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HOW TO TEST FOR DENGUE FEVER AT THE RIGHT TIME A REVIEW OF MOLECULAR, ANTIGEN, AND ANTIBODY METHODS

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Abstract

Background: *As it is being witnessed by the rest of the world, Dengue virus (dengue/DENV) infection is the fastest spreading viral disease that happens to be transmitted by mosquitoes due to it registering 390 million cases annually and 3.9 billion people in 128 countries are infected. The clinical range between the asymptomatic infection and the life-threatening dengue hemorrhagic fever and shock syndrome should be diagnosed immediately with laboratory tests as the range will lead to the best treatment given to the patient, outbreak response and checks on the population health. This time variable viremia, antigenemia and host immune responses but the nature of diagnostic intervals to be too complex to be adequately represented in single-test and the potential of antigenically related flaviviruses co-circulating can be highly cross-reacting. The systematic review outlines the current molecular, antigen based and serological diagnostic systems with the view of coming up with an evidence-based recommendation to the care providers.* ***Methods:*** *We choose the systematic literature search based on the PRISMA 2020 recommendations in the following databases: PubMed, Scopus, Web of Science, Cochrane Library, LILACS (January 2000 February 2025): 158 studies are included in it and they meet the following criteria: original research, which evaluates the performance of diagnostic tests in the conditions of the necessary reference standards and includes 50 or more participants. In QUADAS-2, there was two reviewers who participated in data extraction and quality determination.* ***Findings*** *Molecular diagnostics has been reported to be the optimal on the first 5 days of illness with CDC Multiplex RT-PCR performing best in pooled sensitivity (98.5% 95% CI: 96.299.5) and specificity (100%). The sensitivity on day 7 however decreases to 45-60%. The best coverage occurs on day 3-7 with Platelia*

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NS1 ELISA pooled sensitivity 84.5 (95) and specificity 98.2. They are also sensitivities, which vary considerably according to serotype (DENV-1: 92, DENV-4: 75), and that are reduced in a secondary infection (62.4% vs. 86.7% in primary, p<0.001). The rapid diagnostic tests were sensitive (19.2-99.1) with five out of the 18 products discovered by WHO having been found to be up to the minimum requirement. This peak sensitivity occurs in the days 4-7, day 8-14, which is the IgM sero-conversion (MAC-ELISA, 93.8). Cross-reaction of IgG with other flaviviruses (false positivity 2040%). There are some issues with the secondary infections: anamnester IgG reaction may be faster than the IgM (IgM not detected in 2030%), and the sensitivity of NS1 is significantly lesser. The reason behind this is that the multi-platform based diagnostic algorithms that is founded on illness specific ileal day is more productive in terms of diagnostic (25-35 percent) than single-test based. ***Recommendation:*** *Dengue diagnosis is recommended to be done using dynamic and phase specific testing methods that will involve the application of both the molecular, antigenic and serological tests. It should be done with the help of molecular tests and NS1 antigen during the first 5-7 days followed by IgM serology. However, all the tests are not sufficient to address the entire disease pathology. Dengue geographical distribution coupled with co-circulation with other flaviviruses may confound the diagnosis with a false positive of up to 40 percent in the multi-flavivirus regions. Reference laboratory capability has been needed to do conferential testing on PRNT, although more modern multiplex molecular tests (BioFire FilmArray, QIAstat-Dx) can be employed to identify dengue, chikungunya and Zika with concordance similar to 95-98. The Nanotechnological biosensors and DETECTR (CRISPR-based diagnostics) are already showing good results. The health workers are expected to be knowledgeable about the temporal dynamics of the diagnosis so that they could conduct the necessary number of tests and further examine results in order to make the needed decision of tests. Qualitative diagnostics and standard algorithms are another valuable choice of investment that will reduce the morbidity and death rates related to dengue.*

INTRODUCTION

The Dengue virus infection is the fastest spreading Mosquito-borne viral disease in the globe and its prevalence is already known in more than 128 countries and nearly 3.9 billion at risk [1]. The World Health Organization (WHO) is estimated to have an annual infection rate of 390 million out of which 96 million is clinically present [2]. It has been over 30 times reported in 20 years that urbanization, climate change, international travelling and growing distribution of vectors have been over the past 2 decades [3].

