

Seroprevalence and Immunity to Dengue Virus Among Women in Punjab: A Public Health Concern

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Abstract

Dengue virus (DENV) infection continues to be a public health concern in Punjab, India, where women are at higher risk because of socioeconomic factors. This cross-sectional study sought to assess the DENV seroprevalence, their neutralizing antibody profiles, and the risk factors associated with DENV among a sample of 1,200 women (aged 18–50 years) in Punjab. Serum samples were processed by IgG/IgM ELISA, and plaque reduction neutralization tests (PRNT90) were done to measure neutralizing antibodies. Findings showed that overall IgG seroprevalence was 48.0%, with higher rates among older age groups (57.8% in 41–50 year old women) and urban dwellers (52.4% vs. rural 41.7%). DENV₂ was the predominant serotype (37.2% multitypic immunity), but only 24.8% displayed monotypic immunity which suggests that most people were exposed to heterogeneous virus types. Urban residence (aOR=1.8), prolonged water storage (aOR=2.4), low income (aOR=1.5), and lack of mosquito nets (aOR=1.9) were other risk factors. This study highlights the need for effective vector control and vaccination programs and socioeconomic policies to reduce dengue burden among women in endemic areas.

Keywords: *Aedes aegypti*; dengue virus; neutralizing antibodies; Punjab; risk factors; seroprevalence; Women's health

INTRODUCTION

Dengue fever is a rapidly emerging arthropod-borne viral disease endemic to tropical and subtropical regions across the globe. It is estimated that more than 390 million dengue infections occur annually worldwide, posing a substantial burden on public health systems, particularly in low- and middle-income countries [1]. The disease is caused by the Dengue virus (DENV), a single-stranded RNA virus belonging to the *Flaviviridae* family and is transmitted primarily through the bite of infected *Aedes aegypti* mosquitoes. The increasing geographic spread of dengue has been attributed to rapid urbanization, population growth, climate change, and inadequate vector control measures.

In India, dengue remains a major public health concern, with recurrent outbreaks reported from several states, including Punjab. In the year 2022 alone, Punjab reported over 15,000 confirmed dengue

cases and approximately 60 dengue-related deaths, representing a nearly three-fold increase compared to the reported incidence in 2018 [2]. This alarming rise underscores the need for targeted surveillance, early diagnosis, and effective prevention strategies. Seasonal variations, monsoon-related water stagnation, and increased vector breeding sites further contribute to the cyclical nature of dengue outbreaks in the region.

Women of childbearing age represent a particularly vulnerable population group with respect to dengue infection. Socioeconomic and gender-related factors play a significant role in increasing their risk of exposure. Limited access to healthcare services, delayed health-seeking behavior, and lack

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of awareness regarding preventive measures disproportionately affect women in rural and semi-urban settings. Additionally, prolonged outdoor exposure during agricultural activities, combined with domestic responsibilities, such as water storage and household maintenance, create favorable conditions for contact with *Aedes* mosquitoes. These factors not only increase the risk of infection but may also complicate disease management, especially during pregnancy, thereby highlighting the need for gender-sensitive public health interventions and community-based awareness programs [3].

There is little documented information available on the female population of Punjab regarding the seroprevalence and the long lasting immunity of DENV. Studies conducted in Northern India suggest a 40 to 60 percent seroprevalence, however most of these studies focus on the urban male demographic [4]. Furthermore, the occurrence of cross-reactive immunity between DENV serotypes poses a challenge for vaccination approaches which makes it crucial to obtain data on the region to tailor the public health response [5]. This study aims to provide data on women from Punjab regarding the seroprevalence of DENV, neutralizing antibodies along with sociodemographic background factors to formulate precise vaccination and prevention strategies.

METHODOLOGY

Study Design and Participants

A cross-sectional study design was employed to assess dengue-related exposure and risk factors among women aged 18–50 years across Punjab, India. A total of 1,200 participants were recruited from 12 districts between June 2022 and March 2023. To ensure geographic and demographic representation, the selected districts were stratified into urban ($n = 6$) and rural ($n = 6$) clusters. Participants within each district were selected using a systematic random sampling technique, ensuring uniform coverage of households across the study area. Eligible women were approached through household visits and community health centers, and informed consent was obtained prior to enrollment. Data collection was conducted using a pretested structured questionnaire that captured sociodemographic characteristics, environmental exposure, occupational activities, and dengue-related awareness and preventive practices. The extended study duration allowed for inclusion of both peak and non-peak dengue transmission seasons, thereby enhancing the robustness of the findings. Ethical approval for the study was obtained from the institutional ethics committee, and confidentiality of participant information was strictly maintained throughout the research process.

- *Inclusion Criteria:* Reside in Punjab for at least five years and are non-pregnant without a history of receiving the dengue vaccine.
- *Exclusion Criteria:* Immunocompromised and/or suffering from an acute febrile illness at the time of enrollment.

Data Collection

Serological Testing

- *IgG/IgM ELISA:* Serum analyzed for past or acute infections using Panbio Dengue IgG/IgM kits from Abbott Laboratories.
- *Plaque Reduction Neutralization Test (PRNT90):* Determined the level of neutralizing antibodies to DENV-1–4 in IgG-positive samples per CDC protocol.

Sociodemographic Survey

Interviews were conducted to evaluate their socioeconomic status including household income, dengue diagnosis made in the past, water storage methods, and Trojan horse measures against mosquitos.

Statistical Analysis

Data was analyzed using STATA version 17. Seroprevalence was estimated using the 95% confidence intervals (CI). Risk factors were determined using logistic regression (adjusted odds ratios, $p < 0.05$).

