Evaluation of Rapid Antigen Test for Detection of SARS-CoV-2 at Narayani Hospital, Birgunj, Nepal: A Cross-sectional Analytical Study

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SARS-CoV-2, the causative agent of COVID-19, rapidly spread worldwide, creating an urgent need for reliable diagnostic tests. While reverse transcription polymerase chain reaction (RT-PCR) remains the gold standard for diagnosis, rapid antigen tests (RATs) have been widely deployed due to their ease of use and faster turnaround time. The main aim of this study is to evaluate the diagnostic performance of commercially available RATs for the detection of SARS-CoV-2 in comparison with RT-PCR. A cross-sectional study was conducted at Narayani Hospital, Birgunj, Nepal. Nasopharyngeal swab collection of 100 individuals was done for RT-PCR and rapid antigen testing. RT-PCR was considered the reference standard. Diagnostic accuracy parameters, including sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and overall accuracy of RAT, were calculated. Sensitivity was recalculated in a Ct-dependent manner. Of the 100 participants, 50 (50%) were RT-PCR positive. The rapid antigen test detected 33 true positives and 50 true negatives, yielding a sensitivity of 66%, specificity of 100%, PPV of 100%, NPV of 75%,

and overall accuracy of 83.2%. RAT performance improved in samples with lower cycle threshold (Ct <20) values, while sensitivity was markedly reduced at higher Ct values (>30). RATs demonstrate high specificity and good sensitivity in patients with high viral loads, making them useful for rapid screening and outbreak control. However, their reduced sensitivity in low viral load cases highlights the need for confirmatory RT-PCR testing when RAT results are negative but clinical suspicion remains high.

Keywords: covid-19, diagnostic accuracy, rapid antigen test, rt-pcr, sars-cov-2.

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) that caused the coronavirus disease 2019 (COVID-19) pandemic has persisted in posing a threat to public health systems around the globe. Millions of people have been infected worldwide since SARS-CoV-2 first appeared in late 2019, causing significant morbidity, mortality, and socioeconomic disruption.[1]

Accurate diagnosis is critical for outbreak control, as it enables isolation, contact tracing, and treatment initiation. Considering its great sensitivity and specificity, real-time reverse transcription polymerase chain reaction (RT-PCR) is still the gold standard.[2,3] However, RT-PCR's drawbacks, such as its high cost, lengthy turnaround times, and requirement for specialised labs and trained staff, limit its applicability in situations with low resources and for mass screening.[4] Therefore, rapid and accurate tests for SARS-CoV-2 screening are essential for disease control, as well as screening during pre-operative management for invasive procedures.

Rapid antigen tests (RATs) detect viral proteins such as nucleocapsid antigens through lateral flow immunoassays. They are inexpensive, provide results within 15–30 minutes, are easy to perform, and therefore, are suitable for point-of-care testing.[5] Despite these advantages, RATs are generally less sensitive than RT-PCR, particularly in asymptomatic individuals or those with low viral loads.[6,7]

Several international health authorities, including the World Health Organization (WHO) and National Testing Guidelines for COVID-19, have included Rapid Antigen Test along with the pre-existing RT PCR for diagnostic purposes.[8,9] It is useful for mass screening, urgent reports at health centres, and for people living in congested areas, e.g., prisons, barracks.

RATs have been widely used in Nepal for extensive screening, especially in border areas and during outbreak investigations. To support public health decision-making, it is crucial to verify the quality of these

quick antigen test kits by assessing their diagnostic performance against RT-PCR and analysing COVID-19 positive cases for efficacy. The requirement for context-specific assessments is highlighted by the fact that RAT diagnosis accuracy differs among brands, populations, and testing situations.[10,11]

This study aimed to assess the diagnostic efficacy of a commonly used RAT in comparison to RT-PCR in patients with suspected COVID-19 who presented to Narayani Hospital in Birgunj. The purpose of the study was to assess the sensitivity, specificity, and predictive values, and to recalculate sensitivity in a CT-dependent manner.

Materials & Methods

Study Design and Setting

A hospital-based cross-sectional study was conducted at Narayani Hospital, Birgunj, Parsa, Nepal, from May to October 2021. This study was approved by the Institutional Review Board of Nepal Health Research Council (Approval No. 357-2021). Informed consent was obtained from all participants.

Participants

A total of 100 suspected COVID-19 patients were enrolled. Individuals presenting with respiratory symptoms, fever, or epidemiological linkage to confirmed COVID-19 cases were included. Patients with poor-quality samples or incomplete results were excluded.

Sample Collection

100 respiratory samples (mainly nasopharyngeal and throat swabs) obtained from COVID-19 suspected cases and contact individuals presented at Narayani Hospital, Birgunj, were collected. A rapid antigen test was performed on 50 swabs from patients previously tested positive by SARS-CoV-2 PCR and 50 swabs from patients previously tested negative by SARS-CoV-2 PCR.

