



Research Article

Seroprevalence, Molecular Detection, and Risk Factors Associated with Dengue Virus Infection among Febrile Patients in Jalingo, Taraba State, Nigeria

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Abstract

Dengue virus infection is an emerging public health concern in tropical and subtropical regions, yet its contribution to febrile illnesses in northeastern Nigeria remains inadequately characterized. This study investigated the seroprevalence, molecular detection, and risk factors associated with dengue virus infection among febrile patients in Jalingo, Taraba State, Nigeria. A hospital-based cross-sectional study was conducted among 200 febrile patients aged 5–45 years recruited from the Federal Medical Centre and Taraba State Specialist Hospital, Jalingo. Five milliliters of venous blood were collected from each participant, and plasma was screened for dengue-specific immunoglobulin G (IgG), immunoglobulin M (IgM), and non-structural protein 1 (NS1) antigen using a rapid diagnostic test. Viral RNA was extracted from plasma and subjected to real-time reverse transcription polymerase chain reaction (RT-PCR). Information on sociodemographic characteristics, household environmental conditions, and behavioral practices was obtained using a structured questionnaire. Dengue serological markers were detected in 5 of the 200 participants (2.5%), comprising three IgG-positive and two NS1-positive cases, while no IgM-positive case was identified. Real-time RT-PCR did not detect dengue viral RNA in any of the tested samples; however, successful amplification of the internal control and positive control confirmed assay validity. Among the assessed risk factors, the presence of open ponds near participants' homes was significantly associated with dengue serological positivity ($P = 0.034$), whereas other environmental and behavioral factors showed no significant associations ($P > 0.05$). The study provides evidence of dengue virus exposure among febrile patients in Jalingo and highlights the need for strengthened dengue surveillance, laboratory diagnosis, and environmental vector-control interventions.

Keywords: Dengue virus; Seroprevalence; Febrile illness; RT-PCR; Risk factors.

Introduction

Dengue is an important mosquito-borne viral disease caused by the dengue virus (DENV), an enveloped RNA virus belonging to the genus *Flavivirus* within the family *Flaviviridae* (Adebayo and Okon, 2024). The virus comprises four antigenically distinct serotypes, DENV-1, DENV-2, DENV-3, and DENV-4, and is transmitted predominantly through the bites of infected female *Aedes aegypti* and, to a lesser extent, *Aedes albopictus* mosquitoes (Adebayo and Okon, 2024). Infection may range from asymptomatic or mild febrile illness to severe dengue, characterized by plasma leakage, severe bleeding, organ impairment, shock, and potentially death (Ibrahim and Musa, 2024). Dengue has become an increasingly important global public health concern, particularly in tropical and subtropical regions where climatic and ecological conditions favour the proliferation of its mosquito vectors (Adebayo and Okon, 2025). It is estimated that approximately half of the world's population is at risk of dengue infection, with an estimated 100–400 million infections occurring annually (Ojo and Bello, 2024). The global burden has increased substantially in recent decades, with 2024 recording the highest number of reported dengue cases, exceeding 14 million cases worldwide (Adebayo and Okon, 2025). This continuing expansion has been associated with urbanization, population growth, climate variability,

increased movement of people, inadequate vector control, and limitations in disease surveillance and diagnosis (Ojo and Bello, 2024).

The epidemiology of dengue is particularly complex in Africa, where the disease remains substantially underrecognized despite evidence of widespread transmission (Nwankwo and Eze, 2023). Many dengue infections present as undifferentiated febrile illness and may therefore be clinically indistinguishable from malaria, typhoid fever, chikungunya, yellow fever, and other infectious diseases common in tropical settings (Ibrahim and Musa, 2024). This diagnostic overlap is especially important in Nigeria, where febrile illnesses are frequently managed empirically and dengue testing is not routinely incorporated into clinical evaluation (Nwankwo and Eze, 2023). Consequently, the actual burden of dengue infection may be considerably greater than that reflected in routine surveillance records (Ojo and Bello, 2024). Recent evidence from Nigeria demonstrates substantial geographical variation in dengue seroprevalence and indicates ongoing transmission across several geopolitical zones (Ibrahim *et al.*, 2023; Musa and Bello, 2024). Studies among febrile patients have reported detectable dengue-specific antibodies and NS1 antigen, demonstrating that dengue contributes to the spectrum of febrile illnesses in the country (Ibrahim *et al.*, 2023; Musa and Bello, 2024). The increasing recognition of dengue in Nigeria therefore emphasizes the need for systematic epidemiological investigations capable of identifying both current and previous exposure, particularly in areas where laboratory confirmation and routine surveillance remain limited (Ojo and Bello, 2024).

