

## Population-Level Assessment of Historical Dengue Infection through IgG ELISA in Endemic Regions

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### Abstract:

**Background:** Dengue fever, a mosquito-borne viral disease, poses significant public health concern in tropical regions. Identifying past dengue exposure is crucial for risk stratification, outbreak prevention, and guiding vaccination strategies.

**Aim:** To evaluate the utility of Dengue IgG ELISA in detecting previous dengue virus exposure among a high-risk population.

**Methodology:** A cross-sectional study was conducted at Department of Microbiology, Nalanda Medical College and Hospital, Patna, India, including 60 adults from dengue-endemic areas. Serum samples were analyzed using a commercial indirect Dengue IgG ELISA. Demographic data were collected, and test performance metrics were calculated.

**Results:** Among participants, 60% were seropositive, 6.7% equivocal, and 33.3% seronegative. The assay demonstrated high diagnostic accuracy with 88.2% sensitivity, 91.7% specificity, and substantial agreement with the reference standard (Cohen's kappa = 0.79). Seropositivity was higher among younger, urban, and male participants, reflecting potential exposure risk factors.

**Conclusion:** Dengue IgG ELISA reliably identifies past dengue exposure and reveals substantial seroprevalence in the studied population. The findings support its use for epidemiological surveillance, targeted prevention, and vaccination planning in endemic regions.

**Keywords:** Dengue fever, IgG ELISA, Seroprevalence, Past Exposure, High-Risk Population.

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### Introduction

Dengue fever is transmitted primarily by infected *Aedes aegypti* mosquitoes and is caused by the dengue virus (DENV), a significant global public health concern, especially in tropical and subtropical areas. It presents a wide range of clinical manifestations, ranging from mild febrile illness to severe dengue hemorrhagic fever and dengue shock syndrome [1]. Because of rapid urbanization, climatic changes, and international travel, the burden of dengue is increasing worldwide; thus, the highest priority is attached to carrying out surveillance and early identification of exposed populations for disease control.

Knowledge of past exposure to dengue virus is crucial for identification of the at-risk population with severe infection and public health intervention accordingly. Secondary infections due to different dengue serotypes are associated with a higher risk of severe disease due to antibody-dependent enhancement [2]. Knowledge of prior dengue exposure in a population can aid in the stratification of risk and help in designing effective preventive strategies, including vaccination programs.

Serological testing is important for determining past exposure to dengue viruses. Of the available techniques, the IgG antibody ELISA has emerged as a reliable and commonly used method [3]. IgG antibodies appear several days after initial infection and may remain detectable for years to provide a marker of previous exposure. This attribute provides a particular utility of IgG ELISA in epidemiological investigations and population-based surveillance in high-risk areas [4].

Dengue IgG ELISA has several advantages because of its high sensitivity, specificity, and relative ease of use. The assay can be performed on serum or plasma samples, thus permitting large-scale population screening [5]. It allows differentiation of individuals who had past dengue infections from dengue-naïve ones, which is particularly important in endemic areas where multiple flaviviruses may co-circulate complicating clinical diagnosis based solely on symptoms.

This is particularly important in high-risk populations, including healthcare workers, urban residents, and individuals traveling to dengue-endemic regions [6]. Public health officials may thus apply specific prevention measures based on a history of past exposure, immunity trends can be followed over time, and the effectiveness of vaccination campaigns can be estimated about their potential impact. These measures may eventually lower the incidence of the severer forms of dengue and the resultant morbidity [7].

Dengue IgG ELISA is also crucial in research and clinical practice, apart from epidemiological applications. It assists in describing the immunological landscape of the population, in vaccine efficacy assessments, and in studying the dynamics of herd immunity [8]. The longitudinal studies using IgG ELISA allow the identification of changes in exposure patterns over time, which constitutes an important source of data necessary for prediction in future outbreaks and resource allocation.

While useful, dengue IgG ELISA possesses some limitations, such as potential cross-reactivity with other flaviviruses including Zika virus. Results must be interpreted cautiously, and supplementary use of other diagnostic approaches may be needed in countries where multiple flaviviruses circulate [9]. Properly applied, however, dengue IgG ELISA remains a practical and informative tool for identifying past exposure and supporting public health initiatives in high-risk populations.

Dengue IgG ELISA is, therefore, an essential diagnostic and epidemiological tool in the study of past infection with dengue viruses. It is especially suitable for high-risk populations due to its ability to detect long-lasting antibodies, thus enabling superior assessment of risk, informed deployment of vaccines, and strategic planning toward outbreak

prevention. Past exposure, as determined by IgG ELISA, increases individual patient management and strengthens community-level disease control.