Testing Procedures

Rapid Antigen Test (RAT): Conducted using Rapid SARS-CoV-2 antigen detection assay Standard Q COVID-19 Ag test (SD Biosensor, Republic of Korea). Results were interpreted within 15-30 minutes according to manufacturer guidelines.

RT-PCR: RNA extraction and detection were performed using Automated ZhongkeBio Brand nucleic acid extraction and purification instrument, ZK 96 (Nanjing Zhongkebio Medical Technology Co., Ltd, China) and real time RT-PCR: Real Time PCR Kit for Novel Coronavirus

2019-nCov (ORF 1ab, N) (Uni- medica, China) which targets nucleocapsid (N) and Open Reading Frame 1ab (ORF 1ab) genes of SARS- Cov-2. The Rotor-Gene Q real-time PCR cycler (Qiagen) was used for amplification. The result was analysed using Qiagen viewer, in which a cycle threshold (Ct) value≤38 and an S-shaped amplification curve for both target genes was defined as a positive result.

Statistical Analysis

The collected data was entered into Microsoft Office Excel 2007 software. Data analysis was done by using SPSS 11.5 (Statistical Package for Social Sciences).

Diagnostic accuracy parameters were calculated using RT-PCR as the reference standard:

Sensitivity = TP / (TP + FN)

Specificity = TN / (TN + FP)

PPV = TP / (TP + FP)

NPV = TN / (TN + FN)

Accuracy = (TP + TN) / Total

Subgroup analysis was performed based on Ct value ranges (<20, 20–30, >30).

Results

Participant Characteristics

A total of 100 participants were tested, with 50 (50%) RT-PCR positive and 50 (50%) RT-PCR negative. We evaluated the performance characteristics of the SARS-CoV-2 antigen detection assay (Standard Q COVID-19 Ag test). The results were interpreted as positive when both control (C) and SARS-CoV-2 antigen (T) lines appeared within 30 min, as shown in **Figure 1**.

Among 50 RT-PCR positive cases tested, 17 were negative, and 33 were strongly positive in the Antigen test, as illustrated in **Table 1**. The antigen test showed moderate sensitivity (66%) and excellent specificity (100%), correctly identifying all true negatives but missing some positives. With a PPV of 100%, NPV of 75%, and overall accuracy of 83.2%, the test is reliable for confirming positive cases but less effective for ruling out infection.

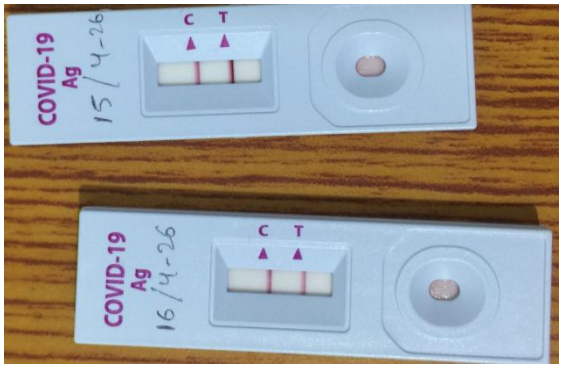

Figure 1. A positive result in the Standard Q COVID-19 Rapid Antigen Test is indicated by the appearance of both the control (C) and test (T) lines within 30 minutes

Table 1. Sensitivity, specificity, true positive, false positive, true negative, and false negative results of the diagnostic test

	PCR (Positive)	PCR (Negative)	Total
Antigen (Positive)	33 (TP)	0 (FP)	33
Antigen (Negative)	17 (FN)	50 (TN)	67
Total	50	50	100

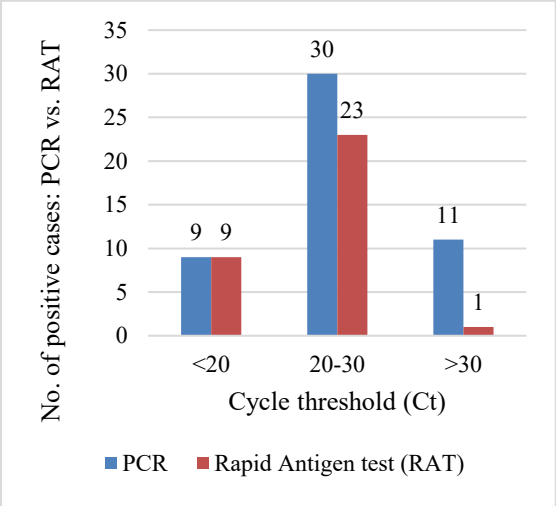

Figure 2. Number of SARS-CoV-2 positive cases: PCR vs. RAT with respect to the viral load

Those negative cases found had a CT value of more than 30 in 10 cases and 20-30 in 7 cases, as shown in **Figure 2**.