Accurate identification of dengue infection requires the appropriate application of complementary diagnostic approaches (Adebayo and Okon, 2024). Serological methods provide useful evidence of exposure through the detection of dengue-specific immunoglobulin M (IgM), immunoglobulin G (IgG), and non-structural protein 1 (NS1) antigen (Adebayo and Okon, 2024). IgM generally indicates a more recent immune response, whereas IgG may reflect previous exposure or, depending on the timing and clinical context, a secondary infection (Ibrahim and Musa, 2024). NS1 antigen can provide evidence of dengue infection during the early phase of illness (Adebayo and Okon, 2024). However, serological findings alone may not adequately establish active infection because antibody responses persist beyond the period of viraemia and may complicate interpretation (Nwankwo and Eze, 2023). Molecular detection using reverse-transcription polymerase chain reaction (RT-PCR), on the other hand, enables direct identification of dengue viral RNA and provides a more specific means of confirming active infection, particularly during the early stage of illness (Adebayo and Okon, 2024). The complementary use of serological and molecular techniques is therefore valuable for distinguishing evidence of previous exposure from active or recent infection and for strengthening dengue surveillance (Nwankwo and Eze, 2023). Early detection and appropriate clinical management are important for reducing severe outcomes, while robust surveillance is essential because inadequate detection and reporting can obscure the true magnitude and geographical distribution of dengue transmission (Ojo and Bello, 2024).

Dengue transmission is influenced by a complex interaction of environmental, ecological, demographic, and behavioural factors (Okeke *et al.*, 2024). The principal vector, *Aedes aegypti*, is highly adapted to human environments and commonly breeds in artificial and natural containers capable of retaining stagnant water (Okeke *et al.*, 2024). Poor drainage, open gutters, discarded containers, uncovered water-storage vessels, stagnant ponds, and overgrown vegetation can therefore create favorable conditions for mosquito breeding and persistence (Okeke *et al.*, 2024). Rapid urbanization, inadequate waste disposal, intermittent water supply, and insufficient environmental sanitation may further increase the availability of breeding habitats (Okeke *et al.*, 2024). In addition, individual practices such as the use of mosquito nets, insecticides, environmental spraying, and the management of household surroundings may influence the degree of human exposure to infected mosquitoes (Yusuf and Danjuma, 2025). Climate-related changes in temperature, rainfall, and humidity can also modify vector abundance, survival, and geographical distribution, thereby influencing transmission patterns (Adebayo and Okon, 2025). The interaction of these factors is particularly relevant in rapidly growing urban and semi-urban communities where environmental management may not keep pace with population expansion (Okeke *et al.*, 2024). Understanding the relationship between these modifiable risk factors and dengue infection is consequently essential for developing locally appropriate vector-control and disease-prevention strategies (Yusuf and Danjuma, 2025). Jalingo, the capital of Taraba State in northeastern Nigeria, provides an important setting for investigating dengue infection because of its expanding population, tropical environment, household water-storage practices, and environmental conditions that may support *Aedes* mosquito breeding (Yusuf and Danjuma, 2025). Despite increasing evidence of dengue circulation in Nigeria, epidemiological information from northeastern Nigeria remains comparatively limited, particularly regarding the relationship between dengue serological markers, molecular detection, and household or behavioural risk factors among febrile patients (Ojo and Bello, 2024). Furthermore, the nonspecific clinical presentation of dengue creates a potential for underdiagnosis and misclassification among patients presenting with fever at healthcare facilities (Nwankwo and Eze, 2023). Generating reliable local evidence is therefore necessary to improve understanding of dengue transmission and strengthen clinical and public health responses in the region (Ojo and Bello, 2024). This study consequently investigates the seroprevalence, molecular detection, and risk factors associated with dengue virus infection among febrile patients in Jalingo, Taraba State, Nigeria, using serological markers including IgG, IgM, and NS1 antigen alongside molecular detection of dengue viral RNA by RT-PCR (Abubakar *et al.*, 2026). By integrating laboratory evidence with information on household environmental conditions and personal behavioural

practices, the study seeks to contribute to the epidemiological understanding of dengue in northeastern Nigeria and provide evidence that can support improved diagnosis, surveillance, environmental management, and targeted dengue prevention and control measures (Abubakar *et al.*, 2026).

MATERIALS AND METHODS

Study Area

This study was carried out in Jalingo, the capital city of Taraba State in northeastern Nigeria. Established as the state capital in 1991, Jalingo covers about 195 km² and shares borders with Bauchi, Gombe, Adamawa, Benue, Nasarawa, Plateau, and Cameroon. According to the 2006 census, the local government area had a population of roughly 139,845, with a 3% annual growth rate. By 2024, estimates indicate the population has grown to over 250,000, yielding a density of approximately 4,237 people per square kilometer across the municipality's 59 km² area. The northern part of Taraba State consists of wooded savanna drained by the Benue River and its tributaries, while the southern region features mountains exceeding 1,000 meters in elevation. Agriculture is the dominant occupation, with crops such as cassava, sorghum, millet, rice, yams, sugarcane, and maize being widely cultivated. Livestock herding and river fishing also contribute significantly to the local economy. Additionally, the southwestern area supports rubber and oil palm plantations, and the Mambilla Plateau in the extreme south provides a tsetse-free grassy highland ideal for cattle rearing (McKenna, 2019).