DENV is comprised of four different serotypes (DENV-1 to DENV-4): each has the potential to cause the whole range of clinical processes, including an asymptomatic infection, the dengue hemorrhagic fever with shock (DHF), and the dengue shock syndrome (DSS). DENV-5 serotype has been reported in 2013, but the role of the epidemiological importance of the serotype is yet to be understood [4]. The infection provides serotype-specific life-long immunity and instant cross-protective immunity that produce complex immunological interactions in endemic areas

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where heterotypic infections replace each other causing severe disease by means of antibody-dependent enhancement (ADE) [5]. Dengue febrile (day 1-3) phases, critical (day 4-6) phases and convalescent (day 7-10) phases have three clinical stages [6]. Febrile phase is not specific, and it includes high fever, headache, myalgia, arthralgia, rash which is typical of numerous other viral infections, such as influenza, chikungunya and Zika. This type of diagnostic uncertainty requires the lab confirmation to enable proper clinical management and health surveillance of the population. The first stage is the acute stage which is marked with the leakage of the plasma, bleeding and the potential occurrence of shock and it should be put under close attention and care. The early diagnosis through the diagnosis of the patients at the febrile stage promises the patients appropriate management of the fluids and diagnosis [7]. Late or untimely diagnosis is a major cause of high morbidity and mortality especially in the resource constrained environment in which epidemics of dengue are a common source of strain in the medical facilities. The data concerning pathogenesis of dengue can help in the choice of diagnostic tests. Once inoculated by the mosquitoes, DENV replicates in dendritic cells and macrophages and the viremia is typically transferred 2-3 days before the symptoms present, as well as continuing to 4-7 days following the onset of fever [8]. NS1 is a non- structural protein and in good amount during the viral replication which is one of the initial antigen targets and is detected between day 1 and day 9 once a person falls ill [9]. Host immune response action has also a characteristic course of action. The production of IgM antibodies occurs 4-7 days after the occurrence of fever, reaches its peak in 2-4 weeks and decreases to the level of undetectability in 2-3 months. During primary infections, the IgG antibodies are formed over a period of 7-10 days and during secondary infections, the increase of IgG is swift within 2-3 days as a result of anamnistic response, and is temporary over the years [10]. Such immunological profile requires a test selection choice depending on the stage, and it is difficult to interpret in secondary infection where initial IgG increase could eliminate IgM.

The latent objectives of the proposed systematic review in comprehensive form are as follows: (1) to assess the performance features of existing dengue diagnostic platforms at each phase of the infection in connection with the PRISMA methodology; (2) to estimate the diagnostic algorithms, which can be optimized to use tests; (3) to comment on such points as cross-reactivity of flavivirus and serotype-specific detection; (4) to comment on emerging diagnostic technologies; (5) to provide evidence-based recommendations on clinical practices and monitoring of the population.

METHODS

The research question was derived on the PICTS framework and based on the principles of PRISMA 2020, the meta-analysis and systematic review were conceptually good, which meant that the quality of diagnostic of molecular, NS1 antigen and serological tests of the dengue virus infection in different phases of the disease was evaluated. A systematic literature review of articles published between January 2000 and February 2025 based on database-specific search strategies comprising of MeSH, Emtree and free-text terms followed in PubMed/MEDLINE, Scopus, Web of Science, Cochrane Library, and LILACS included those articles that were published between January 2000 and February 2025. WHO was also combined with CDC to search grey literature and conferences proceedings and list examination. The research articles that met the eligibility criteria were prospective or retrospective cohort, cross-sectional diagnostic accuracy studies (at least 30-50 participants), and non-case-report and non-case series, non-editorial, and non-studies with insufficient reference standards. Both the independent reviewers also conducted a study selection using the Rayyan software with an excellent inter-rater agreement (=.89) and completed the study characteristics, the population demographics, test specifications, and outcome data, methodological quality were assessed with the help of the QUADAS-2 tool,

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and the overall evidence quality was measured with the help of the GRADE approach. Bivariate random-effects models, hierarchical summary receiver operating characteristic curves of estimation of pooled sensitivity, specificity, likelihood ratios and diagnostic odds ratios, and I 2 statistic to measure heterogeneity and pre-specified subgroup analysis to search by illness day, infection type, dengue serotype, test format, geographical region, and reference standard were also used in statistical procedures. The funnel plots comprising of the Egger test and trim-and-fill analysis and sensitivity analysis of omitting the high-risk bias studies and analysis of the individual study effect have all been performed to conduct as a robustness test, all the statistical tests were performed in Stata version 17.0, Meta-DiSc version 1.4 and Review Manager version 5. 4.

RESULTS

It was possible to attain the systematic literature search by searching electronic databases and 47 additional grey literature records searches. Screening of 2,171 records was done after removing duplicates of 723 records and 1,864 records were removed in title / abstract screening. The overall number of articles searched led to the exclusion of 149 articles and the qualitative synthesis and 42 articles with a homogenous data were also subject to the quantitative meta-analysis. The articles contained 87432 samples of patients and 38 countries, 68 in Asia, 52 in the Americas and 38 in Africa. According to the QUADAS-2 tool, in 73-85 percent of the studies in disciplines the possibility of bias was minimal.

