Truenat Beta CoV E-gene screening assay and Truenat SARSCoV-2 RdRp gene- confirmatory assay (Molbio Diagnostics, India) were earlier validated as a two-step test.The assays were deployed for COVID-19 testing in various parts of India between April and June 2020.3A multiplex assay combining E-gene screening and Orf1a-gene confirmatory assay has also been validated recently.

The extraction of RNA using Trueprep takes 20 min, and each of the assays requires 45 minutes. This is quicker than RT-PCR, which takes around 4-6 h for the entire process. Therefore, these assays would be valuable in rapidly confirming COVID-19 cases in field settings.

Truenat assays were initially designed for testing TB samples; in the advent of ongoing Covid pandemic, Truenat assays have been modified to test Covid models, and hence this study is undertaken to evaluate the efficiency of Truenat assays as a source of rapid point of care diagnosis, in the event of an ongoing pandemic. It has various advantages like it needs fewer consumables s and also, these assays do not require technically trained personnel to work compared to RTPCR.

Material And Methods:

This study was undertaken at the Department of Microbiology, RTPCR referral lab of a tertiary care hospital, Bangalore. This study includes a total of 1026 samples received in our lab sent for covid testing for a period of 6 months, from June 2021 to November 2021. The samples were selected for this study following ICMR guidelines. Therefore, spilled samples, mismatched label samples, inadequate samples were excluded from the study.

Truenat Beta CoV works on the principle of RTPCR which is based on Taqman chemistry. RNA extraction was done using Trueprep Auto/Auto v2 universal cartridge-based sample prep device and prep kit. Truelab real time micro-PCR analyser uses a chip to analyse the sample. 6µl of the extracted RNA is mixed with the microtube containing freeze dried PCR reagents. 6µl of this clear solution is then added to the reaction well of the Truenat Beta Cov chip which is the tested using Trurlab Real time micro-PCR analyser. At the end of the test, results will be displayed as detected for positive and not detected for negative samples.

This comparative study compares the efficiency of Truenat Beta Cov and Truenat Sars Cov with RTPCR for covid positive samples. Tests were done as per the kit manufacturer's literature.

Results

This study was undertaken at the department of Microbiology, RTPCR lab, Bowring and Lady Curzon Medical college and Research Institute, Bangalore from June 2021 to November 2021. Institutional Ethics Committee (IEC NO: BLCMCRI/ IEC/ RP/ 033/2021- 22 dated 8.04.2021) approval was taken before this study.

A total of 1026 patients were included in the study. There was a total of 609 (59.3%) males and 417 (40.6%) females in this study. All the patients were divided into 0-20, 21-40, 41-60 and >61 years of age group. 426 patients were in 21-40 years of age group followed by 411 patients were in 41-60 years of age group, 124 patients were in > 61 years age group, and 62 patients were in the age group of 0-20 years. (Table 1)

Out of 221 positive patients, 46.2% (n=102) were asymptomatic followed by 42% n (n=93) were symptomatic, 4.9% (n=11) were contacts with positive patients, and 3.6% (n=8) of all the positive patients were follow up positives. (Graph 1)

Among total of 1026 patients tested, 221(21.5%) of them tested positive. The data from the study shows that there are more E gene-positive patients who belong to the age group of 21 - 40 years (n=125) followed by 60 patients in the age group 41 – 60 years, followed by 2 patients in the age group > 60 years and 15 in the age group 0-20 years. (Graph 2)

Among all the E gene positives detected by Truenat 4.5% (n=10) was very low detected followed by 10.3% (n=23) low detected, 39.4% (n=88) medium detected and 45.7% (n=102) high detected. (Table 2)

Overall, the E gene was found to be positive in 21.5% (n=221) and 21.7% (n=223) using both Truenat and RTPCR tests. Rdrp gene was found to be positive in 21.5% (n=221) using both Truenat and RTPCR tests. Considering RTPCR as gold standard, two false positives were detected for E gene using Truenat. 807 and 805 patients were found to be negative using

Truenat and RTPCR tests respectively. Considering RTPCR as gold standard, two false negatives were reported in Truenat. Sensitivity and specificity of Truenat Beta Cov (E gene detection) testing was found to be 100% and 99.7% respectively. Whereas Truenat SARS- Cov-2 (Rdrp gene) testing was found to be 100% sensitive and specific. (Table 3)

© 2022 IJMSCR. All Rights Reserved

Discussion

ICMR recommends Truenat testing as a comprehensive assay for screening and confirmation of SARSCoV-2 cases. Sample is collected in the viral buffer solution which minimizes the biosafety and biosecurity requirements. Singleplex Truenat assay has two steps, step1 includes screening test which detects E gene. All negatives are to be considered as true negatives. All positive samples should be subjected to confirmation by step 2 assay. Step 2 assay has RdRp gene confirmatory assay. All samples that test positive by this assay is considered as true positives.

This study was done to do comparative evaluation of Truenat screening and confirmatory assays with RTPCR testing for the diagnosis of SARSCoV-2 infections. Accurate diagnosis of SARSCoV-2 is very important for early identification, isolation and for tracing the patient contacts.

In our study out of a total of 1026 patients, males were 59.3% compared to females which was 40.6%. This finding is similar to a study by Sodani et.al showed 60.4% males out of 1000 total samples.4 Another study by Qun Li et.al showed that 56% were males out of total of 425 patients.5 Another study by Huang C et.al showed 73% males in their study which is correlating with our study.6 Reason for male predominance could be because of increased testing among males compared to females.

