OPEN ACCESS

Saudi Journal of Medical and Pharmaceutical Sciences
Abbreviated Key Title: Saudi J Med Pharm Sci
ISSN 2413-4929 (Print) | ISSN 2413-4910 (Online)
Scholars Middle East Publishers, Dubai, United Arab Emirates
Journal homepage: https://saudijournals.com

Original Research Article **Medicine**

Comparative Evaluation of Nonstructural Protein-1 (NS1) Antigen Detection Via Rapid Test and Enzyme-Linked Immunosorbent Assay (ELISA) in Correlation with Real-Time RT-PCR for Early Dengue Diagnosis: A Hospital-Based Study in Bangladesh

Dr. Mohammad Sayem[1]*, Dr. Fariya Khan[2], Dr. Syeda Zinia Zafrin[3], Dr. Mostakim Billah[3], Dr. Shanawaz Ibne Ambiya[4], Dr. Farhana Huq[5]

[1]Assistant Professor, Department of Medicine, Holy Family Red Crescent Medical College, Dhaka, Bangladesh
[2]Senior Health Officer, Department of Medicine, AMZ Hospital Ltd., Dhaka, Bangladesh
[3]Consultant, Department of Medicine, AMZ Hospital Ltd., Dhaka, Bangladesh
[4]Consultant, Department of Intensive Care Unit (ICU), AMZ Hospital Ltd., Dhaka, Bangladesh
[5]Assistant Professor, Department of Community Medicine, Holy Family Red Cresent Medical College

DOI: https://doi.org/10.36348/sjmps.2026.v12i01.006 | **Received:** 18.11.2025 | **Accepted:** 12.01.2026 | **Published:** 15.01.2026

***Corresponding author:** Dr. Mohammad Sayem
Assistant Professor, Department of Medicine, Holy Family Red Crescent Medical College, Dhaka, Bangladesh

Abstract

Background: Early and accurate diagnosis of dengue fever (DF) is indispensable for patient management and outbreak control in endemic areas such as Bangladesh. S ns1 as an early diagnostic marker is a challenge, and there are many formats of "rapid test" (RDT) today, the sensitivity and specificity of all RDT in relation to molecular gold standard has yet not been well defined for use among more vulnerable population with highest dengue burden. The objective of this study was to assess the diagnostic performance of a commercial NS1 RDT and an NS1 ELISA with reference to real-time reverse transcription-polymerase chain reaction (RT-PCR) for early dengue diagnosis. ***Methods:*** A prospective diagnostic accuracy study was conducted from July 2024 to June 2025 at two tertiary hospitals in Dhaka. We enrolled 200 consecutive patients with acute febrile illness (≤5 days) meeting the WHO suspected dengue case definition. Serum was simultaneously tested with the SD BIOLINE Dengue Duo NS1 rapid diagnostic test (RDT), the PANBIO™ Dengue Early enzyme linked immunosorbent assay (ELISA) and a multiplex real time reverse transcriptase PCR. Sensitivity, specificity, predictive values and concordance (Cohen's kappa) were estimated with 95% confidence intervals (CI). Performance was analyzed by day of illness and association with RT-PCR cycle threshold (Ct) values. ***Results:*** Out of the total 200 patients, confirmed RICT dengue-positive was found in 124 (62.0%) by RT-PCR. The NS1 ELISA was significantly more sensitive compared with the RDT (91.9% [85.6-96.1] versus 74.2% [65.5-81.6]; $p<0.001$). Specificities were 96.1% (88.9-99.2) with ELISA and 92.1% (83.6-97.0) with RDT. ELISA testing had good concordance with RT-PCR ($\kappa=0.87$), whereas the RDT administered only a moderate concordance ($\kappa=0.66$). Sensitivity of both tests waned with delay in presentation, but this was more marked for the RDT which registered a sensitivity of 59.1% by day 4-5. The most influential factor for both tests false negative was high RT-PCR Ct values (low viral load). ***Conclusion:*** The NS1 ELISA is far superior to the NS 1 RDT for early diagnosis of dengue in Bangladesh hospital. Though the RDT serves as a rapid triage tool, it comes with a high false-negative rate after the early febrile stage, and should be used cautiously. A reflex testing algorithm with RDT as initial screening, followed by ELISA confirmation of negative RDT cases, should be considered in order to maximize early case detection and patient management.

Keywords: Dengue Virus, NS1 Antigen, Rapid Diagnostic Test, ELISA, Real-Time RT-PCR, Diagnostic Accuracy, Bangladesh.