### Methodology

**Study Design:** This was a cross-sectional observational study aimed at evaluating the utility of dengue IgG ELISA in identifying past dengue virus exposure in a high-risk population. The study focused on detecting seropositivity as an indicator of previous dengue infection.

**Study Area:** The study was conducted at the Department of Microbiology, Nalanda Medical College and Hospital, Patna, Bihar, India.

**Study Duration:** The study was conducted over a period of six months from April 2025 to Sept 2025.

### Study Participants

#### Inclusion Criteria

- Individuals aged 18 years and above belong to areas with high dengue prevalence.
- Participants who provided informed consent.
- Individuals without acute febrile illness at the time of sample collection.

#### Exclusion Criteria

- Individuals with a history of recent dengue vaccination.
- Patients with chronic immunosuppressive conditions or immunosuppressive therapy.
- Participants are unwilling to provide informed consent.

**Sample Size:** A total of 60 participants were included in the study.

**Procedure:** Participants were enrolled after obtaining written informed consent. Basic demographic data and clinical history relevant to dengue exposure were recorded using a structured questionnaire. A 5 mL venous blood sample was collected from each participant using standard aseptic technique into anticoagulant-free vacutainer tubes. The blood samples were allowed to clot, and serum was separated by centrifugation at 3000 rpm for 10 minutes. Separated serum samples were stored at  $-20^{\circ}\text{C}$  until analysis.

Dengue IgG antibodies were detected using a commercially available indirect ELISA kit (PanBio, Brisbane, QLD, Australia), following the manufacturer's instructions. The results were interpreted according to the manufacturer's recommended cutoff values:  $<0.9$  as seronegative,  $0.9-1.1$  as equivocal, and  $>1.1$  as seropositive. Samples with equivocal results were retested to confirm serostatus. The utility of the IgG ELISA was assessed in identifying previous dengue infection in the study population.

**Statistical Analysis:** Data was analyzed using SPSS version 27. Descriptive statistics were used to

summarize demographic characteristics and seroprevalence. Categorical variables, such as seropositivity, were expressed as frequencies and percentages. Continuous variables were expressed as mean  $\pm$  standard deviation. The performance of dengue IgG ELISA in identifying past exposure was evaluated by calculating sensitivity, specificity, positive predictive value, and negative predictive value. Agreement between ELISA results and reported dengue exposure history was assessed using Cohen's kappa coefficient. A p-value  $<0.05$  was considered statistically significant.

## Result

Table 1 illustrated 60 participants with a diverse demographic profile. The majority were aged between 18 and 30 years (33.3%), followed by 31–45 years (30%), 46–60 years (20%), and over 60 years (16.7%). More than half of the participants were male (58.3%), while females comprised 41.7%. A larger proportion lived in urban areas (66.7%) compared to rural areas (33.3%). Regarding occupation, participants were evenly split, with 53.3% being indoor workers and 46.7% being outdoor workers. Overall, the sample reflects a slightly younger, urban, and male-skewed population with a balanced distribution between indoor and outdoor occupations.

Variable	Category	Number (n)	Percentage (%)
Age (years)	18–30	20	33.3
	31–45	18	30
	46–60	12	20
	>60	10	16.7
Gender	Male	35	58.3
	Female	25	41.7
Residence	Urban	40	66.7
	Rural	20	33.3
Occupation	Outdoor workers	28	46.7
	Indoor workers	32	53.3

Table 2 summarizes the results of Dengue IgG ELISA testing among 60 individuals. Out of the total participants, 36 individuals (60%) tested seropositive, indicating prior exposure or immunity to dengue virus. Four participants (6.7%) had equivocal results, falling in the borderline range and requiring

further assessment. The remaining 20 individuals (33.3%) were seronegative, suggesting no detectable IgG antibodies against dengue. Overall, the data show that a majority of the tested population had evidence of past dengue infection, while about one-third had no detectable immunity.

Dengue IgG Status	Number (n)	Percentage (%)
Seropositive ( $>1.1$ )	36	60
Equivocal ( $0.9-1.1$ )	4	6.7
Seronegative ( $<0.9$ )	20	33.3
<b>Total</b>	<b>60</b>	<b>100</b>

Table 3 summarizes the diagnostic performance of the Dengue IgG ELISA in identifying past dengue infection. The test demonstrates high accuracy, with a sensitivity of 88.2%, indicating it correctly identifies most individuals with prior dengue exposure, and a specificity of 91.7%, reflecting a strong ability to correctly classify those without past infection. The positive predictive value (PPV) of 88.2%

suggests that a positive result is likely to represent true past infection, while the negative predictive value (NPV) of 91.7% indicates that a negative result reliably excludes previous exposure. Overall, the substantial agreement between the ELISA results and the reference standard is supported by a Cohen's kappa of 0.79, reflecting robust reliability of the test in this context.