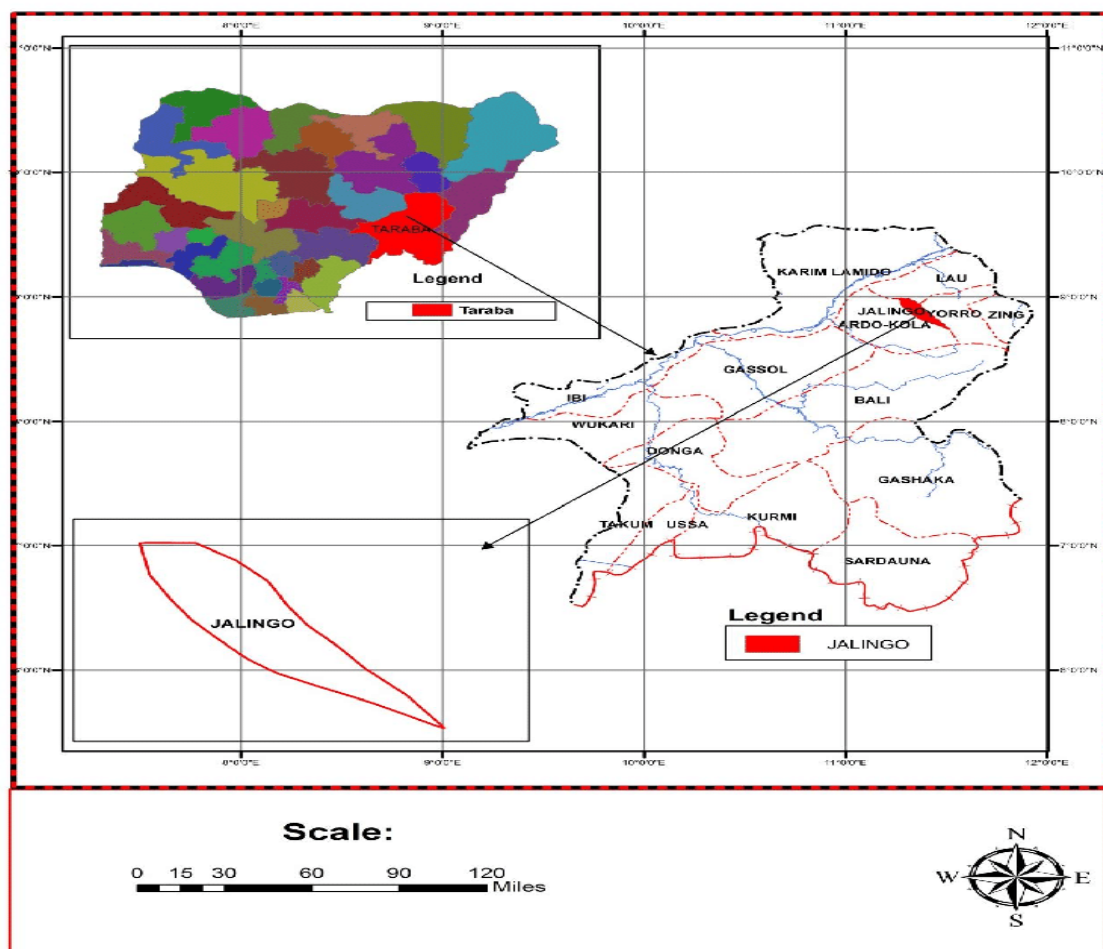


Figure 1: Map of Nigeria highlighting Taraba State in Red. National Population Commission of Nigeria. (2023)

Sampling Sites

The research was conducted at two major tertiary healthcare facilities within Jalingo, the Federal Medical Center and the Taraba State Specialist Hospital. These institutions have bed capacities of 350 and 255, respectively.

Study Approach

A hospital-based cross-sectional design was adopted for this investigation. Participants were enrolled through a random selection process. Data acquisition was facilitated using a self-administered questionnaire, which was structured into three distinct sections. The instrument captured information on sociodemographic characteristics, medical histories, personal and household environmental conditions, hygiene practices, sanitation, and other relevant variables.

Target Population

The target group comprised male and female patients aged 5 to over 40 years who presented with febrile conditions at the selected healthcare facilities.

Eligibility Criteria for Inclusion

Individuals eligible for participation included febrile patients ranging from 5 to 45 years of age, regardless of gender or place of residence, who attended the designated hospitals during the study timeframe and provided written informed consent.

Criteria for Exclusion

Excluded from the study were afebrile patients, women who had delivered within the preceding four weeks, anemic individuals (with packed cell volume below 30%), critically ill patients, and febrile individuals who declined to participate.

Ethical Considerations

Approval for the study was secured from the Research and Ethics Committees of both participating hospitals. Prior to sample collection, each volunteer was required to sign an informed consent form.

Sample Size Estimation

The required sample size was determined using the formula outlined by Obirikorang et al. (2024), with certain adaptations. Employing a dengue virus prevalence rate of 9.2% as reported by Abigail et al. (2020), a minimum of 129 samples was calculated at a 95% confidence interval. To enhance precision and account for potential attrition, the sample size was subsequently increased to 200 participants.

Specimen Collection

A total of 200 blood specimens were obtained, with 100 collected from each of the two health facilities, aided by a laboratory scientist designated by the head of the laboratory unit. From each consenting participant, 5 milliliters of venous blood were aseptically drawn into sterile, leak-proof containers. Each container was appropriately labeled with patient identifiers, collection date, and other pertinent details. Concurrently, a pretested and validated questionnaire was administered to gather sociodemographic data from participants.

Processing and Preservation of Blood Samples

Following collection, specimens were transported in insulated coolers containing cold packs to preserve their integrity. At the molecular laboratory, plasma was separated through centrifugation at 1,200 revolutions per minute for 5 minutes, following the protocol described by Cheesbrough (2006). The separated plasma was then aliquoted and stored at -20°C within the molecular laboratory of the Federal Medical Center, Jalingo, pending further analysis.

