Comparative Study of the Efficacy of the Abbott ID Now Rapid Assay with Real Time PCR for the Diagnosis of SARS-CoV-2 RNA

Pratiksha Chheda

Sir HN Reliance Foundation Hospital and Research Centre

Dama Tavisha

Sir HN Reliance Foundation Hospital and Research Centre

Bhalerao Rahul

Sir HN Reliance Foundation Hospital and Research Centre

Bagwan Jamir

Sir HN Reliance Foundation Hospital and Research Centre

Bhat Devdatta

Sir HN Reliance Foundation Hospital and Research Centre

Shashikala Shivaprakash (✉ Shashikala.Shivaprakash@rfhospital.org)

Sir HN Reliance Foundation Hospital and Research Centre <https://orcid.org/0000-0003-2370-6244>

Short Report

Keywords: SARS-CoV-2, Emergency Use Authorization, Point of care test

Posted Date: August 2nd, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-731426/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Abstract

Rapid diagnostic tests are of great importance in hospital settings during the current outbreak of SARS-CoV-2. The clinical patient management and spread of infection is critically dependent on molecular assays with shortest possible turn-around time. Here we report performance of a point of care Abbott ID NOW COVID-19 assay in comparison to routinely used real-time RT-PCR assay on 205 clinical specimens.

Overall agreement of ID NOW was found to be 93.7% with positive percent agreement (PPA) of 91.8% and negative percent agreement (NPA) of 95.4%. Based on our findings, low turnaround time, minimal infrastructure need and ease of performing the assay, Abbott ID NOW COVID-19 assay can be considered as a point of care test in hospital settings.

Introduction:

Severe acute respiratory virus coronavirus 2 (SARS-CoV-2) that had emerged in Wuhan, China in December 2019 has penetrated world over [1]. This has led to global pandemic of coronavirus disease (COVID-19) with India being the second most affected nations owing to its dense population. Public health efforts to contain the infection was mediated with accelerated large-scale testing by reference laboratories and healthcare centres across the globe. Commercial kit manufacturers and FDA's emergency use authorization (EUA) process ramped up availability of reliable testing kits. Several rapid assays such as ID NOW COVID-19 (Abbott), Xpert Xpress SARS-CoV-2 (Cepheid) and Simplexa™ COVID-19 Direct (Diasorin) offers detection of SARS-CoV-2 RNA in 15 minutes to 90 minutes directly from dry nasopharyngeal swabs or swabs transported in viral transport media (VTM). EUA for diagnostic kits is obtained by minimal test validation and hence it is the responsibility of a molecular diagnostic laboratory to carry out in-house verification of the assay prior to clinical use. The aim of our study was to compare the Abbott ID NOW COVID-19 point of care test (POCT) with Real Time RT-PCR-based methods to assess its efficacy for patient testing.

Materials And Methods:

The performance of ID NOW was evaluated on a set of 205 specimens at the molecular diagnostics laboratory of our hospital. and the results were compared against Real Time based RT-PCR assay from SD Biosensor. The specimens were collected from 114 individuals presenting in the outpatient department (OPD cases) who had signs and symptoms indicative of COVID or had history of contact with COVID-19 positive patient, 57 were from a population of positive patients from COVID ward and 34 were from other hospitalized patients (IPD cases) from non covid ward. The study was conducted as a part of instrument validation and the data obtained was retrospectively analysed. Hence informed consent was not obtained from the patients.

Two nasopharyngeal swabs (NP) were collected in parallel from each of the subjects for both ID Now and RT-PCR assay. For ID Now assay dry swabs were transported in 15 ml sterile tubes to molecular

diagnostics department within the hospital. These swabs were stored at 4°C and tested on ID NOW within 2 hours of collection, consistent with the manufacturer's instructions. For RT-PCR analysis the swabs were transported in viral transport medium (Himedia; VTM) and were analysed within 24 hours of collection.

ID Now COVID-19 (Abbott Diagnostics) is an automated assay based on isothermal nucleic acid amplification technology that targets RdRp region of the genome. It is a rapid assay that utilizes dry swabs and gives qualitative results within 5 to 13 minutes. The assay method comprises of insertion of Orange Test Base (containing sealed lyophilized reaction) into the orange test base holder, followed by placing the Blue Sample Receiver (containing elution/ lysis buffer) into the corresponding Blue Sample Receiver holder. The dry NP swab was vigorously mixed in the sample receiver buffer for 10 seconds. The white transfer cartridge is pressed into the sample receiver, lifted and connected to the Test Base. ID NOW contains an internal control that has been designed to control for sample inhibition, amplification, and assay reagent function.