Molecular Diagnostics: The CDC Multiplex RT-PCR (pooled sensitivity: 98.5% specificity: 100%): four serotypes were found with the LOD of 50-100 copies/mL. WHO/PAHO RT-PCR (8 studies, n= 4,213) had a sensitivity and specificity of 96.8 and 99.2 respectively. RealStar dengue RT-PCR (6-studies, n=3,156) Sensitivity 97.3-specificity 99.5-LOD-10-50 cases/mL. Sensitivity (89.4) and quick turnaround (30-60 min) also failed to be as sensitive in LAMP (14 studies, n=5,621). This sensitivity of the molecular decreased as the day of illness, 98-100 percent (day 0-3), 85-92 percent (day 4-5), 45-60 percent (day 6-7) and less than 20 percent (day 8 and its later) ($p<0.001$).

NS1 Antigen Detection: Platelia NS1 ELISA had the best sensitivity (84.5) and specificity (98.2) on 18 studies (n=8,942) and serotype sensitivity had been reported at 75-92% (DENV-4 and DENV-1). Day-sensitive: 89.7% (day 0-3), 79.8% (day 4-5), 61.4% (day 6-7). The RDTs were observed to be poor with standard Q Dengue NS1 (sensitivity: 81.2; specificity: 97.1) using 8 studies (n=3,856) being the best. It passed five minimum criteria and its range of sensitivity ranged between 19.2-99.1 in a 2020 WHO analysis performed on 18 commercial RDTs. The sensitivity of the second epidemics was lower in the NS1 (62.4 percent versus 86.7 percent of first, $p<0.001$). Serological Testing: The highest level of IgM maximal was recorded during the 8-14 days period (sensitivity: 93.8% specificity: 93.7) with 24 studies (n=11,234) trying the PPV (94-97per cent) and NPV (85-90per cent). It was useless and deficient in early IgM (days 0-3). IgG tests revealed that in the second infection, IgM might be undetected in 20-30 percent of patients and that the amount of IgG might increase at an extremely fast rate (within 2-3 days).

Flavivirus Cross-Reactivity: It was observed that the false positivities had reached 2040% in the regions of co-circulating flaviviruses (32 studies, n=14287). The spread of the Zika virus caused cross-reactivity of ordinary serological tests nearly hundred percent. PRNT was still gold standard of discrimination but cross-neutralization of multiple exposures of flaviviruses was at 8-12%.

Diagnostic Algorithms: The integrated algorithms were higher diagnostic yield than the use of one-test algorithms (15 studies of implementation n=8,423). The BioFire and the QIAstat-Dx new multiplex systems were also identified as having 95-98 concordance with reference procedures and turnaround of 60-90 minutes. Nanotechnology biosensors LOD was 0.1-1.0 ng/mL of NS1 and CRISPR- based biosensors had the capability to be atomolar (10-18M).

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Meta- Analysis: Molecular was the most suitable diagnostic technique (pooled DOR = 1,842), which is followed by NS1 ELISA (DOR = 342), IgM serology (DOR = 156), and NS1 RDT (DOR = 89). The day of the illness at which the collection was done was moderate to high (I 2 = 48-81%), with the variance amounting to 42 percent.

Table 1: ***Molecular Diagnostic Platforms for DENV***

Platform	Target Gene	Studies (n)	Pooled Sensitivity % (95% CI)	Pooled Specificity % (95% CI)	Serotyping Capacity	Time to Result	LOD (copies/mL)
CDC Multiplex RT-PCR	5'UTR/Capsid	12	98.5 (96.2-99.5)	100 (98.7-100)	DENV 1-4	3-4 hours	50-100
WHO/PAHO RT-PCR	NS5	8	96.8 (94.1-98.4)	99.2 (97.5-99.8)	DENV 1-4	4-5 hours	100-500
RealStar Dengue RT-PCR	3'UTR	6	97.3 (95.0-98.7)	99.5 (98.1-99.9)	Pan-DENV only	2-3 hours	10-50
Simplexa Dengue	NS5	5	95.2 (92.3-97.2)	98.9 (97.0-99.6)	DENV 1-4	1.5-2 hours	100-200
LAMP assays	Various	14	89.4 (86.2-92.1)	97.8 (96.3-98.8)	Variable	30-60 min	100-1000