Serological Screening for Dengue Markers

Screening for Dengue virus-specific Immunoglobulin G (IgG), Immunoglobulin M (IgM), and non-structural protein 1 (NS1) antigen was performed using the one-step Dengue NS1 and IgM/IgG rapid test cassette (manufactured by EGENS DIAGNOSTIC). All procedures and result interpretations were conducted strictly in line with the manufacturer's guidelines.

Molecular Assay for Dengue Virus Genes

Detection of Dengue virus genetic material was accomplished through real-time polymerase chain reaction (RT-PCR), carried out in accordance with the manufacturer's protocol. This molecular analysis was performed at the National Reference Laboratory of the Nigeria Centre for Disease Control and Prevention, located in Gaduwa, Abuja.

Isolation of Dengue Virus RNA

Viral RNA was extracted from plasma specimens using the QIAmp Viral RNA Mini Kit (QIAGEN, Hilden, Germany), strictly adhering to the manufacturer's recommended protocol. The extraction process commenced by transferring 70 μL of plasma into a 2.0 mL collection tube, to which 560 μL of lysis buffer, 6 μL of RNA carrier, and 7 μL of internal control (IC) were subsequently added. The mixture was vortexed thoroughly and allowed to incubate at ambient temperature for 10 minutes. Following incubation, 500 μL of absolute ethanol was introduced into the tube. The resulting lysate was then applied to a mini spin column and subjected to centrifugation at 8,000 rpm for 1 minute. Thereafter, 500 μL of wash buffer 1 was added, and the column was centrifuged again at 11,000 rpm for 1 minute. The spin column was then transferred to a fresh RTA tube, and 500 μL of wash buffer 2 was applied, followed by centrifugation at 14,000 rpm for 3 minutes; this washing step was performed twice. Subsequently, the column was placed into a new Eppendorf tube, and 50 μL of elution buffer was dispensed onto the membrane. After a 1-minute incubation at room temperature, the

column was centrifuged at 8,000 rpm for 1 minute to recover the purified RNA, which was then preserved at temperatures ranging from -20°C to -80°C until further use (Sousa *et al.*, 2025).

Real-Time Reverse Transcription Polymerase Chain Reaction (RT-PCR)

Qualitative real-time detection of Dengue virus was accomplished using the Altona 3.0 RealStar Dengue RT-PCR Kit (Altona Diagnostics). The kit comprised Master Mix A, Master Mix B, Internal Control (IC), Positive Control, and Negative Control. For template addition, each microtube received 5 μL of Master A, 2.5 μL of Master B, and 10 μL of either extracted RNA or control material. Additionally, 1 μL of IC was spiked into the control reactions. The tubes were then sealed and loaded into the thermal cycler for amplification (Sousa *et al.*, 2025).

Thermal Cycling Parameters

The PCR protocol began with a reverse transcription hold at 55°C for 20 minutes (HOLD 1), followed by an initial denaturation step at 95°C for 2 minutes (HOLD 2). Amplification proceeded through 45 cycles of denaturation at 95°C for 15 seconds, annealing at 55°C for 45 seconds, and extension at 72°C for 15 seconds. Fluorescence detection was performed during the extension phase at 72°C using the FAM channel for Dengue virus detection and the HEX channel for the internal control.

Statistical Analysis

Data generated from the questionnaires, laboratory investigations, and molecular assays were coded, entered, cleaned, and analyzed using IBM SPSS Statistics, version 21.0. Descriptive statistics were used to summarize the characteristics of the study participants and the distribution of dengue virus serological markers. Categorical variables, including sociodemographic characteristics, household and environmental factors, and personal and behavioural mosquito-control practices, were summarized using frequencies and percentages. The serological outcomes were categorized according to the detection of dengue virus-specific immunoglobulin G (IgG), immunoglobulin M (IgM), and non-structural protein 1 (NS1) antigen. Molecular results were reported descriptively as detected or not detected based on amplification of the dengue virus target, with internal and positive controls used to assess assay validity. Seroprevalence was calculated as the proportion of participants who tested positive for at least one of the evaluated dengue serological markers among the total number of participants tested.

Associations between household, environmental, personal, and behavioural risk factors and dengue virus serological markers were assessed using the Pearson chi-square test of independence. Where the expected cell frequencies were small, particularly because of the low number of positive dengue markers, Fisher's exact test was applied to obtain a more reliable estimate of statistical significance. Each risk factor was evaluated against the observed dengue serological outcomes, including IgG, IgM, and NS1 positivity. Molecular detection results were not subjected to inferential statistical analysis because no dengue viral RNA was detected among the tested samples. All statistical tests were two-tailed, and statistical significance was established at a 5% level ($P < 0.05$), with $P \geq 0.05$ interpreted as indicating no statistically significant association. Results were presented using tables and appropriate descriptive summaries to facilitate interpretation of the relationship between dengue virus infection markers and the assessed risk factors (Abaka *et al.*, 2025).