This study included 53.8% of symptomatic patients. Since we have included data from ICMR portal directly, we are not certain if we have missed out real data. Proper history taking will help us to avoid missing out on categories of patients.

In our study, 21-60 age group was the most affected age group. Finding is similar to a study done by Priya et.al shows highest affected age group was between 50-69 years.7 Our study showed maximum positives in symptomatic group which is 53.8%. In a study done by ICMR, positives were observed more in symptomatic group.7

E gene positivity of this study was 21.5% (n=221) and 21.7% (n=223) using RTPCR and Truenat respectively. 21.5% (n=221) were tested positive using Truenat SARS CoV-2 RdRp confirmatory gene. We observed 100% sensitivity and 99.7% specificity in this study. In another study, 75 samples (30 positives and 45 negatives) were tested using Truenat and reported sensitivity and specificity to be 100%.8

To our knowledge, this is the first study which was conducted in Bangalore to know the performance of Truenat testing. RTPCR is the gold standard method used for the detection of SARS COV 2 infection. Truenat screening test and confirmatory tests are highly recommended testing methods during the pandemic owing to rapid detection the results being available in a very short period. Further, the Truenat testing method does not require skilled person to perform testing which is a major advantage over RTPCR testing.

Conclusion

To our knowledge, this is the first study which was conducted in Bangalore to know the performance of Truenat testing. RTPCR is the gold standard method used for the detection of SARS COV 2 infection. Truenat screening test and confirmatory tests are highly recommended testing methods during the pandemic owing to rapid detection the results being available in a very short period. Further, the Truenat testing method does not require skilled person to perform testing which is a major advantage over RTPCR testing.

Limitations Of Study

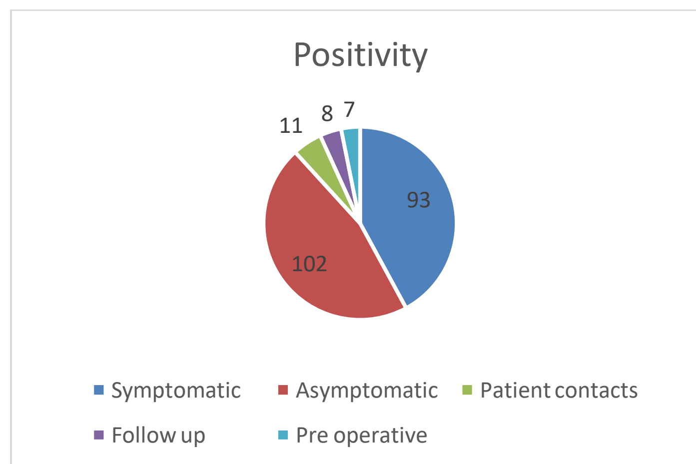
This study has some limitations, First, patient history was taken from the ICMR portal, no direct interaction or diagnosis has been possible with patients who have given their samples owing to the same. Second, Truenat SARS-COV-2 detects only Rdrp confirmatory genes which increases the chance of missing out other confirmatory genes. Epidemiology history was taken in to consideration to increase the sensitivity for early detection of the cases.

© 2022 IJMSCR. All Rights Reserved

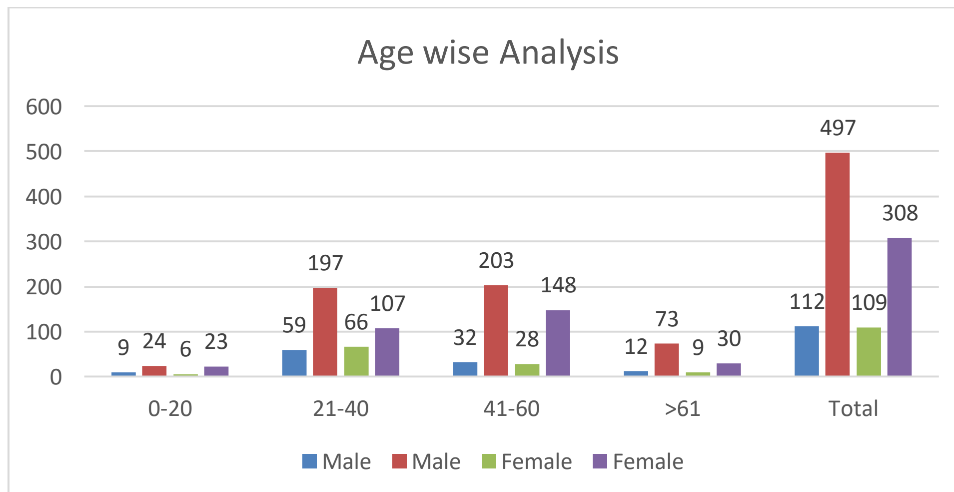
Table1. showing patient demographic data

AGE (years)	Male	Female	Total
0-20	33	29	62
21-40	256	173	429
41-60	235	176	411
>61	85	39	124
Total	609	417	1026

Graph 1. showing different categories of positive patients



Graph 2. showing age wise distribution of E gene



© 2022 IJMSCR. All Rights Reserved

Table 2. showing level of detection of E gene and Rdrp in Truenat

Levels	E gene	Rdrp
Very low detected	10	9
Low detected	23	22
Medium detected	88	88
High detected	102	102

Table 3. showing E gene comparison using Truenat and RTPCR

E gene detection tests	Positives	Negatives
Truenat	223	803
RTPCR	221	805

Volume 5, Issue 1; January-February 2022; Page No 863-867

© 2022 IJMSCR. All Rights Reserved