StandardM nCoV Real-Time Detection (SD BIOSENSOR) target regions of envelope (E) and RNA dependent RNA polymerase (RdRp). The RNA was extracted using 200 µl of VTM by Qiasymphony DSP Virus Pathogen mini kit (Qiagen GmbH, Germany) as per the manufacturer's protocol and 10 µl of eluted RNA was used for RT-PCR reaction. Exogenous internal control provided in the StandardM nCoV kit was spiked in the specimen during extraction. The Real Time PCR was carried out in Rotor-Gene Q platform (Qiagen Inc.) and ORF1ab (RdRp), E Gene and Internal control targets were detected in FAM, HEX and Cy5 channels respectively. The work was carried out inside a Class II Biological Safety Cabinet (BSC) by a certified laboratory personnel.

Assuming RT-PCR to be the reference method, we calculated the positive percent agreement (PPA) and negative percent agreement (NPA) of rapid assay in our study population.

Results:

In total 205 samples were tested prospectively by Abbott ID Now as well as by StandardM nCoV Real-Time PCR (reference method). SARS-CoV-2 RNA was detected in 97 (47.3%) and 94 (45.8%) cases by RT-PCR and POCT assays respectively. The overall agreement between the two assays was observed in 93.7% (192/205) (95% CI, 89.5 to 96.3%) cases (Table 1). The PPA of ID Now in comparison to StandardM nCoV Real-Time was found to be 91.8% (95% CI, 84.6 to 95.8%) and NPA was 95.4% (95% CI, 89.6 to 98.0%).

Table 1
Performance of Abbott ID NOW in comparison to StandardM nCoV Real-Time PCR (SD Biosensor) Assay

Molecular Assay	StandardM nCoV Real-Time PCR				
	Positive	Negative	PPA (± 95% CI)	NPA (± 95% CI)	OPA (± 95% CI)
Abbott ID NOW					
Positive	89	5	91.8%	95.4%	93.7%
Negative	8	103	(84.6–95.8)	(89.6–98.0)	(89.5–96.3)

There were 8 samples that were false negative by ID Now assay. Similarly, StandardM nCoV kit failed to detect SARS-CoV-2 RNA in 5 clinical specimens. Overall, non-concordance of results was observed between the two methods in 13/205 (6.34%) cases.

The mean Ct value for concordant positive samples was 22.0 (95% CI, 20.6–23.3), ranging from 18–33, with a standard deviation of 6.42. The mean Ct for discordant samples (RT-PCR + / ID NOW -) was 27.5 (95% CI, 24–31). The data indicates that majority of the discordant samples (false negative on ID NOW) exhibited comparatively higher Ct values or lower viral load.

The claimed sensitivity of detection of ID NOW is 125 genomic equivalents/ mL which is lower than that of STANDARD M nCoV Real-Time Detection kit which is 250–500 copies/ ml upper respiratory specimen. This is quite surprising as the 3.9% (8 cases) with higher Ct values were missed by ID NOW assay. Similarly there were 5 cases that were missed by Real Time PCR, but were picked up on ID NOW.

Discussion:

Accurate results along with rapid turnaround time of testing is of utmost importance for SARS-CoV-2 testing not only for patient management but also to curtail community spread of the infection [2]. There was a huge demand for diagnostic kits and reagents which were made available to the healthcare centres through emergency use approval (EUA) mode. Abbott ID NOW COVID-19 POCT assay claimed to provide accurate results in less than 15 minutes, our study was intended to verify its accuracy and to evaluate its efficacy for clinical use.

Our study showed PPA and NPA of 91.8% and 95.4% respectively. Though testing was done strictly as per the manufacturer's instructions, i.e. dry swab was processed within 1–2 hours of sample collection, discrepancy was observed in 13 of the samples. Out of the 8 samples that were missed on ID NOW, four of them were known COVID positive patients and were put on treatment. The remaining 4 were had reported to OPD for testing, three of which had relevant clinical symptoms and one patient had close contact with another COVID-19 positive patient. There were 5 samples that were missed by RT-PCR but

picked up by ID NOW assay, 4 of these were again known COVID positive patients on treatment and one was asymptomatic patient admitted in the hospital for some surgical procedure. The discrepancy between the two assays could be attributed to variability in sampling and the fact that majority of the samples were COVID-19 positive patients on treatment and displayed comparatively higher ct values indicating lower viral burden.