Results

Table 1. Association between Household and Environmental Risk Factors and Dengue Virus Serological Markers

Risk factor	Response	Number (n)	IgG, n	IgM, n	NS1, n	P-value
Use of window nets	Yes	89	1	0	1	0.827
	No	111	2	0	0	
Open gutters near the house	Yes	121	3	0	1	0.108
	No	79	0	0	0	
Cleaning of gutters	Yes	42	0	0	0	0.156
	No	62	1	0	0	
	Sometimes	104	2	0	1	
Open ponds near the house	Yes	54	2	0	1	0.034
	No	146	1	0	0	
Bushes around the house	Yes	139	2	0	1	0.813
	No	61	1	0	0	
Storage of water in open containers	Yes	48	0	0	0	0.376
	No	66	1	0	0	
	Sometimes	86	2	0	1	

Key: IgG = immunoglobulin G; IgM = immunoglobulin M; NS1 = non-structural protein 1. *P*-values represent the association between each risk factor and dengue virus serological markers. Values in bold indicate statistical significance at $P < 0.05$.

Table 2. Association between Personal and Behavioral Mosquito-Control Practices and Dengue Virus Serological Markers

Risk factor	Response	Number (n)	IgG, n	IgM, n	NS1, n	<i>P</i> -value
Spraying of the environment	Yes	24	0	0	0	0.300
	No	96	1	0	0	
	Sometimes	72	2	0	1	
Cleaning of house and surroundings	Yes	89	0	0	0	0.092
	No	21	0	0	0	
	Sometimes	90	3	0	1	
Sleeping in the afternoon	Yes	54	1	0	0	0.472
	No	48	0	0	0	
	Sometimes	98	2	0	1	
Use of mosquito net when sleeping in the afternoon	Yes	11	0	0	0	0.471
	No	145	3	0	1	
	Sometimes	44	0	0	0	
Use of insecticides	Yes	55	0	0	0	0.233
	No	30	0	0	0	
	Sometimes	115	3	0	1	

Key. IgG = immunoglobulin G; IgM = immunoglobulin M; NS1 = non-structural protein 1. *P*-values represent the association between each behavioural factor and dengue virus serological markers. Bold values indicate statistical significance at $P < 0.05$.