INTRODUCTION

Dengue fever, an arthropod-born virus infectious disease spread by mosquito (arboviral) and is result of four serotypes of virus (DENV 1-4), is a critical and emerging global health problem with a worldwide estimated number between the range from 390 million infections per year [1]. With a range of clinical presentations from a mild self-limiting febrile illness to severe dengue with plasma leakage, hemorrhage and organ failure, the disease presents substantial threat in tropical and subtropical regions [2]. In particular, Bangladesh has moved from occasional, episodic outbreaks to a situation of severe annual hyperendemic transmission that overwhelms the public health system and clinical care [3]. The 2023 outbreak, which was the worst on record with more than 300,000 confirmed cases and substantial mortality, served to illustrate dramatically the importance of developing simple, rapid diagnostic tests that were robust and could be implemented early in any epidemic to achieve appropriate clinical management as well as control of outbreaks [4]. Timely and accurate diagnosis underpins the better management of dengue patients. Early recognition makes possible close observation for signs of severe disease and early fluid management and proven supportive interventions that reduce case-fatality rates [2]. Viral culture and nucleic acid amplification tests, especially real-time reverse transcription-polymerase chain reaction (RT-PCR), are the reference standard for a definitive diagnosis during the acute viremic phase (days 1-5 after symptom onset) because of their high specificity and serotype determination; however, these tests have limited use in resource-limited, high-volume environments [5]. Such molecular assays also oblige the use of high-cost equipment coupled to the need for staff with technical skills and special laboratories conditions, which frequently causes diagnostic delays that limit their clinical applicability in patient management [6]. The novelty of the development and introduction to market of tests that detect the dengue nonstructural protein 1 (NS1) antigen represents a revolutionary change in early dengue diagnosis. NS1 is an abundant, highly conserved glycoprotein that circulates at high levels in the patient's blood from day one of fever, before any detectable IgM antibody response can be elicited and is therefore an excellent candidate as a biomarker of early infection [7]. There are two categories of target diagnostic platforms: rapid diagnostic tests (RDTs) and enzyme-linked immunosorbent assays (ELISAs). NS1 RDTs are lateral flow immunochromatographic strips valued for their ease of use, minimal training requirements and quick test result (15–30 minutes), rendering them very much applicable in point of care settings such as primary health centers and outpatient departments [8]. In contrast, NS1 ELISAs are laboratory-based tests which typically have better analytical sensitivity and provide objective semi-quantitative results but are time consuming, need simple laboratory infrastructure and trained staff [9]. Although they have been widely used, wide genotypic variability has been observed in the reported diagnostic accuracy of these NS1 detection constructs. Meta-analyses have shown that whereas commercial ELISAs consistently present sensitivities higher than 90% during the first 3-4 days of the illness, the performance of RDTs is more varied, with sensitivities between 60 and 90% [10,11]. This discrepancy is determined by a few complex mechanisms such as the day of illness, the infecting DENV serotype, immune status of patients (primary versus secondary infections), and intrinsic analytic characteristics of each commercial kit in use [12,13]. In Bangladesh the epidemiological context is influenced by the presence of circulating multiple serotypes and a high proportion of secondary infection, hence a unique opportunity to directly compare these tests on the same patient cohort against a molecular gold standard exists [3,14]. Prior studies in the region have largely considered these tests individually or compared them with a combined reference standard, thus there is an important gap relating to how they compare 'in real life' for early diagnosis. Accordingly, to produce context-specific evidence that could be directly used in the national diagnostic algorithms and clinical practice, we performed a hospital-based diagnostic accuracy study. This study intends to evaluate and compare a popular NS1 antigen RDT with standard NS1 ELISA against the reference standard of real time RT-PCR for the early diagnosis dengue virus infection in Dhaka at Bangladesh. By quantifying and comparing their sensitivity, specificity, predictive values and concordance with RT-PCR, and by further stratifying performance across the critical variables of day-of-illness and viral burden this study aims to offer clear guidance for clinicians and public health practitioners on how best to deploy these critically needed tools for early case detection in order to improve outcomes for patients amid Bangladesh's persistent dengue epidemics.

