

To Estimate the Prevalence of Dengue at Tertiary Care Center in Darbhanga

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Received: 01-01-2026 / Revised: 15-02-2026 / Accepted: 21-03-2026

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Conflict of interest: Nil

Abstract

Background: Dengue fever is a mosquito-borne viral disease caused by dengue virus (DENV) with four distinct serotypes (DENV-1, DENV-2, DENV-3, and DENV-4). It poses a significant public health burden globally, particularly in tropical and subtropical regions. This study aimed to estimate the prevalence of dengue infection at a tertiary care center in Darbhanga, Bihar, India.

Methods: A clinic-based prospective study was conducted at Darbhanga Medical College and Hospital (DMCH) from September 2022 to April 2024. A total of 1076 blood samples from suspected dengue patients were collected and tested using enzyme-linked immunosorbent assay (ELISA) for NS1 antigen, IgM, and IgG antibodies.

Results: Out of 1076 patients tested, the overall seroprevalence of dengue was 13.10% (141 cases). Among the positive cases, NS1 antigen was detected in 86 (7.80%) patients, IgM antibodies in 46 (4.28%) patients, and IgG antibodies in 9 (0.83%) patients. The highest incidence was observed in the 21-30 years age group (31.50%), followed by 11-20 years (28.44%). Males (55.58%) were more commonly affected than females (44.42%). Fever was the most common presenting symptom (33.27%).

Conclusion: The study demonstrates a significant prevalence of dengue infection in the Darbhanga region. The higher prevalence among young adults and males highlights the need for targeted preventive measures. Early serological diagnosis using ELISA remains crucial for effective management and control of dengue outbreaks.

Keywords: Dengue fever, ELISA, IgM antibodies, IgG antibodies, NS1 antigen.

DOI: 10.25258/ijcpr.18.4.138

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Introduction

Dengue fever is a viral disease transmitted by mosquitoes of the *Aedes* genus, primarily *Aedes aegypti* and *Aedes albopictus*. It has emerged as a significant global health concern, particularly in tropical and subtropical regions. According to the World Health Organization (WHO), approximately 390 million dengue infections occur annually worldwide, with nearly 50% of the global population at risk of contracting this disease. [1,2]

The dengue virus belongs to the family Flaviviridae and genus Flavivirus, comprising four distinct but closely related serotypes: DENV-1, DENV-2, DENV-3, and DENV-4. Infection with one serotype provides lifelong immunity against that specific serotype but only partial and temporary protection against the others. Secondary infection

with a different serotype increases the risk of severe dengue, including dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS), due to antibody-dependent enhancement. [3-5] The clinical presentation of dengue fever ranges from asymptomatic infection to mild febrile illness and severe, life-threatening complications. Common symptoms include high fever, severe headache, retro-orbital pain, muscle and joint pain, rash, and mild bleeding manifestations.

The incubation period typically ranges from 4 to 10 days following the bite of an infected mosquito. [6-8] India has witnessed a significant increase in dengue cases over the past few decades, with Bihar being one of the endemic states. The state experiences periodic outbreaks, particularly during

the monsoon and post-monsoon seasons when vector breeding conditions are favorable. The Darbhanga region, located in northern Bihar, presents unique epidemiological characteristics due to its geographical location, climate patterns, and demographic factors. [9-11] Laboratory diagnosis of dengue infection relies on the detection of viral components or host immune response. The NS1 antigen test detects non-structural protein 1, which is secreted by infected cells during the early phase of infection. Serological tests for IgM and IgG antibodies help distinguish between recent and past infections. The IgM antibodies typically appear 3-5 days after symptom onset and persist for 30-60 days, while IgG antibodies develop later and may persist for years. [12,13]

This study was conducted to estimate the prevalence of dengue infection at Darbhanga Medical College and Hospital, a tertiary care center serving the Darbhanga region. The objectives were to determine the seroprevalence of dengue using antigen and antibody detection tests, understand the demographic distribution of cases, and assess the clinical presentation patterns among suspected patients.