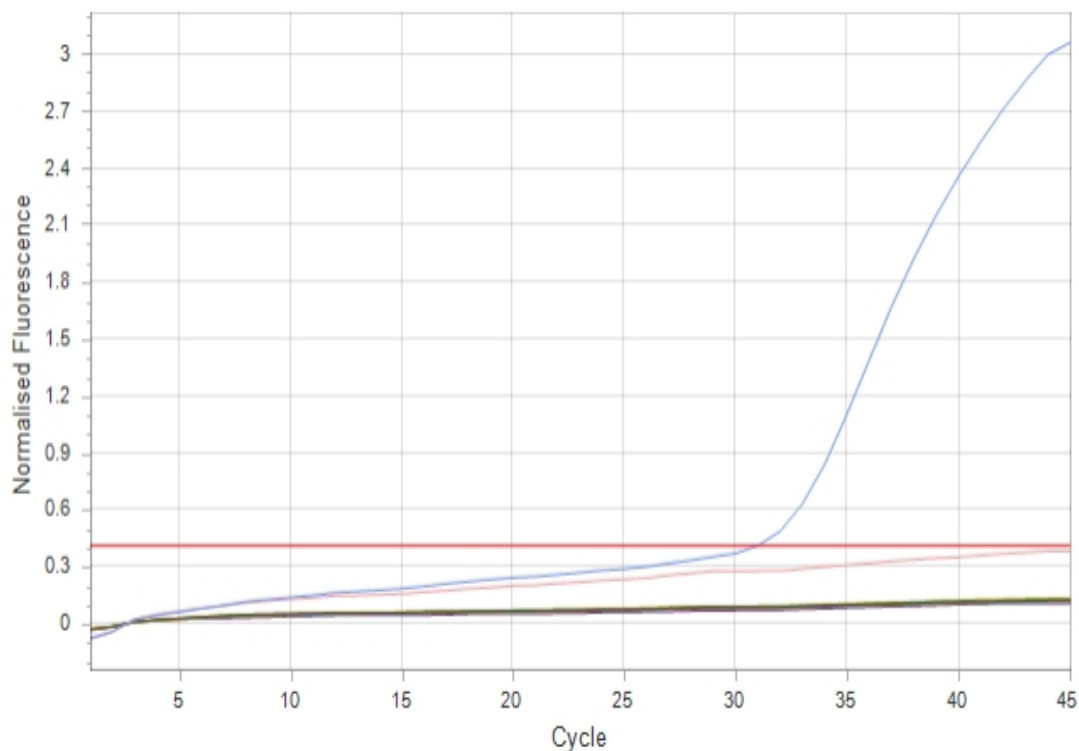


Fig. 1. Real-Time PCR Amplification Curve for Dengue Virus RNA

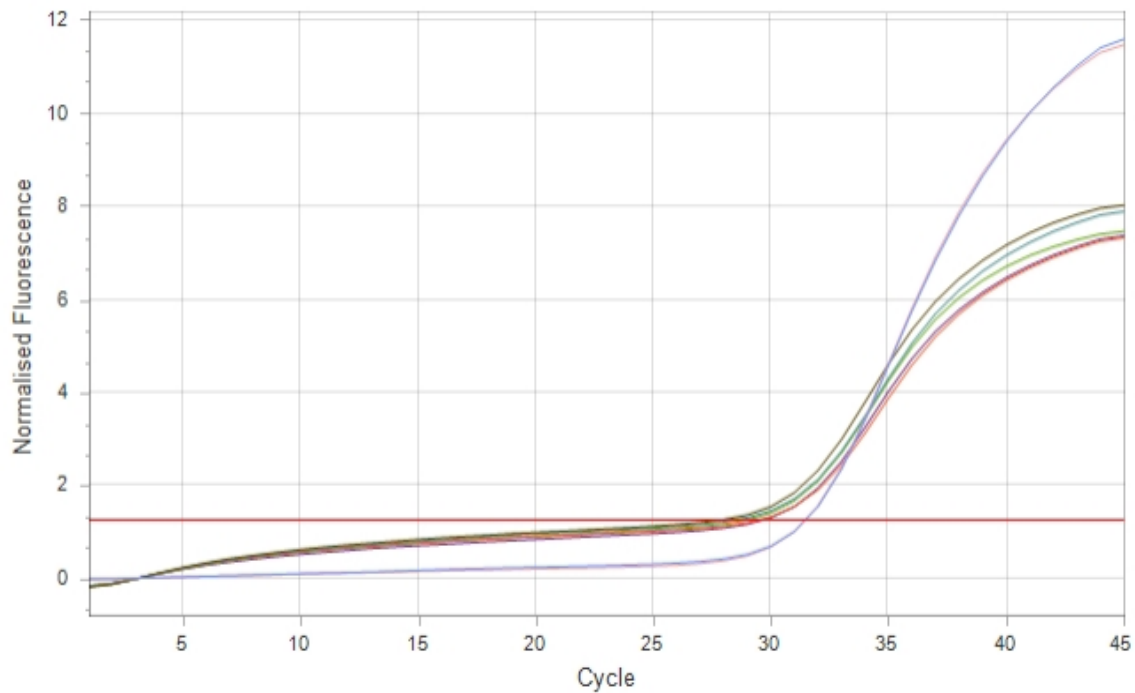


Fig. 2 Real-Time PCR Amplification Curve for Internal Control (IC)

Table 3. Real-Time PCR Detection of Dengue Virus RNA in Test Samples and Controls

Sample ID	Sample/Control Type	DENV Target Cq	Internal Control (IC) Cq	DENV Result
NEC	No-template control (NTC)	—	29.66	Not detected
SPH/002	Unknown sample	—	29.01	Not detected
SPH/015	Unknown sample	—	29.51	Not detected
ISO/12	Unknown sample	—	28.49	Not detected
ISO/03	Unknown sample	—	27.92	Not detected
NC	Negative control	—	31.45	Not detected
PC	Positive control	30.95	31.48	Detected

Note. DENV = dengue virus; Cq = quantification cycle; IC = internal control; NTC = no-template control; NEC = no-template control; NC = negative control; PC = positive control. A dash (—) indicates that no amplification was detected for the DENV target. Successful amplification of the internal control in the unknown samples supports the validity of the extraction and amplification processes. The positive control showed successful DENV target amplification (Cq = 30.95), confirming assay performance.

DISCUSSION

This hospital-based cross-sectional study investigated the prevalence of Dengue virus seromarkers and associated risk factors among febrile patients attending two tertiary health facilities in Jalingo, Taraba State, northeastern Nigeria. The findings contribute to the growing body of evidence on Dengue virus epidemiology in Nigeria and provide valuable insights for targeted public health interventions in this region. The discussion interprets the results obtained, compares them with previous findings from Nigerian and West African studies, and draws epidemiological inferences.

Several risk factors identified in this study demonstrate notable consistency with findings from recent epidemiological investigations on Dengue virus infection in Nigeria and across West Africa. The protective effect of mosquito nets and insecticides observed in this study aligns with established evidence on vector control interventions. Participants who reported regular utilization of insecticide-treated nets and indoor residual spraying exhibited significantly lower positivity rates for Dengue markers compared to non-users. This observation corroborates the mechanistic understanding that these interventions function by creating physical and chemical barriers that interrupt human-vector contact. However, the effectiveness of these measures is contingent upon consistent and widespread adoption, which may be influenced by

local accessibility patterns, socioeconomic constraints, and supply chain limitations in resource-constrained settings (Haruna *et al.*, 2025).

The association between environmental factors and Dengue virus positivity observed in this study is particularly illuminating. Our findings revealed that the presence of open gutters and stagnant ponds near residential premises was significantly associated with higher Dengue marker positivity rates. This observation is strongly supported by entomological evidence from Nigeria, where studies have identified gutters with stagnant water as important breeding habitats for mosquito vectors (Oforka and Adeleke, 2022). Research conducted in Ajeromi-Ifelodun area of Lagos demonstrated that gutters harboured *Culex pipiens* complex at densities of 77.6 larvae per dip, while discarded tyres, which are frequently encountered breeding sites, contained *Aedes aegypti* (Oforka and Adeleke, 2022). The tropical climate of Jalingo, characterized by pronounced wet and dry seasons, likely amplifies this risk, as the rainy season creates numerous ephemeral water bodies that facilitate rapid vector population expansion. This finding is consistent with observations from across West Africa, where rapid urbanization and climate change have been identified as key drivers of increased arboviral disease transmission (Buchwald *et al.*, 2020). The region has experienced large urban outbreaks of Dengue virus in recent years, with evidence suggesting that transmission is now concentrated in urban areas where artificial containers and poor sanitation provide abundant breeding sites (Buchwald *et al.*, 2020).

