ACTA SCIENTIFIC MICROBIOLOGY (ISSN: 2581-3226)

Volume 4 Issue 2 February 2021

Short Communication

Ensuring Quality of Covid-19 Testing in a State of Emergency

Ashok Rattan*

Professor, Advisor, Pathkind Labs, India

***Corresponding Author:** Ashok Rattan, Professor, Advisor, Pathkind Labs, India.

DOI: 10.31080/ASMI.2020.04.0763

Received: November 26, 2020

Published: January 22, 2021

The high infectivity of Severe Acute Corona Virus 2 (SARS CoV 2) made Tedros Adhanom Ghebreyesus of World Health Organization (WHO) urge all member states to "Test, Test, Test". Indian Council of Medical Research (ICMR) was charged with the responsibility of stewardship of 5 T approach, "Test, Track, Trace, Treat and Technology". ICMR has successfully ramped up testing facilities to have 1122 government facilities and 867 private National accreditation board for testing and calibration laboratories (NABL) accredited laboratories conduct diagnostic tests for Covid-19 and more than 10 crore samples have been tested. Earlier RT PCR kits were obtained from international In vitro devices (IVD) companies but subsequently many of the RT PCR and Rapid Antigen tests (RAT) and more recently RT LAMP tests have been manufactured by Indian companies and made available for clinical use.

Registration of IVD is complex and expensive, but has been put in place to minimise the risk of poorly performing tests. IVDs for HIV and hepatitis and all test kits to screen the blood supply are highly regulated, as a false result can cause harm to both the individual and the community.

Sensitivity and specificity are important performance characteristics for almost all test kits. To estimate these characteristics, access to large numbers of known positive and negative samples is required. The European Common Technical Specifications currently require about 500 positive samples and 5,000 negative samples to assess specificity and sensitivity for blood screening assays. For Rapid Diagnostic Tests (RDT) for HIV, HCV and HBsAg, 500 positive and 1,500 negative samples are required; with an expected sensitivity of > 99.0%. Samples obtained from blood donors, clinical samples, pregnant women and samples with potentially interfering substances need to be sourced and tested. NAT for HIV RNA, HCV RNA and HBV DNA need to have the LOD estimated, as well as the LOQ for quantitative assays, by testing against an international standard. At least 10 samples for each HIV genotype must be tested to demonstrate the ability to detect each genotype equally. WHO Prequalification protocols have similar but often less stringent criteria. By the time an IVD is released to the market, extensive performance evaluations have been conducted and are in the public domain for potential users to assess.

In the face of global spread of SARS CoV 2 and the availability of many RT PCR kits for its detection, regulators waived the regulatory requirements to allow use of kits without the complete manufacturer evidence normally required. WHO implemented an Emergency Use Listing Procedure for COVID-19 NAT assays (but not serology), requiring limited evidence of performance. Similarly USA FDA; Canada; Japan; Korea and Singapore have also implemented Emergency Use Listing for NAT and serology without requiring complete performance evidence. India initially requested NIV for validation of COVID-19 but all EU or USA FDA approved tests were expediously registered.

Many of the testing laboratories had little experience of RT PCR much less in house validation of approved kits and at the same time have been overburdened by the heavy sample load with even courts having a say on turn around time of reports. No approved EQAS scheme was initially available.

There are some technical difficulties faced when designing evaluation protocols for COVID-19 tests. Unlike HIV, syphilis and

HBsAg serology tests, there are no acknowledged reference or confirmatory methods. In HIV serology, a testing strategy using multiple tests including screening tests followed by supplemental and/or confirmatory tests such as western blots are used to confirm positivity. Similarly syphilis testing used multiple specific anti-treponemal tests such as EIAs, CHLIA and TPPA in the testing strategy. HBsAg and HIV p24 positivity can be confirmed using neutralisation testing. Unfortunately none of this has been done with tests for Covid19 diagnosis. Without this level of discipline, our understanding of the performance of COVID-19 test kits will remain limited.

In India over 9 million confirmed cases have been reported of which 134,218 could not survive. With availability of ample RT PCR kits, the focus has been to make the test available at lower price rather than focusing on quality of the results and reliability of decision making. With availability of Rapid Antigen Test, which in two studies had been shown to be specific but of low sensitivity and since the results were available in 30 minutes, RAT has been the test of choice in many situations, thus missing a large number of real cases as the initial advisory of performing RT PCR on all symptomatic patients who test RAT negative, was not implemented.

There are multiple reasons to explain the wild fire like spread of this infection, but the pandemic has brought forth the importance of rapid and definitive diagnosis to improve containment and treatment of Covid-19 infection. It also provides opportunities to improve the reliability of laboratory testing, some of the measures could be:

- Since all private labs which have been allowed to test for Covid 19 are accredited by NABL, all government labs too should be encouraged to follow the same discipline.
- All staff should be continuously trained and have their competency assessed in their ability to discharge the duties assigned to them.
- All testing should be performed as per approved SOPs.
- All samples must have positive control (with known viral genome load) and negative control run on each of the 96 well RT PCR plate.
- Results should be reported only when both positive and negative controls perform satisfactorily.
- The CT value of positive control should be plotted on an L J chart and used for monitoring the day to day or inter run variation.
- Delta CT should be reported. Delta CT is obtained by subtracting the CT value of the clinical sample from the positive control. Since the number of viral genomes present in positive control are known, the viral genomes in the clinical sample can be derived and an estimation of viral load provided to the clinician.
- All labs should participate in EQAS run by an accredited PT supplier.

WHO and ICMR has recently drawn up essential diagnostics list. ICMR should identify the high impact diseases which these newly upgraded molecular biology laboratories would be able to perform once Covid-19 subsides. Great help in rapid diagnosis of tuberculosis and tropical fever can be anticipated.