Materials and Methods

Study Design and Setting: This was a clinic-based prospective observational study conducted at the Department of Microbiology, Darbhanga Medical College and Hospital (DMCH), Laheriasarai, Bihar, India, over a period of 18 months from September 2022 to February 2024.

The study was approved by the institutional research ethics committee.

Study Population: A total of 1076 patients suspected of having dengue fever were included in the study. Blood samples were collected from patients presenting to the virology laboratory from various departments, including medicine, as well as from individuals visiting the blood bank during the study period.

Inclusion Criteria

- Individuals of all age groups
- Symptomatic and asymptomatic patients
- Patients with acute onset of fever lasting more than 4 days
- Clinically suspected cases of dengue virus infection

Exclusion Criteria

- Patients with trauma
- Patients with proven febrile illnesses such as malaria, typhoid, congestive cardiac failure, or renal disease

Sample Collection and Processing: Blood samples (3-5 mL) were collected aseptically from each patient in sterile vacutainers. The samples were allowed to clot at room temperature and then centrifuged at 1500 rpm for 15 minutes to separate serum. Serum samples were stored at 4-8°C for up to one week or at -20°C for longer periods until testing. Repeated freeze-thaw cycles were avoided to preserve antibody integrity.

Laboratory Methods: All serum samples were tested using commercially available ELISA kits according to the manufacturer's instructions. The following parameters were assessed:

NS1 Antigen Detection: The NS1 antigen capture ELISA was performed to detect dengue virus NS1 protein, indicating acute or early infection.

IgM Antibody Detection: IgM antibody capture ELISA (MAC-ELISA) was performed to detect recent dengue infection. IgM antibodies typically appear 3-5 days after symptom onset.

IgG Antibody Detection: IgG antibody capture ELISA (GAC-ELISA) was performed to detect past or secondary dengue infection. IgG antibodies appear later and persist for years.

Statistical Analysis: Data were compiled in Microsoft Excel and analyzed using descriptive statistics by SPSS-25. Categorical variables were expressed as frequencies and percentages. The sample size was determined using the formula $n = (Z\alpha/2)^2 \times p(1-p) / d^2$, where n represents the sample size, Z denotes the confidence level, p indicates the expected prevalence, and d signifies the margin of error. Seroprevalence was calculated as the proportion of ELISA-positive cases relative to total suspected cases. Age and gender distributions were analyzed through frequency tabulation. Clinical symptom patterns and travel history were categorized and reported as percentages. NS1 antigen, IgM, and IgG antibody detection rates were computed separately to distinguish acute, recent, and past infections.

Results

A total of 1076 patients suspected of dengue infection were enrolled in the study. The age of patients ranged from 8 months to 95 years. Demographic characteristics, clinical presentation, and laboratory findings are summarized below.

Age Distribution: The age-wise distribution of patients showed that the highest number of cases was observed in the 21-30 years age group (339 patients, 31.50%), followed by the 11-20 years age group (306 patients, 28.44%). The distribution across different age groups is presented in Table 1 and Figure 1.

Table 1: Age-wise Distribution of Patients

Age Group (Years)	Number of Patients	Percentage (%)
0-10	58	5.39
11-20	306	28.44
21-30	339	31.50
31-40	166	15.43
41-50	79	7.35
51-60	63	5.86
61-70	48	4.46
71-80	16	1.48
91-100	1	0.09
Total	1076	100