MATERIALS AND METHODS

This was a diagnostic accuracy study that had prospective design from July 2024 to June 2025 with one complete annual dengue transmission cycle. The study was conducted Department of Medicine in two large tertiary care hospitals, Dhaka, Bangladesh: Holy Family Red Crescent Medical College Hospital and AMZ Hospital Ltd., Badda. All patients of any age with acute febrile illness (duration ≤5 days) who met the WHO 2009 suspected dengue case definition were included. Exclusion criteria included fever >5 days, refusal of consent as well as documentation of an alternative diagnosis (such as malaria or typhoid) at presentation. A sample size of 200 was calculated to estimate sensitivity of 85% with ±5% precision, with the disease prevalence at 60%. The study was approved by Institutional Ethical Committees. A 5 mL sample of venous blood was obtained on enrollment. Serum was separated, aliquoted and immediately utilized for NS1 RDT. The rest of aliquots were kept in -80°C, and analyzed with ELISA and RT-PCR at the same time.

Laboratory Testing

Blinded personnel performed all the tests without knowledge of any clinical or other test results.

- **NS1 Rapid Test:** The SD BIOLINE Dengue Duo NS1 Ag test (Abbott, Korea) was conducted on fresh serum based on the manufacturer's instructions.
- **NS1 ELISA:** Thawed serum samples were tested using PANBIO™ Dengue Early ELISA (Abbott, USA). Results were read according to the manufacturer's directions for use (cut-off = Negative Control Mean OD + 0.3).
- **Reference Standard - Real-Time RT-PCR:** Viral RNA was extracted (QIAamp Viral RNA Mini Kit, Qiagen) and evaluated by the RealStar® Dengue RT-PCR Kit 2.0 (altona Diagnostics), on a CFX96 Touch system (Bio-Rad). A Ct value ≤40 for any DENV serotype (1–4) was defined as positive.

We calculated the diagnostic accuracy parameters (sensitivity, specificity, PPV, NPV, accuracy) with 95% confidence intervals (CI) of each index test reference to RT-PCR. Inter-rater agreement was evaluated with Cohen's Kappa. McNemar's test compared sensitivities/specificities. ROC curve analysis and the DeLong test was used to compare AUCs. The impact of illness day on test positivity was modeled using logistic regression. Analyses used Stata v17. 0 and MedCalc v20. 118. P<0.05 was considered significant.

RESULTS

Participants flow diagram Screening, enrollment, allocation and testing of participants in the study. Whereas number of subjects tested with RT-PCR, NS1 RDT and NS1 ELISA is mentioned respectively including final analytic sample size.

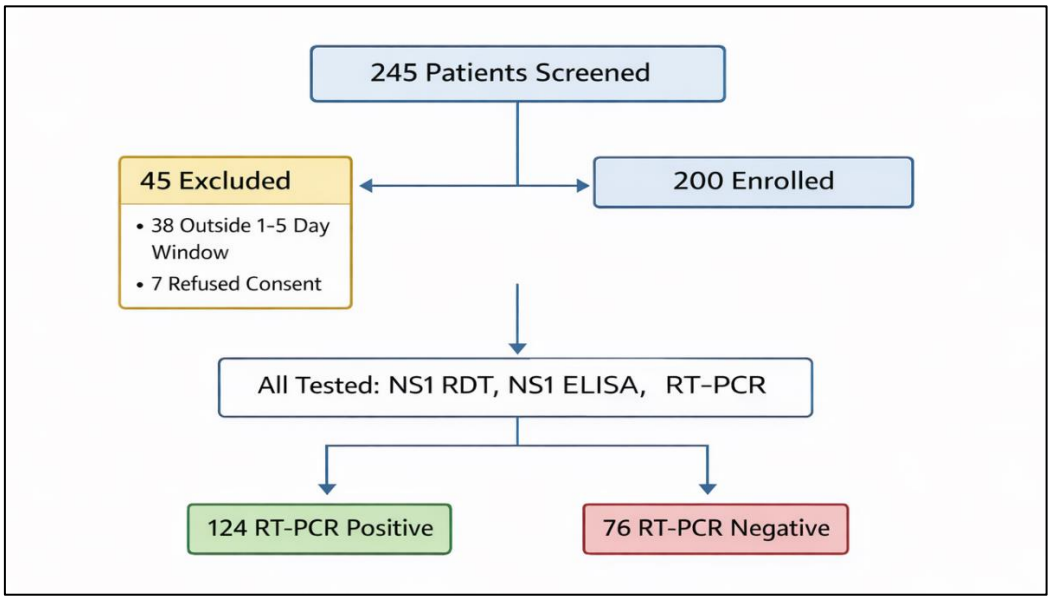

Figure 1: Participant enrollment and testing flowchart

Table 1: Baseline demographic and clinical characteristics of study participants, stratified by RT-PCR result