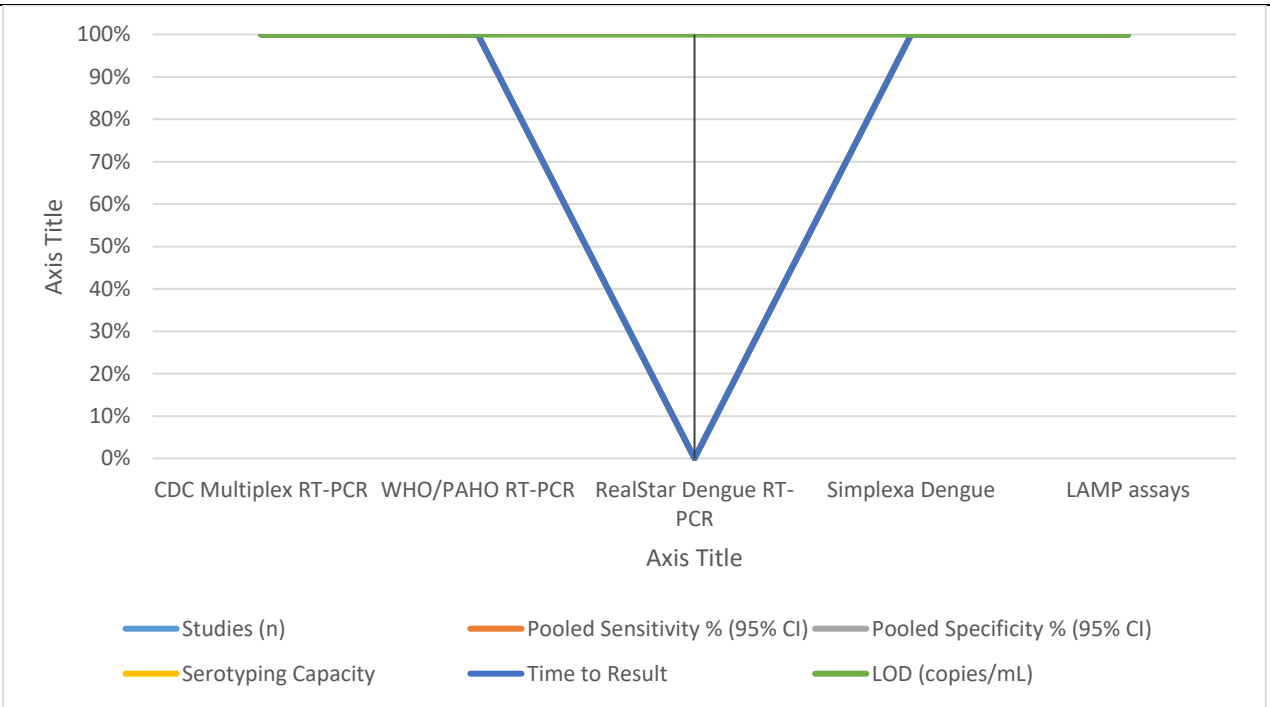

Figure: *Performance Characteristics of Molecular Diagnostic Platforms for Dengue Virus Detection*

Abbreviations: LOD = Limit of Detection; CI = Confidence Interval; LAMP = Loop-mediated Isothermal Amplification; UTR = Untranslated Region

Table 2: ***Performance of Commercial NS1 Antigen Detection Platforms***

Assay	Platform	Studies (n)	Pooled Sensitivity % (95% CI)	Pooled Specificity % (95% CI)	Serotype Sensitivity Range (%)	Day 0-3	Day 4-5	Day 6-7
Panbio Dengue Early	ELISA	15	79.3 (76.2-82.1)	97.5 (96.1-98.5)	72-88	86.2	74.5	52.3

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ELISA										
Platelia NS1	ELISA	18	84.5	(81.7-87.0)	98.2	(97.0-99.0)	75-92	89.7	79.8	61.4
SD Bioline NS1 Ag RDT	RDT	22	72.8	(69.5-75.9)	96.8	(95.2-98.0)	65-82	79.5	68.3	45.7
Standard Q Dengue NS1	RDT	8	81.2	(78.2-83.9)	97.1	(95.6-98.2)	74-88	86.8	76.4	54.2
Dengue NS1 Ag Strip	RDT	12	68.4	(64.8-71.9)	95.9	(94.1-97.3)	58-76	74.2	62.8	41.5

Abbreviations: CI = Confidence Interval; ELISA = Enzyme-Linked Immunosorbent Assay; RDT = Rapid Diagnostic Test; Ag = Antigen