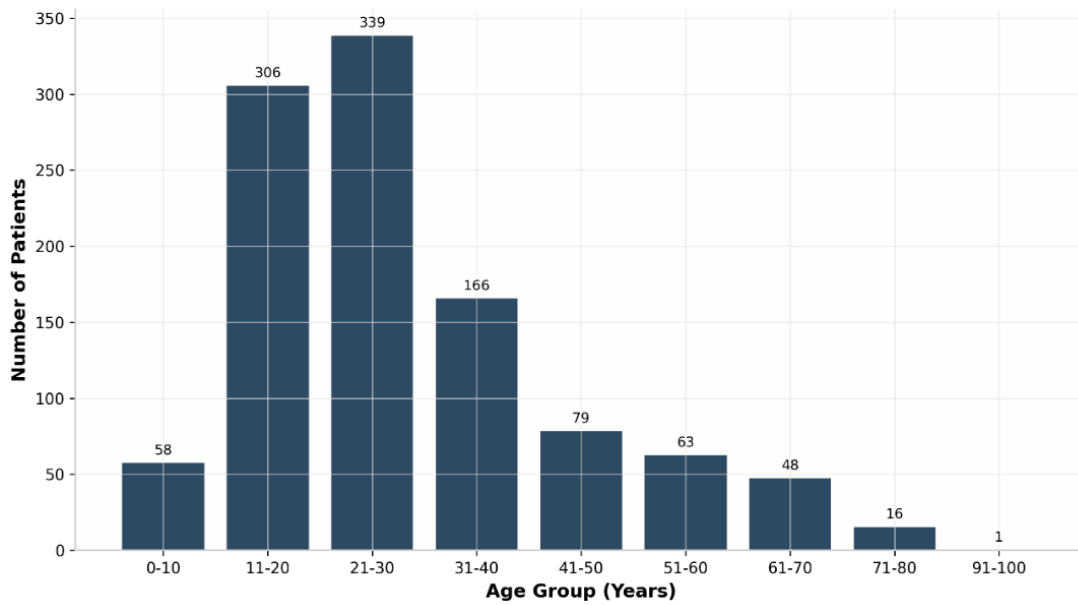


Figure 1: Age-wise Distribution of Dengue Suspected Patients

Gender Distribution: Out of 1076 patients, 598 (55.58%) were male, and 478 (44.42%) were female, indicating a higher prevalence among males. The gender distribution is shown in Table 2 and Figure 2.

Table 2: Gender Distribution of Patients

Gender	Number of Patients	Percentage (%)
Male	598	55.58
Female	478	44.42
Total	1076	100

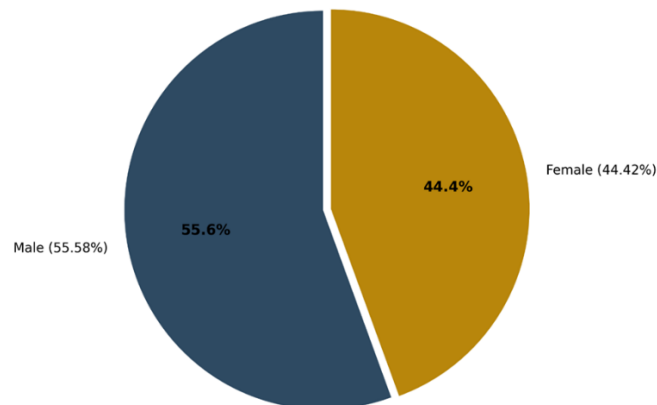


Figure 2: Gender Distribution of Dengue Suspected Patients

Clinical Presentation: The most common presenting complaint was fever and related symptoms (358 patients, 33.27%), followed by pain-related complaints (219 patients, 20.35%) and cough-related symptoms (217 patients, 20.16%). The distribution of clinical complaints is summarized in Table 3 and Figure 3.

Table 3: Distribution of Patients by Clinical Complaints

Patient Complaints	Frequency	Percentage (%)
No Complaint	92	8.55
Cough and Related	217	20.16
Fever and Related	358	33.27
Diarrhea	16	1.49
Vomiting	174	16.18
Pain and Related	219	20.35
Total	1076	100.0