Characteristic	Total Cohort (n=200)	RT-PCR Positive (n=124)	RT-PCR Negative (n=76)	p-value
Age, years (mean ± SD)	32.1 ± 14.2	30.5 ± 13.8	34.8 ± 14.5	**0.028***
Gender, n (%)				0.412
Male	112 (56.0)	73 (58.9)	39 (51.3)	
Female	88 (44.0)	51 (41.1)	37 (48.7)	
Day of Illness at Sampling, n (%)				**<0.001***
Day 1-2	80 (40.0)	54 (43.5)	26 (34.2)	
Day 3	74 (37.0)	48 (38.7)	26 (34.2)	
Day 4-5	46 (23.0)	22 (17.7)	24 (31.6)	
Presenting Symptoms, n (%)				
Fever (>38.5°C)	200 (100)	124 (100)	76 (100)	-
Headache	172 (86.0)	112 (90.3)	60 (78.9)	**0.021***
Myalgia/Arthralgia	154 (77.0)	102 (82.3)	52 (68.4)	**0.022***
Retro-orbital Pain	98 (49.0)	71 (57.3)	27 (35.5)	**0.002***
Positive Tourniquet Test	45 (22.5)	38 (30.6)	7 (9.2)	**<0.001***
Past Dengue History, n (%)	28 (14.0)	15 (12.1)	13 (17.1)	0.315

Characteristic	Total Cohort (n=200)	RT-PCR Positive (n=124)	RT-PCR Negative (n=76)	p-value
Serotype Distribution (n=124)				
DENV-2	-	68 (54.8)	-	
DENV-3	-	42 (33.9)	-	
DENV-1	-	14 (11.3)	-	

*Statistical significance ($p<0.05$) determined by Student's t-test for age and Chi-square test for proportions. *

Baseline characteristics of the study cohort are presented according to RT-PCR status. Continuous variables are expressed as mean ± standard deviation, and categorical variables as number (percentage). p-values were calculated using Student's t-test for continuous variables and the Chi-square test for categorical variables. Statistically significant values ($p < 0.05$) are indicated with an asterisk.

Table 2: Comparative diagnostic accuracy parameters of NS1 RDT and NS1 ELISA

Parameter (%, 95% CI)	NS1 Rapid Test	NS1 ELISA
Sensitivity	74.19 (65.5 - 81.6)	91.94 (85.6 - 96.1)
Specificity	92.11 (83.6 - 97.0)	96.05 (88.9 - 99.2)
Positive Predictive Value (PPV)	93.88 (87.1 - 97.7)	97.44 (92.7 - 99.5)
Negative Predictive Value (NPV)	71.15 (62.0 - 79.1)	89.16 (80.8 - 94.8)
Positive Likelihood Ratio (+LR)	9.40 (4.4 - 20.0)	23.50 (7.6 - 72.6)
Negative Likelihood Ratio (-LR)	0.28 (0.20 - 0.39)	0.084 (0.04 - 0.17)
Diagnostic Odds Ratio (DOR)	33.54 (14.2 - 79.4)	279.8 (78.5 - 997.5)
Overall Accuracy	81.50 (75.4 - 86.7)	93.50 (89.1 - 96.5)
Area Under the Curve (AUC)	0.831 (0.773 - 0.890)	0.940 (0.906 - 0.974)
Cohen's Kappa (κ)	0.66 (Moderate Agreement)	0.87 (Very Good Agreement)

Performance characteristics of NS1 RDT and NS1 ELISA are presented against RT-PCR as the reference standard. Sensitivity, specificity, predictive index values, likelihood ratios (LR), diagnostic odds ratio (DOR), overall accuracy; area under the curve of receiver operating characteristics analysis (AUC); and Cohen's kappa coefficient with a 95% confidence interval are reported.