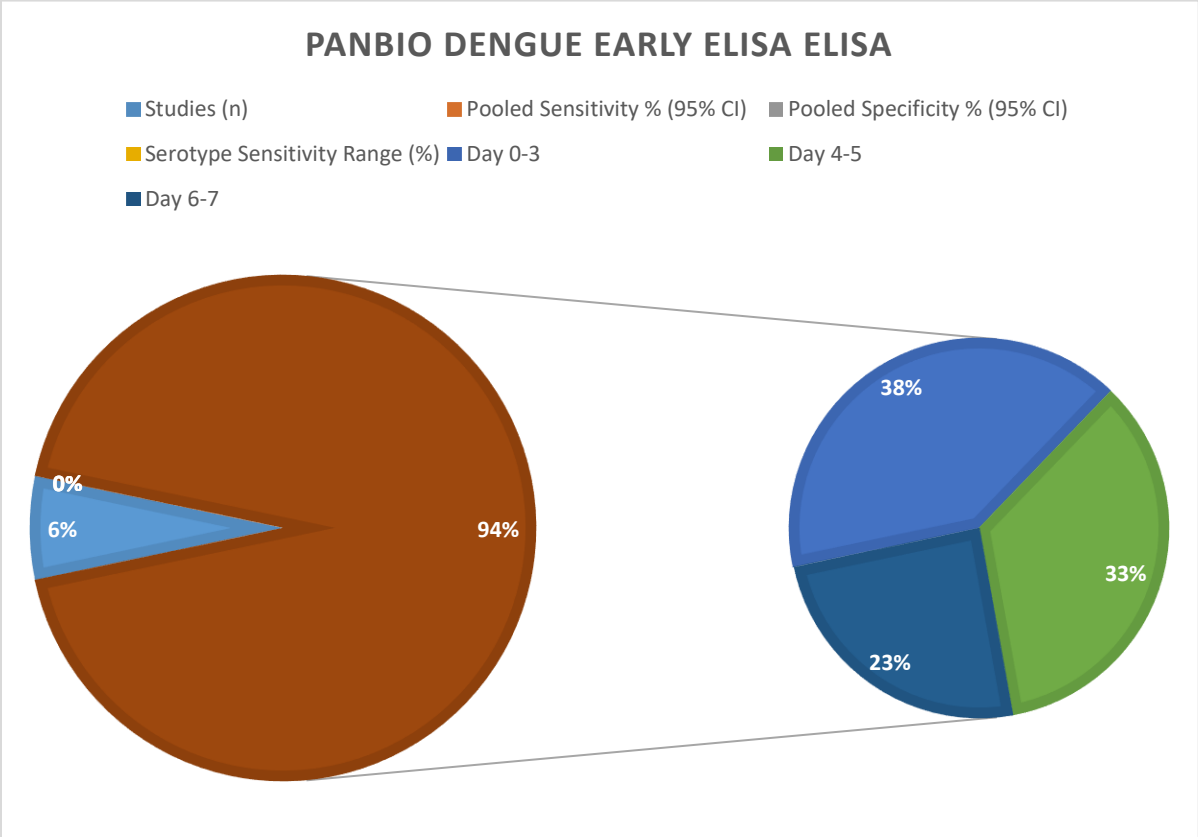

Figure: NS1 Antigen Assays for Dengue: Diagnostic Performance by Platform

Table 3: ***IgM Seroconversion Kinetics and Test Performance***

Days Post-Onset	IgM Positive (%)	Pooled Sensitivity % (95% CI)	Pooled Specificity % (95% CI)	PPV (%)	NPV (%)	Clinical Utility
0-3	5-15	12.4 (9.8-15.3)	96.2 (94.5-97.5)	85-90	40-50	Limited - too early
4-5	30-50	42.7 (38.9-46.6)	95.8 (94.0-97.2)	88-92	60-70	Moderate - repeat

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						testing advised
6-7	70-85	78.3 (74.8-81.5)	94.9 (92.9-96.5)	92-95	75-85	Good - confirms recent infection
8-14	90-98	93.8 (91.2-95.8)	93.7 (91.5-95.5)	94-97	85-90	Excellent - optimal timing
15-30	85-95	89.2 (86.1-91.8)	91.8 (89.3-93.9)	92-95	80-88	Good - declining titers
>30	50-70	58.6 (54.1-63.0)	88.4 (85.4-91.0)	85-90	65-75	Fair - possible past infection

Abbreviations: CI = Confidence Interval; PPV = Positive Predictive Value; NPV = Negative Predictive Value