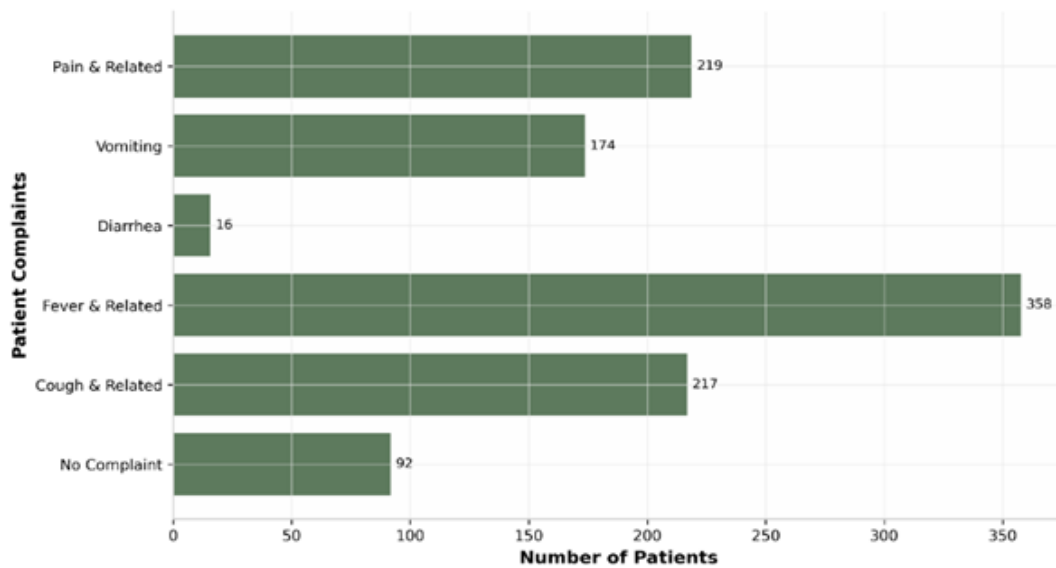


Figure 3: Distribution of Patients by Clinical Complaints

Travel History: Among the study population, 978 patients (90.90%) had no travel history, indicating local transmission of the disease. Only 98 patients (9.10%) reported travel to other cities or states, with Patna (19 patients, 1.77%) and Delhi (17 patients, 1.58%) being the most common destinations (Table 4 and Figure 4).

Table 4: Distribution of the subjects as per Patient's Travel history

Travel history	Frequency	Percent
No	978	90.90
Delhi	17	1.58
Patna	19	1.77
Surat	1	0.09
Jawalpur	1	0.09
Bhopal	1	0.09
Aurangabad	1	0.09
culcutta	3	0.27
Bikaner	1	0.09
Bombay	1	0.09
Hyderabad	5	0.47
Others	48	4.47
Total	1076	100.0

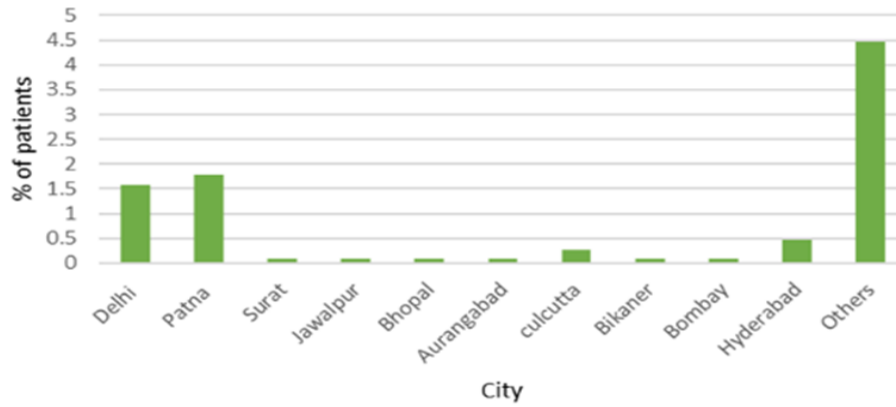


Figure 4: Distribution of the subjects as per Patient's Travel history

Sample Collection Source: The majority of samples (985, 91.54%) were collected from the laboratory, while 91 samples (8.46%) were obtained from the blood bank, reflecting the primary source of patient enrolment (Table 5).