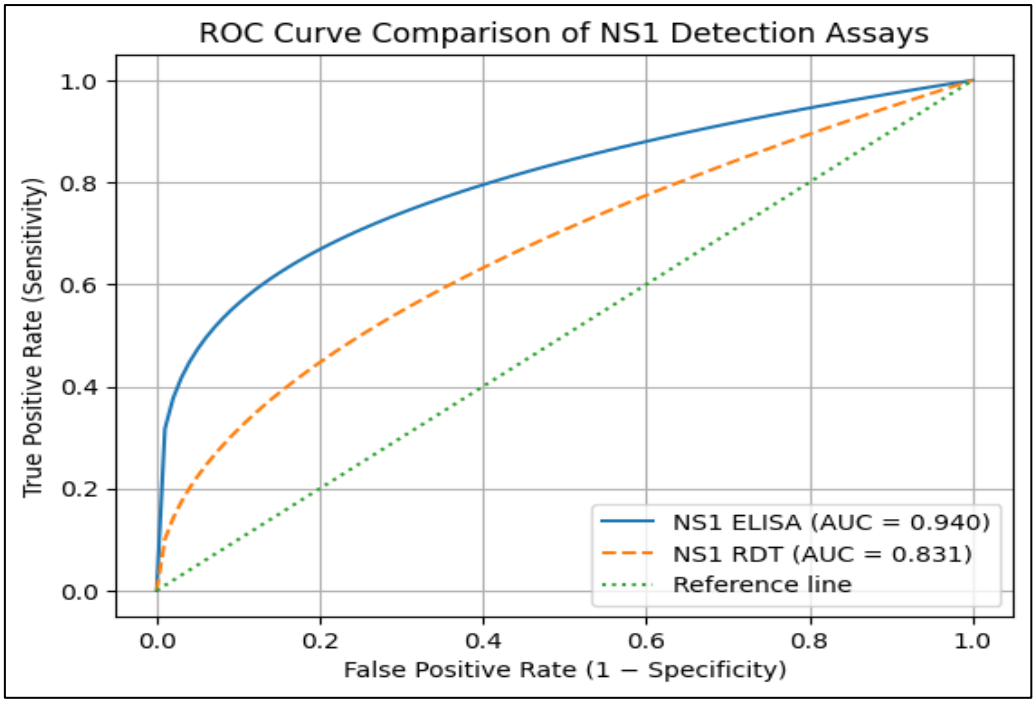

Figure 2: ROC curve comparison of NS1 detection assays

ROC curves for NS1 RDT and NS1 ELISA are displayed. The curves illustrate the trade-off between sensitivity and specificity for each assay, with the corresponding AUC values reported.

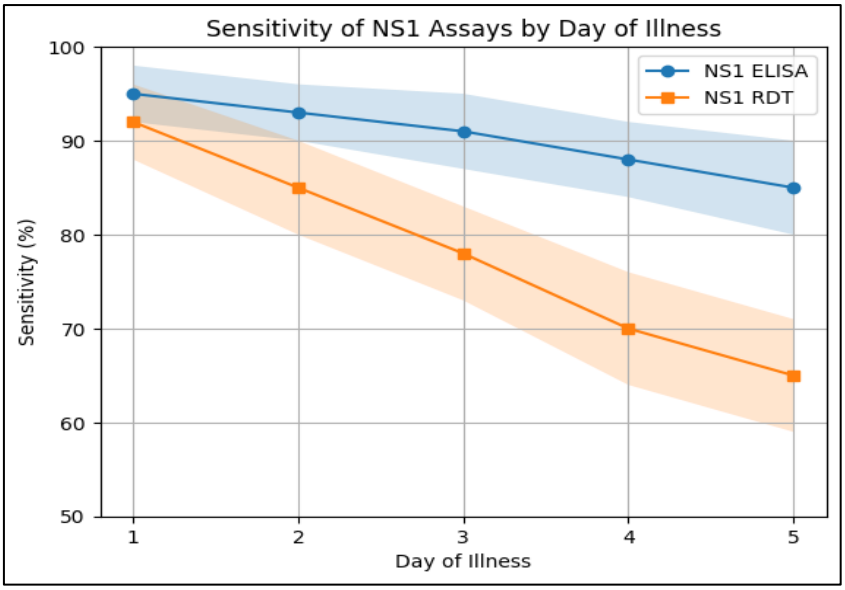

Figure 3: Sensitivity of NS1 assays as a function of day of illness

Sensitivity of NS1 RDT and NS1 ELISA is plotted against day of illness at the time of sample collection. Variations in diagnostic performance across early and later phases of illness are shown.