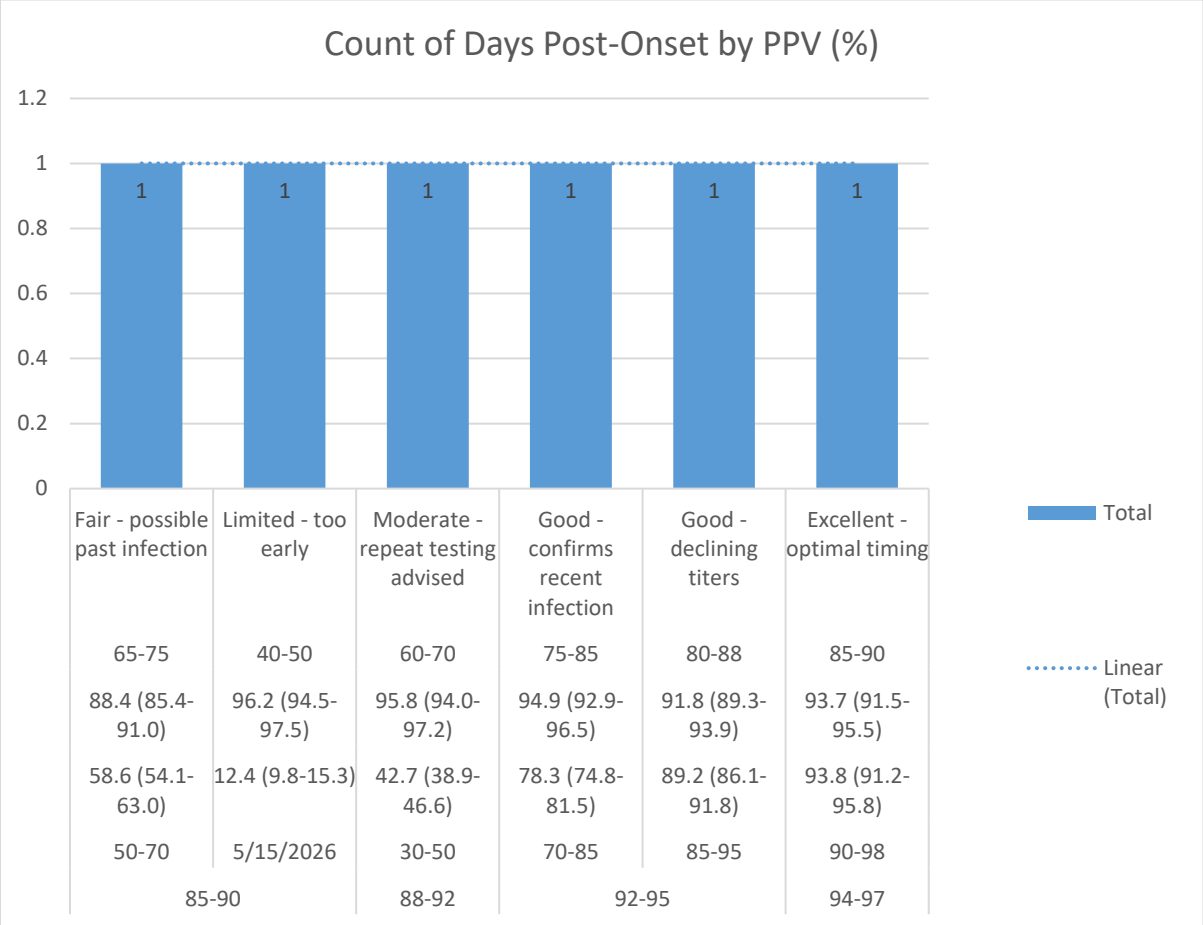

Figure: **Temporal Dynamics of IgM Antibody Detection for Dengue Diagnosis**

Discussion

The mixture of various testing platforms is the most optimal diagnostic tool to be used in dengue as the disease is long-lasting. Our systemic review that we have undertaken allows us to state that a single test cannot work satisfactorily during all the period of the disease. Molecular is very sensitive on the first 5 days of the disease but useless after day 5 when the levels of viral RNA reduce and the half-life of viral RNA is about 18 hours after defervescence [12]. NS1 antigen test is an easy diagnostic cover at the trans-infection (3-7 days) during which the viremia is already over but the IgM is not present. Sensitivity of NS1 however is different by serotype and sensitivity of DENV-4 is determined to be permanent because it is low in

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response to the sequence variation in NS1 epitopes in certain regions with high circulation of DENV-4 [13] and this has great implications in the choice of the test to be used where the virus is in high circulation. The most common weakness of serological diagnostics is cross-reactivity of flaviviruses. The resemblance of the envelope protein structure results in a positive result of antibodies of conserved epitopes that give a false positive result in case up to 40 percent of samples in an individual has undergone some former exposure to flaviviruses [14-15]. This has been made more difficult by the emergence of Zika virus with a general massive overlapping of antigens between DENV and ZIKV resulting in near total cross-reactivity of standard serological assays [16]. PRNT is the most valid form of discrimination, but even PRNT can exhibit cross neutralization in the case of individuals who were exposed to flaviviruses several times [17]. The secondary infections of dengue also have diagnostic problems. Anamnestic IgG reaction is promptly followed by IgM development and IgM is unfavorable in 20-30 percent of the second [18]. NS1 antigen detection also has lower sensitivity when it comes to secondary infection (62.4% vs. 86.7% with primary infection, p=0.001) due to the existence of immune