Table 5: Distribution of the subjects as per Patient's Travel history

Sample collection	Frequency	Percent
Laboratory	985	91.54
Blood bank	91	8.46
Total	1076	100.0

Laboratory Findings: The overall seroprevalence of dengue was 13.10% (141 out of 1076 patients). Among the positive cases, NS1 antigen was detected in 86 (7.80%) patients, indicating primary or early infections. IgM antibodies were positive in 46 (4.28%) patients, suggesting recent infections, while IgG antibodies were detected in 9 (0.83%) patients, indicating past or secondary infections. The ELISA test results are summarized in Table 4 and Figure 4.

Table 5: ELISA Test Results for Dengue Diagnosis

ELISA Test	Result	Number of Patients (%)
NS1 Antigen	Positive	86 (7.80%)
NS1 Antigen	Negative	552 (51.30%)
IgM Antibody	Positive	46 (4.28%)
IgM Antibody	Negative	307 (28.53%)
IgG Antibody	Positive	9 (0.83%)
IgG Antibody	Negative	82 (7.62%)

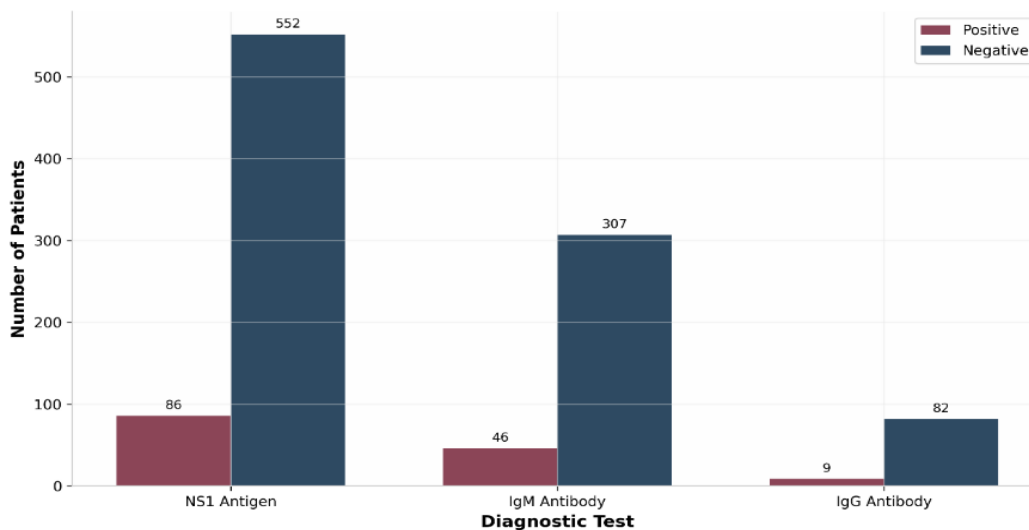


Figure 5: ELISA Test Results for Dengue Diagnosis

Discussion

This prospective study conducted at a tertiary care center in Darbhanga, Bihar, revealed an overall dengue seroprevalence of 13.10% among suspected cases. This finding is consistent with studies from other parts of India, where prevalence rates have ranged from 3.55% to 31.3% depending on the geographical location, study period, and diagnostic methods used.

The age distribution analysis revealed that young adults (21-30 years) and adolescents (11-20 years) were the most affected groups, accounting for 31.50% and 28.44% of cases, respectively. This pattern aligns with findings from Tripathi et al. [14] and other Indian studies, which have reported higher dengue incidence among young adults. The increased exposure risk in this age group may be attributed to greater outdoor activity and occupational exposure during daytime hours when *Aedes* mosquitoes are most active.

The male predominance (55.58%) observed in our study is consistent with previous reports from India and other dengue-endemic regions. Males may have higher exposure to mosquito vectors due to increased outdoor activities for work or other purposes. Tripathi et al. [14] reported similar findings, with males accounting for 62.63% of positive cases. However, some studies have shown a more balanced gender distribution, suggesting that local factors may influence this pattern.