Published studies on ID NOW COVID-19 assay have documented PPA of 48.0–94.0% and NPA of 98.6 to 100% (Table 2) [3–12]. A study by Basu et al. had shown comparison of Abbott ID NOW with Cepheid Xpress Xpert SARS-CoV-2 assay on 101 specimens and had observed low PPA of 54.8% and NPA of 98.6% [3]. The study had raised concerns regarding utility of this assay for diagnostic purpose. There was another study by Lephart et al. that had reported very low PPA of 48.0% when compared with composite reference standard (m2000, Simplexa, Xpert) [4]. Sensitivity however, improved from 48–64% when nasopharyngeal swab in VTM was considered instead of dry swab [4]. Thwe et al showed overall agreement of 96.2% and PPA of 53.3% when compared against collective data set from all RT-PCR platforms (Abbott RealTime SARS-CoV-2, Panther Fusion SARS-COV-2, Cepheid Xpert Xpress SARS-CoV-2 and a laboratory developed test [5].

Smithgall et al., had evaluated performance of ID NOW assay using specimens in transport media and not direct nasal swabs and had demonstrated PPA of 73.9% and NPA of 100% with Roche Cobas as the reference method [6]. Authors concluded that PPA was 100% for high and medium viral load, however, it dropped considerably (34.3%) for lower viral load samples with ct values > 30. Another study by Harrington et al. on large number of samples (n = 524) showed lower PPA 74.73% against Abbott Real Time SARS-CoV-2 assay. In addition they also conducted in-house study on limit of detection and concluded that low PPA was attributed to higher limit of detection on ID NOW assay and preanalytical sampling error [7]. Moore et al compared ID NOW assay on 200 samples against two different assays – Modified CDC assay and Abbott Molecular RealTime SARS-CoV-2 assay with positive percent agreement of 80.30% and 75.2%) respectively [8].

Couple of studies demonstrated comparatively good PPA of greater than 90%.^[9,10] The study by Rhoads on 96 NPS collected in normal saline showed PPA of 94% when compared against modified CDC assay [9]. The authors conclude that besides PPA, limit of detection of the assay along with other variables should be considered while implementing molecular assays for clinical use. Cradic et al had used consensus standard to evaluate Abbott ID NOW assay in Nasopharyngeal Swab Specimens Collected in Universal Viral Transport Medium and demonstrated PPA of 91% [10]. Also, Abbott ID NOW was evaluated on matched specimens - swabs collected in UVT and dry swab and no difference in the performance was noted.

Our study has shown improved PPA of 91.80% in comparison to most of the other studies. The current study has a couple of limitations – the discrepancy between the two assays was not discerned by utilising the third assay and limit of detection (LoD) assay for ID NOW was not determined. However, we could resolve discrepant results through detailed review of clinical records.

Table 2
Performance of Abbott ID NOW Assay for detection of SARS-CoV-2 against other Molecular Methods

Sr. No.	Reference	No. of cases	Reference Method	PPA	NPA
1	Current study	205	SD Biosensor (Real Time PCR)	91.80%	95.40%
2	Basu et al., 2020	101	Cepheid Xpert Xpress SARS-CoV-2	54.80%	98.60%
3	Smithgall et al., 2020	113	Roche Cobas Assay	73.90%	100.00%
4	Harrington et al., 2020	524	Abbott RealTime SARS-CoV-2	74.73%	99.00%
5	Rhoads et al., 2020	96	Modified CDC Assay (RT-PCR)	94.00%	NA
6	Mitchell et al., 2020	61	Real Time PCR	71.70% (33/46)	100.00% (15/15)
7	Zhen et al., 2020	108	Hologic Panther Fusion SARS-CoV-2 assay	50/57 (87.7%)	50/50 (100%)
8	Lephart et al., 2021	88	Composite reference standard	48.00%	100.00%
9	Cradic et al., 2020	184	Consensus standard	91.00%	100.00%
10	Thwe et al., 2020	129	Panther Fusion® SARS-COV-2	53.30%	100.00%
11	Moore et al., 2020	200	Modified CDC Assay (RT-PCR)	80.30%	100.00%
		200	Abbott Molecular RealTime SARS-CoV-2 assay	75.20%	100.00%

Conclusion:

Overall, the performance of ID NOW COVID-19 assay was found to be satisfactory and comparable to RT-PCR assay. In addition, it is a POC test that offers exceptionally good turnaround time of testing and can be considered not only in the healthcare set up but also as a screening tool for travellers.

Declarations

Funding: This study was not supported by any of the funding agencies.

Conflicts of interest/Competing interests: The authors report no conflicts of interest.

