DST institute develops new multiplex RT-PCR kit with novel gene targets to facilitate detection across various mutant strains of COVID-19

A newly developed multiplex RT-PCR kit has a higher accuracy of detecting covid19 across the various mutant strains of the virus responsible for the global pandemic.

As the pandemic is going through a second wave with multiple variants, the selection of target genes in multiplex RT-PCR assay is becoming critical for accurate detection of the virus.

Even though coronaviruses make far fewer errors than other RNA viruses, the mutations in S, R, and N genes often interfere with RT-PCR assay. For example, the “variant of concern” B1.1.7 (also known as the UK variant) has a 69-70del, due to deletion of 6 bases in the RNA, which resulted in S gene drop out from RT-PCR assay.

The new multiplex RT-PCR kit developed Sree Chitra Tirunal Institute for Medical Sciences and Technology (SCTIMST), an Institute of National Importance under the Department of Science and Technology, Government of India targets two SARS CoV2 genes: RdRp and ORFb-nsp14, and the human RNAse P gene as the internal control to help detect a range of mutant strains.

Various studies have shown that RdRp and ORF1b-nsp14 genes are more sensitive in detecting Covid19. In order to target the multiple variants in the second wave, using two highly accurate confirmatory genes like RdRp and ORF-nsp14, can give precise results. The ORFb-nsp14 is one of the least mutated genes in Covid19 and currently, there are no kits in the market with ORF-nsp14 as the target.

The new kit is based on multiplex Taqman chemistry, amplifying all three genes in a single reaction. The amplification time for the assay is 45 minutes, apart from the time required for the RNA isolation from nasopharyngeal swab samples. Multiplexing two confirmatory genes will help shortlist possible new variants if one of the genes fails to amplify and can be marked for sequence analysis.

ICMR has validated this kit at the National Institute of Virology, Pune, and found that it has 97.3% sensitivity and 100% specificity in covid19 detection.

SCTIMST has signed a non-exclusive license MoU with Huwel Lifesciences, Hyderabad, on 14th May 2021 to commercialize the kit.

“This unique RT-PCR kit will be a significant weapon in our fight against COVID-19 by a facile detection of SARS-CoV-2 mutations which are becoming increasingly important,” said Prof Ashutosh Sharma, Secretary, DST.