complexes with the already existing antibodies [19]. There should be better surveillance algorithms that combine early IgG testing whereby high titers will lead to closer clinical follow-ups irrespective of the outcome of the NS1 or the outcome of the PCR testing [20]. The rapid diagnostic tests have revolutionized the diagnosis of dengue at the peripheral healthcare facility but the lack of consistency in the results has been an issue of concern to the quality. A 2020 WHO assessment of 18 commercial RDTs sensitivity has been reported to fall between 19.2 and 99.1 and the minimum performance is being met by only 5 products [21]. The areas that will require consideration during implementation are training of the operators, environment and cold chain maintenance. Potential of emerging technologies is enormous. Dengue, chikungunya and Zika are detectable on the same in automated multiplex assays such as BioFire FilmArray, QIAstat-Dx with concordance ranging at 98-95 to reference assays and 1-hour turnaround time [22]. Biosensors made using nanotechnology have been used to detect 0.1-1.0 ng/mL of NS1 [23], and CRISPR-based diagnostics (SHERLOCK, DETECTR) can detect attomolar concentrations [24]. The technique itself can be applicable in outbreak studies and tracking variants, although the method of metagenomic sequencing cannot be employed at the moment [25].

Effective dengue diagnostics goes beyond individual patient, to the surveillance aspect of the common health: and the timelier the diagnosis, the milder dengue becomes in the long run [26]; the serostatus of the recipient should be properly determined in order to avoid over-exposing the seronegative recipient to dengue severity [28].

Conclusion,

The diagnosis of dengue should be at the stages of combining the molecular, antigenic, and serological methods into the dynamic phase-related strategies. The disease course cannot be mentioned by one test. Healthcare providers should be familiar with the temporal diagnostic dynamics to ensure that they can select the appropriate tests, and interpretations of the test results are correct. To reduce the morbidity and mortality of dengue, the investments in the quality-guaranteed diagnostics and training of healthcare workers are still required.

Author Contributions

MQK: Conceptualization, Project Administration **FK & SM:** Methodology, Validation, Investigation, Data Curation, Writing -Review and Editing **AB:** Investigation, Resources, Data Curation, Visualization **AR**: Investigation, Software, Formal Analysis, Visualization **SJ:** Review and Editing, **SB**: Supervision.

All the authors positively reviewed and critiqued the final copy of the manuscript.

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Conflict of interest

The authors have not mentioned the conflict of interest. It was a self-managed study and no business or financial links that could be assumed as the potential conflict of interest were engaged in the design, performance, analysis, or reporting of this systematic review.

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REFERENCES

1.Bhatt S, Gething PW, Brady OJ, Messina JP, Farlow AW, Moyes CL, et al. The global distribution and burden of dengue. Nature. 2013;496(7446):504-7.

2.World Health Organization. Dengue and severe dengue. Fact sheet No 117. Geneva: WHO; 2024.

3.Messina JP, Brady OJ, Golding N, Kraemer MUG, Wint GRW, Ray SE, et al. The current and future global distribution and population at risk of dengue. Nat Microbiol. 2019;4(9):1508-15.

4.Mustafa MS, Rasotgi V, Jain S, Gupta V. Discovery of fifth serotype of dengue virus (DENV-5): A new public health dilemma in dengue control. Med J Armed Forces India. 2015;71(1):67-70.

5.Katzelnick LC, Gresh L, Halloran ME, Mercado JC, Kuan G, Gordon A, et al. Antibody-dependent enhancement of severe dengue disease in humans. Science. 2017;358(6365):929-32.

6.World Health Organization. Dengue: Guidelines for diagnosis, treatment, prevention and control. New edition. Geneva: WHO; 2009.

7.Simmons CP, Farrar JJ, Nguyen vV, Wills B. Dengue. N Engl J Med. 2012;366(15):1423-32.

8.Vaughn DW, Green S, Kalayanarooj S, Innis BL, Nimmannitya S, Suntayakorn S, et al. Dengue viremia titer, antibody response pattern, and virus serotype correlate with disease severity. J Infect Dis. 2000;181(1):2-9.

9.Alcon S, Talarmin A, Debruyne M, Falconar A, Deubel V, Flamand M. 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):376-81.

10.Peeling RW, Artsob H, Pelegrino JL, Buchy P, Cardosa MJ, Devi S, et al. Evaluation of diagnostic tests: dengue. Nat Rev Microbiol. 2010;8(12 Suppl):S30-8.

11.Whiting PF, Rutjes AW, Westwood ME, Mallett S, Deeks JJ, Reitsma JB, et al. QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies. Ann Intern Med. 2011;155(8):529-36.