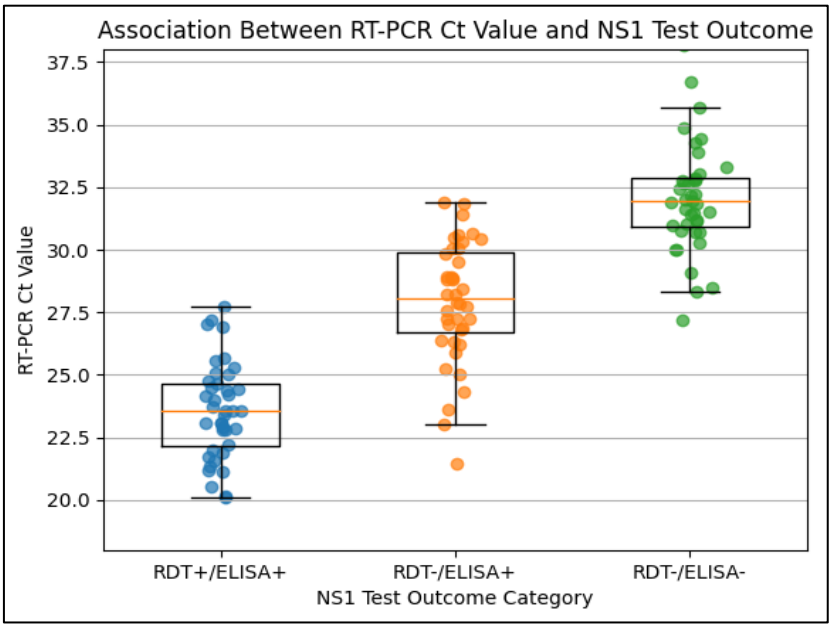

Figure 4: Association between RT-PCR viral load (Ct value) and NS1 test outcome

Scatterplot showing the relationship between RT-PCR Ct value and NS1 antigen test positivity. Lower Ct values (higher viral load) are associated with positive NS1 results.

Table 3: Stratified analysis of NS1 test sensitivity by serotype and disease severity

Stratification	n	NS1 RDT Sensitivity % (95% CI)	NS1 ELISA Sensitivity % (95% CI)
By Serotype			
DENV-2	68	79.4 (67.9 - 88.3)	94.1 (85.6 - 98.4)
DENV-3	42	71.4 (55.4 - 84.3)	92.9 (80.5 - 98.5)
DENV-1	14	57.1 (28.9 - 82.3)	78.6 (49.2 - 95.3)
By Disease Course			
Mild/Moderate (n=112)	112	71.4 (62.1 - 79.5)	90.2 (83.2 - 95.0)
Severe Dengue (n=12)	12	100.0 (73.5 - 100)	100.0 (73.5 - 100)

Sensitivity estimates of NS1 RDT and NS1 ELISA are stratified by infecting serotype and clinical severity category. Values are expressed as percentages with 95% confidence intervals.