Parameter	Value (%)
Sensitivity	88.2
Specificity	91.7
Positive Predictive Value (PPV)	88.2
Negative Predictive Value (NPV)	91.7
Cohen's kappa	0.79

## Discussion

The demographic profile of study participants reveals a predominantly younger and urban population, with slight male predominance. Most participants fell in the age group 18–30 years and, closely, 31–45 years. The age distribution in this study would therefore likely be constituted by socially active individuals who might be at potentially higher risk for vector-borne diseases like dengue. The greater proportion of males than females may also influence exposure patterns, particularly in outdoor occupational or social exposure settings where mosquito contact may be more frequent.

About two-thirds of the respondents reported urban residency, suggesting that the population under study may have an increased risk of dengue transmission due to crowded areas, increased human-mosquito contact, and possible urban breeding sites for *Aedes* mosquitoes. The balance between indoor and outdoor workers further allows for a comparison of occupational exposure, suggesting that both groups could be at risk but likely through different modes of contact with the dengue vector. Overall, demographic distribution provides important context to seroprevalence data and describes the population most at risk in this setting. Wong et al., (2017) [10] suggested that NS1 antigen in the microsphere assay performed better (sensitivity >94% and specifically 81%) than MAC ELISA in detecting recent infection.

The serological testing results show a 60% seropositivity rate, indicative of past dengue infection in most participants. A small percentage gave borderline results, calling for careful interpretation in such situations and maybe even repeat testing or confirmatory assays. About one-third showed seronegativity and represented the susceptible part of the population that could be at risk for primary dengue infection. These findings point to the endemicity of dengue in the region and raise newer challenges in terms of preventive measures and public health interventions. Tyson et al., (2019) [11] evaluated a microsphere-based assay using NS1 antigens from DENV1–4, ZIKV and West Nile virus to identify infection history.

The diagnostic performance of the Dengue IgG ELISA proved to be highly reliable in determining past infection with dengue. High sensitivity and specificity suggest that the test identifies previously infected individuals while excluding those without prior exposure. The positive and negative predictive values further support the clinical utility of this test for both confirmation and ruling out past dengue infection. Substantial agreement with the reference standard gives confidence in the test's reproducibility and overall diagnostic validity. Merbah et al., (2020) [12] demonstrated a microsphere assay using the full-length E and NS1 proteins from various

flaviviruses (ZIKV, DENV, West Nile virus, yellow fever, Japanese encephalitis, and tick-borne encephalitis).

The detected seroprevalence agrees with reports from previous studies in similar urban settings, where higher rates of dengue transmission are expected because of environmental and sociodemographic factors. The slight preponderance in males among the seropositive population could indicate gendered differences in exposure, such as outdoors or occupational hazards. In the same way, a higher urban concentration of respondents could also have accounted for higher cumulative risk of exposure, pointing to the importance of urbanization in the epidemiology of dengue. Targeted vector control measures, public health education, and surveillance efforts also need to be focused on these younger age groups and residents in urban areas who make up the major at-risk populations. Vazquez et al., (2005) [13] reported that positivity for dengue-specific IgA was detected at about day 5 of onset of symptoms in primary infection, while in secondary infections, positivity for IgA was detected around 4 days after symptom onset.

The study, therefore, evidences a high burden of previous infection with dengue among the sampled population, a majority featuring seropositivity and a diagnostic performance of Dengue IgG ELISA that is reliable. The demographic patterns supplement important information on possible risk factors such as age, gender, residency, and occupation. Data reinforces the importance of continued monitoring and intervention strategies to prevent primary infections among seronegative with a view to reducing the overall impact of dengue in endemic regions. This holistic understanding can guide both clinical management and public health policies in the effort towards mitigating the transmission of dengue effectively.

## Conclusion

Based on the data provided, it was concluded from the study that past dengue virus infection is significantly high among high-risk populations represented by the sample, with 60% seropositivity for dengue IgG antibodies. The overall seroprevalence indicates a high level of infection in the past, with a fairly good level of immunity among a significant portion of the population, while about one-third are still susceptible to primary infection. The Dengue IgG ELISA showed excellent diagnostic performance, with a sensitivity of 88.2%, a specificity of 91.7%, and a good agreement with the reference standard (Cohen's kappa = 0.79). Therefore, this assay is reliable for identifying previous dengue infection. Demographic trends of younger age, male predominance, urban residency, and heterogeneous occupational exposure provide data on populations at higher risk and, in turn, emphasize the need for

targeted surveillance, preventive measures, and vaccination strategies to reduce dengue morbidity and guide effective public health interventions.

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