- *Ethical Approval:* Approval was granted by Punjab Health Research Ethics Committee, with written informed consent acquired prior.

RESULTS

From the (Table 1) presented, Seroprevalence (IgG) took women between the ages of 41 to 50 years, peeking at 57.8%. This indicates that the level is cumulative ($p < 0.001$). Women in urban regions had higher levels of positivity in IgG (52.4% vs. 41.7%) and IgM (8.2% vs 3.6%) ($p=0.002$). This suggests that high populations and water stagnation in overhead tanks are contributing factors.

Table 1. Dengue seroprevalence by age and residence.

Variable	IgG-Positive (%)	IgM-Positive (%)	Total (N=1,200)
<i>Age Group</i>			
18–30 years	34.2	5.1	480
31–40 years	48.6	7.3	420
41–50 years	57.8	4.9	300
<i>Residence</i>			
Urban	52.4	8.2	720
Rural	41.7	3.6	480

Above (Table 2) DENV₂ was noticed to have the highest rate of being serotyped multitypic (37.2%), which is in line with its prevalence in the outbreaks in Punjab in recent years [2]. Merely 24.8% of women showed having monotypic immunity which shows widespread exposure to heterotypic and the chances of ADE [5].

Table 2. Neutralizing antibody profiles (PRNT90 $\geq$ 1:40).

DENV serotype	Monotypic (%)	Multitypic (%)	Total IgG+ (n=576)
DENV-1	12.4	24.8	576
DENV-2	18.9	37.2	576
DENV-3	8.5	16.7	576
DENV-4	6.3	12.1	576

The chances of dengue for people who store water for 7 days are doubled (aOR = 2.4) because stagnant water is a boon for Aedes breeding. These figures among low-income households put them at 1.5 fold odds, which suggests their lack of access to preventative means (Table 3) [3].

Table 3. Risk factors for dengue seropositivity (logistic regression).

Factor	aOR	95% CI	p-value
Urban residence	1.8	1.3–2.5	0.001
Stored water >7 days	2.4	1.7–3.4	<0.001
Low household income	1.5	1.1–2.1	0.01
No mosquito nets	1.9	1.4–2.7	0.003

DISCUSSION

The present study demonstrates a high DENV seroprevalence of 48.0% among women in Punjab, indicating sustained viral circulation and significant population-level exposure to dengue virus. This elevated seroprevalence suggests that a large proportion of women have experienced prior dengue infection, either symptomatic or asymptomatic, highlighting the endemic nature of dengue in the region. A clear age-related trend was observed, with seropositivity increasing progressively across older age groups. The stepwise rise in dengue-specific IgG antibodies with advancing age reflects cumulative exposure over time and is consistent with patterns reported from other dengue-endemic regions. In such settings, repeated exposure to different dengue virus serotypes across multiple transmission seasons contributes to higher antibody prevalence in older individuals. This finding underscores the long-term risk of dengue transmission and the potential for secondary infections, which are often associated with more severe clinical outcomes. Additionally, the observed age-related increase in seroprevalence may

be influenced by prolonged occupational and environmental exposure, particularly among women engaged in outdoor activities and domestic water storage practices. Together, these findings emphasize the need for sustained vector control efforts and targeted public health interventions, especially for high-risk demographic groups [4–6]. Urban women showed much higher IgG and IgM rates compared to rural women, indicating greater wealth with urban women owing to heavy population concentration, improper sanitation, and dependency on roof water tanks which encourage *Aedes aegypti* breeding [3]. These findings support recent reports of urban-centered dengue outbreaks in Punjab [2, 7–9].

Neutralizing antibodies to DENV₂ were predominant (37.2% multitypic immunity), which is consistent with the assumptions on its epidemiological importance in recurrent outbreaks, indicating continued circulation of this serotype. However, the small proportion of monotypic immunity (24.8%) is troubling due to the possibility of antibody dependent enhancement (ADE), where cross-reactive antibodies from earlier infections may worsen the disease during secondary heterotypic infections [5, 10]. This highlights the challenges posed by multitypic dengue transmission in an epidemic context for the vaccination strategy. It has been shown that using vaccines, such as Dengvaxia® could be dangerous for seronegative individuals in highly endemic regions [6].

Prolonged storage of water and low-income households highlight the blend of socioeconomics and environmental susceptibilities. Women from poorer communities do not have means for mosquito prevention strategies and, therefore, remain stuck in the cycle of infection [11, 12]. In addition, water storage which is required in these parts because of irregular supply becomes an unwanted breeding place requiring community solutions, such as container covers and larvicidal treatments.

Limitations of the study include exclusion of pregnant women, deemed to be potentially more susceptible to severe dengue, and the non-causal assumption due to the cross-sectional design. The focus of future longitudinal studies ought to be the impacts of DENV on maternal and child health [7–9].

CONCLUSION

The remarkable DENV seroprevalence and multitypic immunity in women of Punjab necessitates immediate actions towards integrated public health approaches. Active control of dengue vectors, such as covering water storage containers and deliveries of insecticide nets, should be more aggressive in urban and poor areas. Additionally, sero surveillance should determine vaccination policies to reduce the risk of ADE, especially in areas with strong heterotypic transmission. Policymakers need to subsidize these preventive strategies to improve women's healthcare access, thereby closing the socioeconomic inequality gap. Punjab requires integrated environmental strategies, community education, and equitable health policies to effectively manage dengue, while also alleviating its disproportionate impacts on women's health.

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