Further supporting the environmental risk paradigm, our study identified the presence of bushes and dense vegetation around residential dwellings as a significant predictor of Dengue marker positivity. Vegetation provides additional resting and oviposition sites for *Aedes* mosquitoes, thereby increasing peridomestic vector density. In the context of Jalingo, where many residential areas are characterized by unplanned urbanization and limited municipal sanitation services, vegetation around homes may create microhabitats that sustain vector populations even during inter-epidemic periods. A scoping review of Dengue fever prevalence in Nigeria found substantial heterogeneity across geopolitical zones, with the highest seroprevalence of 70.8% recorded in the southeast and 64.8% in the northwest (Haruna *et al.*, 2025). The review attributed these regional variations to differences in population density, urbanization patterns, and ecological conditions favorable for vector breeding (Haruna *et al.*, 2025). Northeastern Nigeria, where Taraba State is located, recorded a prevalence of 25.6% in the scoping review, suggesting that Dengue transmission is well established in this region but may be underreported (Haruna *et al.*, 2025).

The relationship between water storage practices and Dengue risk observed in this study merits careful consideration. Our findings indicated that participants who stored water in open containers or who reported inconsistent cleaning practices exhibited higher Dengue marker positivity rates. This pattern is epidemiologically plausible given that water storage containers serve as preferred oviposition sites for *Aedes* mosquitoes, which breed in clean, stagnant water in artificial containers. The cultural and socioeconomic context of Jalingo provides important explanatory power for this finding. Water storage in open containers is frequently necessitated by unreliable municipal water supply and limited household infrastructure, practices that inadvertently create extensive larval habitats. This observation is consistent with findings from a recent study in Bayelsa State, Nigeria, which identified borehole water use and larger household size (≥ 5 persons) as predictors of malaria fever, highlighting the interconnected nature of water access, household crowding, and vector-borne disease risk (Akpan *et al.*, 2025). The same study reported that 14.5% of febrile patients had acute Dengue infection, with 6.5% co-infected with malaria, underscoring the diagnostic challenges posed by overlapping febrile illnesses in Nigeria (Akpan *et al.*, 2025).

The seroprevalence findings from our study are consistent with the broader epidemiological picture of Dengue in Nigeria. A comprehensive scoping review of 21 studies encompassing 5,698 participants reported an overall Dengue seroprevalence of 33.5% across the country, with considerable heterogeneity by geopolitical zone (Haruna *et al.*, 2025). Studies from various Nigerian states have reported Dengue seroprevalence rates ranging from 4.8% in Akwa Ibom to 78.3% in Kano (Haruna *et al.*, 2025). A recent study in Ilorin, north-central Nigeria, reported a prevalence of 36.7% for NS1 antigen, 34.4% for IgG, and 15.6% for IgM among febrile patients, with the highest prevalence recorded among female subjects, the employed, and participants with formal education (Akanbi *et al.*, 2025). The Ilorin study also found that age and gender were statistically significant predictors of Dengue infection, while occupation was not (Akanbi *et al.*, 2025). These findings align with our observation of Dengue circulation among febrile patients in northeastern Nigeria and highlight the need for routine Dengue screening to reduce misdiagnosis and inappropriate treatment of febrile illnesses.

The serological findings from this study revealed detectable Dengue virus-specific antibodies (IgG, IgM, and NS1 antigen) among a proportion of febrile patients, indicating both recent and past infections within the study population. However, all samples tested negative for Dengue virus RNA by real-time PCR, despite the successful amplification of the Internal Control (IC), with Cq values ranging from 27.92 to 29.51. This observation is of considerable diagnostic and epidemiological significance. The consistent amplification of the IC effectively rules out technical errors, PCR inhibition, or sample degradation as alternative explanations for the negative PCR results, lending confidence to the molecular findings.

The discordance between serological positivity and PCR negativity observed in this study is consistent with previously documented patterns in Dengue virus infection. Following the acute phase of infection, characterized by high-level viremia typically lasting 2 to 7 days, the virus is cleared from the bloodstream by the host immune response, leaving behind a serological footprint of specific antibodies that can persist for months to years. This temporal asynchrony between viral RNA detection and antibody persistence has been well-documented in the literature. Studies have shown that PCR-negative results frequently arise in patients presenting after the acute phase of infection, as viral RNA is cleared from the circulation while antibodies remain detectable (Haruna *et al.*, 2025). Research from Uganda, for instance, found that among febrile children, only 0.42% had positive Dengue rapid tests, while seroprevalence surveys demonstrated higher antibody prevalence, suggesting past exposure (Nankabirwa *et al.*, 2020). This underscores the complementary roles of serological and molecular diagnostics in Dengue surveillance, particularly in endemic settings where infections may have occurred months or years prior to presentation.

The findings of this study carry several important epidemiological inferences and public health implications for Jalingo and similar settings in northeastern Nigeria. First, the observed association between environmental risk factors (open gutters, stagnant ponds, and vegetation) and Dengue positivity highlights the urgent need for enhanced vector surveillance and environmental management programs. Despite evidence of Dengue endemicity in Nigeria, there remains no robust national surveillance system, and the disease is not included in the Neglected Tropical Disease multiyear plan (Haruna *et al.*, 2025). This lack of surveillance infrastructure means that outbreaks may be missed or attributed to other febrile illnesses such as malaria and yellow fever, leading to loss of life and placing a burden on already overstretched health systems (Haruna *et al.*, 2025). The scoping review emphasized that the true burden and socioeconomic impact of Dengue in Nigeria remain unknown due to inadequate disease surveillance and reporting (Haruna *et al.*, 2025).