Figure 3 illustrates that the RAT sensitivity was highest (~100%) at Ct <20 (high viral load), moderate (76%) at Ct 20–30, and very low (10%) at Ct >30.

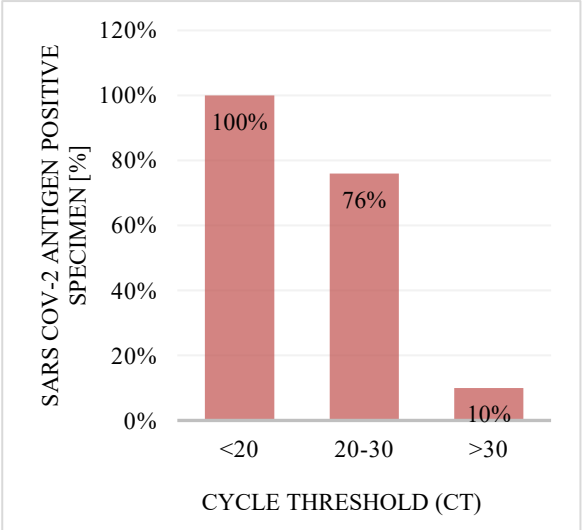

Figure 3. Sensitivity of the SARS-CoV-2 Rapid Antigen Test with respect to the viral load of clinical specimens

Discussion

This study evaluated the diagnostic performance of RAT compared to RT-PCR among suspected COVID-19 cases. RAT demonstrated **excellent specificity (100%) and PPV (100%)**, indicating that a positive result is highly reliable. However, its **sensitivity was only 66%**, consistent with prior reports showing variable antigen test performance.

Performance was strongly influenced by viral load, as indicated by Ct values. Sensitivity reached 100% in samples with Ct <20 but dropped to 10% in cases with Ct >30.

Our results align with prior research. A systematic review by Dinnes et al. reported a pooled RAT sensitivity of 72% and specificity of 99%.[6] Pilarowski et al.

observed similar findings, emphasizing RAT effectiveness in detecting high viral load cases.[12]

Our findings align with those of **Shrestha et al.** in Nepal, who reported 85% sensitivity, 100% specificity, and 93.8% accuracy in a quarantine-based population.[13] The higher sensitivity in their cohort may reflect population differences, as their subjects were close contacts under quarantine, while our hospital cohort included symptomatic patients with variable viral loads.

Similarly, an evaluation by **Corman et al.** in Germany compared seven commercial antigen kits and found that most reliably detected viral loads in the infectious phase (approx. **2.07×10^6 to 2.86×10^7 copies per swab**) but performed poorly at lower viral loads.[14] These results mirror our observation that RAT was more effective at Ct <20 but missed many cases with Ct >30.

Internationally, studies have reported comparable findings. A Chilean study in asymptomatic individuals reported that RAT had lower sensitivity compared to RT-PCR but retained strong performance in high viral load cases.[15] Likewise, a German hospital-based evaluation showed significant variation between RAT and RT-PCR, emphasizing that false negatives were more frequent in samples with low viral loads.[16]

Taken together, these studies confirm that RATs are most useful in detecting individuals with **high viral loads**, who are also the most infectious. Negative RAT results in symptomatic cases should therefore be confirmed with RT-PCR. In resource-limited or high-volume settings, RATs remain valuable as complementary tools to RT-PCR.

Conclusion

RATs are valuable diagnostic tools due to their rapidity, ease of use, and high specificity. They perform best in individuals with high viral loads, making them effective for identifying highly infectious cases and curbing transmission. However, their reduced sensitivity in low viral load cases underscores the importance of confirmatory RT-PCR, particularly in symptomatic patients testing negative by RAT.

Public Health Implications

RAT's high specificity reduces the likelihood of false positives, making it a useful tool in outbreak control. Its rapid turnaround makes it a powerful tool for use in resource-limited settings and enables timely decision-making for mass screening in hospitals, schools, airports, and community testing centers.[8] However, the reduced sensitivity, especially in patients with low viral loads or during

early/late infection, highlights the necessity of confirmatory RT-PCR testing when clinical suspicion is high.[17]

Strengths and Limitations

The large sample size and Ct-based subgroup analysis are strengths. However, this study was limited to one RAT brand; there was a lack of follow-up testing for initially negative cases, and asymptomatic populations were underrepresented. Moreover, sequential testing strategies were not evaluated.

Future Directions

Further studies should evaluate RAT performance in asymptomatic individuals, compare multiple RAT brands, and assess serial testing approaches. Integration of RATs into national testing strategies, particularly in resource-limited countries, warrants exploration.

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