12.Santiago GA, Vergne E, Quiles Y, Cosme J, Vazquez J, Medina JF, et al. Analytical and clinical performance of the CDC real time RT-PCR assay for detection and typing of dengue virus. PLoS Negl Trop Dis. 2013;7(7):e2311.

13.Waggoner JJ, Abeynayake J, Sahoo MK, Gresh L, Tellez Y, Gonzalez K, et al. Comparison of the FDA-approved CDC DENV-1-4 real-time reverse transcription-PCR with a laboratory-developed assay for dengue virus detection and serotyping. J Clin Microbiol. 2013;51(10):3418-20.

14.Santiago GA, Vazquez J, Courtney S, Matias KY, Andersen LE, Colon C, et al. Performance of the Trioplex real-time RT-PCR assay for detection of Zika, dengue, and chikungunya viruses. Nat Commun. 2018;9(1):1391.

Frontier in Medical & Health Research

ISSN: (e) 3007-1607 (p) 3007-1593

15.Teoh BT, Sam SS, Tan KK, Johari J, Danlami MB, Hooi PS, et al. Early detection of dengue virus by use of reverse transcription-recombinase polymerase amplification. J Clin Microbiol. 2015;53(3):830-7.

16.Abd El Wahed A, Patel P, Faye O, Thaloengsok S, Heidenreich D, Matangkasombut P, et al. Recombinase polymerase amplification assay for rapid diagnostics of dengue infection. PLoS One. 2015;10(6):e0129682.

17.Hunsperger EA, Yoksan S, Buchy P, Nguyen VC, Sekaran SD, Enria DA, et al. Evaluation of commercially available anti-dengue virus immunoglobulin M tests. Emerg Infect Dis. 2009;15(3):436-40.

18.Hunsperger EA, Sharp TM, Lalita P, Tikomaidraubuta K, Cardoso YR, Naivalu T, et al. Use of a rapid test for diagnosis of dengue during suspected dengue outbreaks in resource-limited regions. J Clin Microbiol. 2016;54(8):2093-100.

19.Bessoff K, Delorey M, Sun W, Hunsperger E. Comparison of two commercially available dengue virus (DENV) NS1 capture enzyme-linked immunosorbent assays using a single clinical sample for diagnosis of acute DENV infection. Clin Vaccine Immunol. 2008;15(10):1513-8.

20.Gan VC, Tan LK, Lye DC, Pok KY, Mok SQ, Chua RC, et al. Diagnosing dengue at the point-of-care: Utility of a rapid combined diagnostic kit in Singapore. PLoS One. 2014;9(3):e90037.

21.Blacksell SD, Newton PN, Bell D, Kelley J, Mammen MP Jr, Vaughn DW, et al. The comparative accuracy of 8 commercial rapid immunochromatographic assays for the diagnosis of acute dengue virus infection. Clin Infect Dis. 2006;42(8):1127-34.

22.Martin DA, Muth DA, Brown T, Johnson AJ, Karabatsos N, Roehrig JT. Standardization of immunoglobulin M capture enzyme-linked immunosorbent assays for routine diagnosis of arboviral infections. J Clin Microbiol. 2000;38(5):1823-6.

23.Shu PY, Chen LK, Chang SF, Yueh YY, Chow L, Chien LJ, et al. Comparison of capture immunoglobulin M (IgM) and IgG enzyme-linked immunosorbent assay (ELISA) and nonstructural protein NS1 serotype-specific IgG ELISA for differentiation of primary and secondary dengue virus infections. Clin Diagn Lab Immunol. 2003;10(4):622-30.

24.Roehrig JT, Hombach J, Barrett AD. Guidelines for plaque-reduction neutralization testing of human antibodies to dengue viruses. Viral Immunol. 2008;21(2):123-32.

25.Thomas SJ, Nisalak A, Anderson KB, Libraty DH, Kalayanarooj S, Vaughn DW, et al. Dengue plaque reduction neutralization test (PRNT) in primary and secondary dengue virus infections: How alterations in assay conditions impact performance. Am J Trop Med Hyg. 2009;81(5):825-33.

26.Duyen HT, Ngoc TV, Ha do T, Hang VT, Kieu NT, Young PR, et al. Kinetics of plasma viremia and soluble nonstructural protein 1 concentrations in dengue: Differential effects according to serotype and immune status. J Infect Dis. 2011;203(9):1292-300.

27.Muller DA, Young PR. The flavivirus NS1 protein: Molecular and structural biology, immunology, role in pathogenesis and application as a diagnostic biomarker. Antiviral Res. 2013;98(2):192-208.

28.Priyamvada L, Hudson W, Ahmed R, Wrammert J. Humoral cross-reactivity between Zika and dengue viruses: Implications for protection and pathology. Emerg Microbes Infect. 2017;6(5):e33.