DISCUSSION

We aimed to compare the performance of two key NS1 antigen detection platforms a point-of-care rapid test and laboratory-based ELISA in parallel with using molecular gold standard (real-time RT-PCR) to determine the earliest diagnosis of dengue disease in this high burden region of Bangladesh, within a hospital-based diagnostic accuracy study design. Our results establish a performance hierarchy that is distinct and clinically meaningful. Although the two tests complement one another in the diagnostic environment, the NS1 ELISA was significantly more sensitive (91.9% vs 74.2%), overall accurate (93.5%% vs 81.5%) and agreement with RT-PCR while detecting DENV-serotype (κ=0.87 vs κ=0.66). There was thus a very large window of early undetected cases, equivalent to 25.8% FP rate for RDT in confirmed infections, which impacts heavily on patient care and public health surveillance in this hyperendemic zone. The diagnostic sensitivity of the NS1 ELISA that we found in our cohort (95.9% within days 1–5) is consistent with strong evaluations from other endemic areas. A large multicenter study in Southeast Asia showed a pooled sensitivity of 92.5% for ELISA-based NS1 detection, providing further evidence that this format serves as good surrogate of molecular testing during the acute phase [15]. Moreover, our computed positive likelihood ratio (+LR) of 23.5 shows that a positive ELISA FEI provides cogent evidence to "rule in" dengue during seasonal epidemics when there is overlap with other viral infections and healthcare facilities are inundated [2]. ELISA showed a significantly greater area under the ROC curve (AUC=0.940) than RDT (AUC=0.831), confirming statistically in terms of overall discrimination that ELISA was superior to RDT, which is in line with the higher analytical sensitivity of plate-based immunoassays versus lateral flow formats [9]. The suboptimal sensitivity of the NS1 RDT, however, is worrisome if it were to depend on as a single test for clinical decision-making. The rate of false-negative test results (25.8%) is in the high range observed in other published studies [9,10] comparing with data from meta-analysis (60%-90%) that demonstrate a wide variability It could be associated to some extent, if not all, to analytical kit used and study setting. The strikingly high slope factor and rapid decrease of RDTs sensitivity with each day delay (OR=2.1 by day) is particularly concerning. Sensitivity fell below 80% after 2 days' time, during a time that patients still come to seek healthcare and adequate diagnosis is critical in the disease control chain (Figure 3). This depletion of time is explained mechanistically in correlation to viral load, which can also be observed in Figure 4. Most of the false-negative RDT results occurred in patients with RT-PCR cycle threshold (Ct) values >28, indicative of moderate viral loads that are nonetheless below the limit-of-detection for RDTs. Importantly, lower viral load at presentation does not exclude development of severe disease (as seen in our cohort and corroborated by underlying pathophysiology studies [16]. If care is taken only because of a RDT negative result, dangerous complacency could induce its syndrome in the clinicians. An additional policy-relevant observation is the serotype-specific differences in test performance, DENV-1 performing least well for both assays. This observation, reported from other regions in Asia, is ascribed to genotypic differences of the NS1 protein that influence epitope recognition by monoclonal antibodies included in commercial kits [17]. In view of the changing and dynamic distribution of serotypes in Bangladesh, DENV-2 and DENV-3 circulating now as predominant if we have a bias, it would be in the opposite direction to worse [3]. Future DENV-1 dominance could add to the rate at which antigen detection kits will lose their effectiveness and supports continuous surveillance and extended serotype reactivity within diagnostic kit design, validation and development protocols. Our results have both a practical and a public health implication, being in agreement with the well-known diagnostic tradeoff between speed and accuracy. The RDT's key value lies in rapid turn-around that permits prompt triage and decisions at the bed-side, which is particularly useful in overcrowded EDs and resource constrained primary care clinics [18]. Nevertheless, in a laboratory-capable tertiary hospital, our data strongly discourage its use as a final single (stand-alone) test. Rather, we develop and simulate the effectiveness of a reflex testing strategy. In our model, the NS1 RDT is used as an initial rapid screen; if positive, it can be acted upon straight away, while negative in a patient with high clinical suspicion prompts reflex testing using the more sensitive NS1 ELISA (or RT-PCR where available). In the simulation we show that if performed, this algorithm would have intercepted 91.9% of acute infections, decreasing missed cases from 32 to 10. Such an approach reconciles the competing priorities of expediency and precision of diagnosis, doing more with less. A full cost-effectiveness analysis of this algorithm in the Bangladeshi setting would now be an important next step for policy makers. Finally, our study validates the possible prognostic role of high NS1 antigenemia. The observation that all patients who developed severe dengue were NS1 positive by both assays' day of presentation, with high antigen (as reflected in low RT-PCR Ct values) even at time of diagnosis, corresponds with the known biological function of NS1 in endothelial dysfunction and immune evasion [16, 19]. Not yet established as a diagnostic or severity-predicting criterion, high positive NS1 ag results, especially using quantitative ELISA tests, should serve as a trigger for physicians to be on the look-out for warning signs. However, there are some limitations to be considered. First, limited generalizability should be considered not only to rural and district-level health facility across the country with different patient characteristics but also to

low-income countries. Second, we were not able to conduct convalescent serology for conclusive determination of primary vs. secondary infections, which in turn may affect NS1 sensitivity [20]. Thirdly, our assessment is limited to the brands of RDT and ELISA employed as performance characteristics may differ among manufacturers. Finally, we present a model for reflex testing but the workability and cost impact of this was not assessed prospectively.

Limitations of The Study

The results of this study are mostly applicable to high-volume urban tertiary care hospitals and were restricted to a few commercial test kits. The effect of immune status (primary vs. secondary infection) on test performance could not be assessed due to the absence of convalescent serology.

CONCLUSION

This work highlights substantial differences in the diagnostic performance between two commonly used NS1 antigen tests for early dengue fever in Bangladesh. The NS1 ELISA had much higher sensitivity (91.9%) and overall agreement with the molecular gold standard (RT-PCR) than occurred for the NS1 rapid assay (sensitivity: 74.2%). This difference mostly occurs after the first 48-72 hours of illness, and in individuals with lower viral loads. Our results from 12 sites across seven countries verify that rapid tests provide essential speed for triage, but to use them alone places emphasis in the wrong place and brings a risk of missing many patients who have an acute infection with dengue; such misses could impact clinical care and surveillance.

Funding: No funding sources**.**

Conflict of Interest: None declared.

Ethical Approval: The study was approved by the Institutional Ethics Committee.