Availability of data and material: The data will be made available upon request.

Code availability: NA

Authors' contributions:

- **PC:** Concepts, design, Manuscript Preparation and Editing
- **TD:** literature search, data analysis
- **RB:** experimental studies, data acquisition, data analysis
- **JB:** literature search, data analysis
- **DB:** experimental studies, data acquisition, data analysis
- **SS:** Concept, definition of intellectual content, Manuscript Review

Consent to participate: NA

Consent for publication: All the authors give consent for publication.

References

1. Shereen MA, Khan S, Kazmi A, Bashir N, Siddique R (2020) COVID-19 infection: Origin, transmission, and characteristics of human coronaviruses. *J Adv Res* 24:91–98. <https://doi.org/10.1016/j.jare.2020.03.005>
2. Ward S, Lindsley A, Courter J, Assa'ad A. Clinical testing for COVID-19 (2020). *J Allergy Clin Immunol* 146:23–34. <https://doi.org/10.1016/j.jaci.2020.05.012>
3. Basu A, Zinger T, Inglima K, Woo K, Atie O, Yurasits L et al (2020) Performance of Abbott ID Now COVID-19 Rapid Nucleic Acid Amplification Test Using Nasopharyngeal Swabs Transported in Viral Transport Media and Dry Nasal Swabs in a New York City Academic Institution. *J Clin Microbiol* 58:e01136–e01120. doi 10.1128/JCM.01136-20
4. Lephart PR, Bachman MA, LeBar W, McClellan S, Barron K, Schroeder L et al (2020) Comparative study of four SARS-CoV-2 Nucleic Acid Amplification Test (NAAT) platforms demonstrates that ID NOW performance is impaired substantially by patient and specimen type. *Diagn Microbiol Infect Dis* 99:115200. doi 10.1016/j.diagmicrobio.2020.115200
5. Thwe PM, Ren P (2020) How many are we missing with ID NOW COVID-19 assay using direct nasopharyngeal swabs? Findings from a mid-sized academic hospital clinical microbiology laboratory. *Diagn Microbiol Infect Dis* 98:115123. doi 10.1016/j.diagmicrobio.2020.115123.
6. Smithgall MC, Scherberkova I, Whittier S, Green DA (2020) Comparison of Cepheid Xpert Xpress and Abbott ID Now to Roche cobas for the Rapid Detection of SARS-CoV-2. *J Clin Virol* 128:104428. doi 10.1016/j.jcv.2020.104428
7. Harrington A, Cox B, Snowdon J, Bakst J, Ley E, Grajales P et al (2020) Comparison of Abbott ID Now and Abbott m2000 Methods for the Detection of SARS-CoV-2 from Nasopharyngeal and Nasal

- Swabs from Symptomatic Patients. *J Clin Microbiol* 58:e00798–e00720. doi 10.1128/JCM.00798-20
8. Moore NM, Li H, Schejbal D, Lindsley J, Hayden MK (2020) Comparison of Two Commercial Molecular Tests and a Laboratory-Developed Modification of the CDC 2019-nCoV Reverse Transcriptase PCR Assay for the Detection of SARSCoV-2. *J Clin Microbiol* 58:e00938–e00920. <https://doi:10.1128/JCM.00938-20>
 9. Rhoads DD, Cherian SS, Roman K, Stempak LM, Schmotzer CL, Sadri N (2020) Comparison of Abbott ID Now, DiaSorin Simplexa, and CDC FDA Emergency Use Authorization Methods for the Detection of SARS-CoV-2 from Nasopharyngeal and Nasal Swabs from Individuals Diagnosed with COVID-19. *J Clin Microbiol* 58:e00760–e00720. doi 10.1128/JCM.00760-20
 10. Cradic K, Lockhart M, Ozbolt P, Fatica L, Landon L, Lieber M et al (2020) Clinical Evaluation and Utilization of Multiple Molecular In Vitro Diagnostic Assays for the Detection of SARS-CoV-2. *Am J Clin Pathol* 154:201–207. <https://doi:10.1093/ajcp/aqaa097>
 11. Mitchell SL, St. George K (2020) Evaluation of the COVID19 ID NOW EUA assay. *J Clin Virol* 128:104429. <https://doi:10.1016/j.jcv.2020.104429>
 12. Zhen W, Smith E, Manji R, Schron D, Berry GJ (2020) Clinical Evaluation of Three Sample-to-Answer Platforms for Detection of SARS-CoV-2. *J Clin Microbiol* 58:e00783–e00720. doi 10.1128/JCM.00783-20