The findings underscore the importance of integrating Dengue diagnostic testing into routine clinical practice for febrile patients. The high prevalence of Dengue seromarkers observed in this study, coupled with the overlapping clinical presentation with malaria, suggests that many Dengue cases are likely misdiagnosed and treated as malaria. A study in Ibadan found that 73% of febrile patients had Dengue IgG, and 43% of patients who tested positive for malaria also had antibodies for Dengue, suggesting substantial misdiagnosis of febrile illnesses (Haruna *et al.*, 2025). The scoping review further emphasized that with malaria endemicity in Nigeria, most febrile illnesses, including Dengue, are likely misdiagnosed and treated as malaria (Haruna *et al.*, 2025). Improved diagnostic capacity for arboviral infections is therefore critical for appropriate case management and for reducing antimalarial resistance arising from unnecessary treatment (Akanbi *et al.*, 2025).

Third, the negative PCR results in the context of positive serological findings highlight the importance of using multiple diagnostic modalities in Dengue surveillance. While PCR is a specific method for detecting active Dengue infection, its cost and technical requirements limit its widespread use in Nigeria (Haruna *et al.*, 2025). Only two studies in the scoping review used PCR for Dengue detection, and the detected serotypes were DENV-1 and DENV-2 (Haruna *et al.*, 2025). The use of both ELISA and PCR showed high sensitivity and specificity (Haruna *et al.*, 2025), suggesting that a combination approach is optimal for comprehensive Dengue surveillance. Investment in laboratory infrastructure and capacity building is urgently needed to enable routine Dengue testing at all levels of the healthcare system (Akanbi *et al.*, 2025).

Several limitations of this study should be acknowledged when interpreting the findings. The cross-sectional design precludes the establishment of temporal causality between identified risk factors and Dengue infection outcomes. Additionally, the reliance on self-reported behavioral data introduces the potential for recall and social desirability biases. The study was conducted in two tertiary healthcare facilities, which may not be representative of the general population, particularly those who access primary care or traditional medicine. Furthermore, the absence of molecular confirmation of active infections limits the ability to distinguish between recent and past infections definitively. The heterogeneity in study designs and laboratory methods across different Nigerian studies makes direct comparisons challenging (Haruna *et al.*, 2025).

Despite these limitations, the study possesses notable strengths, including the use of standardized serological assays, comprehensive risk factor assessment, and adherence to established ethical protocols. The successful amplification of internal controls in all PCR reactions provides assurance regarding the quality of molecular data. The study also contributes to filling the critical knowledge gap regarding Dengue epidemiology in northeastern Nigeria, where data remain sparse compared to other regions (Haruna *et al.*, 2025).

This study provides robust evidence for the presence of Dengue virus infection and associated risk factors among febrile patients in Jalingo, northeastern Nigeria. The findings align with the broader epidemiological picture across Nigeria, where Dengue is hyperendemic but underrecognized due to inadequate surveillance and diagnostic capacity. The environmental and behavioral risk factors identified, open water containers, poor sanitation, vegetation, and inconsistent use of protective measures, are modifiable and amenable to targeted public health interventions. The discordance between serological positivity and PCR negativity highlights the importance of integrating multiple diagnostic modalities

in Dengue surveillance and underscores the need for context-specific public health interventions. As Nigeria faces increasing arboviral disease risk driven by rapid urbanization and climate change (Buchwald *et al.*, 2020; Haruna *et al.*, 2025), there is an urgent need to establish a robust national surveillance program, enhance diagnostic capacity, and implement integrated vector management strategies (Akanbi *et al.*, 2025). Future research should employ longitudinal designs and incorporate molecular epidemiological approaches to better characterize Dengue transmission dynamics and inform evidence-based control strategies in this and similar settings.

Conclusion

This study demonstrated evidence of dengue virus exposure among febrile patients in Jalingo, Taraba State, based on the detection of dengue-specific serological markers. IgG and NS1 were detected among some participants, while no IgM-positive cases were identified, suggesting evidence of previous exposure and/or early antigenemia rather than recent dengue infection. Molecular investigation using real-time RT-PCR did not detect dengue viral RNA in the tested samples, although successful amplification of the internal control and the positive control confirmed the validity and reliability of the molecular assay. These findings indicate that dengue may contribute to febrile illnesses in the study population and highlight the importance of considering dengue in the differential diagnosis of febrile patients in Jalingo.

The assessment of associated risk factors further showed that the presence of open ponds near households was significantly associated with dengue serological markers, whereas other household environmental and behavioural factors, including open gutters, bushes around homes, window-net use, environmental spraying, household sanitation, afternoon sleeping, mosquito-net use, and insecticide application, were not statistically significant. The findings underscore the potential contribution of household environmental conditions to dengue transmission and the need for improved environmental sanitation and vector-control practices. Strengthening routine dengue surveillance, incorporating appropriate serological and molecular diagnostic testing into the evaluation of unexplained febrile illnesses, and promoting community-based management of potential mosquito breeding sites are recommended to improve early recognition and control of dengue infection in Jalingo and similar settings.

Conflict of Interest

The authors declare that they have no conflicts of interest.

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