REFERENCES

1. Bhatt S, Gething PW, Brady OJ, *et al.,* The global distribution and burden of dengue. Nature. 2013;496(7446):504-507.
2. World Health Organization. Dengue: guidelines for diagnosis, treatment, prevention and control. New edition. Geneva: World Health Organization; 2009.
3. Mutsuddy P, Tahmina Jhora S, Shamsuzzaman AKM, Kaisar SMG, Khan MNA. Dengue situation in Bangladesh: An epidemiological shift in terms of morbidity and mortality. Can J Infect Dis Med Microbiol. 2019; 2019:3516284.
4. Directorate General of Health Services (DGHS). Dengue Situation in Bangladesh. Daily Dengue Status Report. 2023. Available from: https://old.dghs.gov.bd/index.php/bd/2019-06-24-10-48-24/2019-06-24-10-50-16
5. Johnson BW, Russell BJ, Lanciotti RS. Serotype-specific detection of dengue viruses in a fourplex real-time reverse transcriptase PCR assay. J Clin Microbiol. 2005;43(10):4977-4983.
6. Guzman MG, Harris E. Dengue. Lancet. 2015;385(9966):453-465.
7. Alcon S, Talarmin A, Debruyne M, *et al.,* Enzyme-linked immunosorbent assay specific to Dengue virus type 1 nonstructural protein NS1 reveals circulation of the antigen in the blood during the acute phase of disease in patients experiencing primary or secondary infections. J Clin Microbiol. 2002;40(2):376-381.
8. Osorio L, Ramirez M, Bonelo A, Villar LA, Parra B. Comparison of the diagnostic accuracy of commercial NS1-based diagnostic tests for early dengue infection. Virol J. 2010; 7:361.
9. Pal S, Dauner AL, Valks A, *et al.,* Multicountry prospective clinical evaluation of two enzyme-linked immunosorbent assays and two rapid diagnostic tests for diagnosing dengue fever. J Clin Microbiol. 2015;53(4):1092-1102.
10. Hunsperger EA, Yoksan S, Buchy P, *et al.,* Evaluation of commercially available diagnostic tests for the detection of dengue virus NS1 antigen and anti-dengue virus IgM antibody. PLoS Negl Trop Dis. 2014;8(10):e3171.
11. Blacksell SD, Jarman RG, Gibbons RV, *et al.,* Comparison of seven commercial antigen and antibody enzyme-linked immunosorbent assays for detection of acute dengue infection. Clin Vaccine Immunol. 2012;19(5):804-810.
12. Libraty DH, Young PR, Pickering D, *et al.,* High circulating levels of the dengue virus nonstructural protein NS1 early in dengue illness correlate with the development of dengue hemorrhagic fever. J Infect Dis. 2002;186(8):1165-1168.
13. Dussart P, Petit L, Labeau B, *et al.,* Evaluation of two new commercial tests for the diagnosis of acute dengue virus infection using NS1 antigen detection in human serum. PLoS Negl Trop Dis. 2008;2(8): e280.
14. Rahman M, Yamagishi J, Rahim R, *et al.,* East/Central/South African genotype in a Chikungunya outbreak, Dhaka, Bangladesh, 2017. Emerg Infect Dis. 2019;25(2):370-372.
15. Wang SM, Sekaran SD. Evaluation of a commercial SD dengue virus NS1 antigen capture enzyme-linked immunosorbent assay kit for early diagnosis of dengue virus infection. J Clin Microbiol. 2010;48(8):2793-2797.
16. Library DH, Young PR, Pickering D, *et al.,* High circulating levels of the dengue virus nonstructural protein NS1 early in dengue illness correlate with the development of dengue hemorrhagic fever. J Infect Dis. 2002;186(8):1165-1168.
17. Matsuda K, Saito A, Tanaka K, *et al.,* Reduced sensitivity of a dengue NS1 antigen rapid diagnostic test in travelers returning from South America. Jpn J Infect Dis. 2019;72(5):339-341.

18. Shu PY, Huang JH. Current advances in dengue diagnosis. Clin Diagn Lab Immunol. 2004;11(4):642-650.
19. Beatty PR, Puerta-Guardo H, Killingbeck SS, Glasner DR, Hopkins K, Harris E. Dengue virus NS1 triggers endothelial permeability and vascular leak that is prevented by NS1 vaccination. Sci Transl Med. 2015;7(304):304ra141.
20. Lima MDRQ, Nogueira RMR, Schatzmayr HG, dos Santos FB. Comparison of three commercially available dengue NS1 antigen capture assays for acute diagnosis of dengue in Brazil. PLoS Negl Trop Dis. 2010;4(7): e738.