Fever was the most common presenting symptom (33.27%), followed by pain-related complaints (20.35%) and cough-related symptoms (20.16%). This clinical presentation is consistent with the typical manifestations of dengue fever, which include high-grade fever, severe headache, retro-orbital pain, muscle and joint pain, and rash. The presence of respiratory symptoms in some patients may reflect concurrent infections or atypical presentations. A similar clinical presentation was observed by Mahesh Kumar et al., [15] fever was present in almost all cases (n=380), followed by headache (n=274), joint pain (n=2432), myalgia (n=144), retro-orbital pain (n=141), backache (n=95), skin rash (n=80).

The finding that 90.90% of patients had no travel history indicates that dengue transmission is occurring locally in the Darbhanga region. This underscores the importance of implementing local vector control measures and community awareness programs. The presence of *Aedes aegypti* mosquitoes in urban and peri-urban areas, coupled with favorable climatic conditions during the monsoon season, creates an environment conducive to dengue transmission. The detection of NS1 antigen in 7.80% of patients indicates ongoing viral replication and acute infection. NS1 antigen

detection is particularly valuable in the early phase of illness, typically within the first 5-7 days of symptom onset, before antibody responses become detectable. The presence of IgM antibodies in 4.28% of patients suggests recent infection, as IgM antibodies appear 3-5 days after symptom onset and persist for 30-60 days.

The relatively low prevalence of IgG antibodies (0.83%) in our study population suggests that most infections were primary rather than secondary. This is an important finding, as secondary infections with a different serotype are associated with a higher risk of severe dengue due to antibody-dependent enhancement. The low IgG prevalence may indicate limited circulation of multiple dengue serotypes in the study area during the study period.

The study has several limitations. First, the study was conducted at a single tertiary care center, which may not be representative of the entire Darbhanga district. Second, the study did not include serotyping of dengue virus isolates, which would have provided valuable information about the circulating serotypes. Third, the correlation between clinical severity and serological markers was not analyzed in detail.

Despite these limitations, our study provides important baseline data on dengue prevalence in the Darbhanga region. The findings highlight the need for continued surveillance, early diagnosis, and appropriate case management. Public health interventions focusing on vector control, community education, and promotion of personal protective measures are essential for reducing the burden of dengue in this region.

Conclusion

This study demonstrates a significant prevalence of dengue infection (13.10%) among suspected cases at a tertiary care center in Darbhanga, Bihar. The findings reveal that young adults (21-30 years) and males are disproportionately affected, highlighting the need for targeted preventive interventions in these demographic groups.

The predominance of NS1 antigen positivity over IgM and IgG antibodies suggests ongoing transmission of the dengue virus in the community. The high proportion of patients without travel history (90.90%) confirms local transmission and emphasizes the importance of implementing effective vector control measures in the Darbhanga region.

Serological diagnosis using ELISA for NS1 antigen, IgM, and IgG antibodies remains a valuable tool for early and accurate diagnosis of dengue infection. The combination of these tests allows for differentiation between acute, recent, and past infections, which is crucial for appropriate

clinical management and epidemiological surveillance. Based on our findings, we recommend the following measures: (1) strengthening vector control programs targeting *Aedes* mosquito breeding sites; (2) enhancing public awareness about dengue prevention and early recognition of warning signs; (3) promoting the use of personal protective measures such as mosquito repellents and bed nets; and (4) establishing continuous surveillance systems to monitor dengue trends and detect outbreaks early.

Further research should focus on serotyping of circulating dengue viruses, evaluation of clinical severity patterns, and assessment of the effectiveness of current prevention and control strategies. Collaboration between healthcare facilities, public health authorities, and communities is essential for reducing the burden of dengue in the Darbhanga region and beyond.

Acknowledgement: The authors thank the hospital staff and patients for their support of this study.

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