



## Original Research Article

# Analysis of clinical and laboratory profile of a recent dengue outbreak in a tertiary care hospital in Puducherry

Pon Roja Murugaraj<sup>1\*</sup>, Shashikala Nair<sup>2</sup>, Moses Marie Ambroise<sup>2</sup>, Nimmy Elizabeth George<sup>2</sup>

<sup>1</sup>Pondicherry Institute of Medical Sciences, Kalapet, Puducherry, India

<sup>2</sup>Dept. of Microbiology, Pondicherry Institute of Medical Sciences, Kalapet, Puducherry, India

## Abstract

**Background:** Dengue fever is a mosquito-borne, self-limiting viral disease, endemic in tropical and subtropical regions around the world. Detection of NS1 antigen and IgM antibody are the basic methods for diagnosis. This study was done to analyze the demographic profile, association of serological markers and platelet count with the severity of dengue infection in a tertiary care hospital in Puducherry.

**Materials and Methods:** This is a retrospective cross-sectional study. Data of 261 serologically positive dengue patients during the period between January 2022 and December 2022 was analyzed for duration of fever, risk factors, treatment modalities, complications, morbidity and mortality. Severity of the disease was correlated with serological markers, platelet count and other platelet indices.

**Results:** Seroprevalence of dengue infection was found to be 33%. Among them, 41% were exclusively positive for NS1 antigen, 38% were positive for both NS1 antigen and IgM antibody, and 21% were exclusively positive for IgM antibody. About 73.9% of the patients developed thrombocytopenia. Various platelet indices were found to have a statistically significant association with bleeding manifestations and serological markers. About 7.2% of the patients developed complications, the most common being dengue hemorrhagic fever.

**Conclusion:** Dengue infection is of major concern in and around Puducherry. Various platelet indices were found to have a statistically significant association with complications of dengue fever. A longer study period and a larger sample size would have helped us achieve better results and probably a significant association between the parameters.

**Keywords:** Dengue fever, NS1 antigen, IgM antibody, Thrombocytopenia, Platelet indices

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## 1. Introduction

Dengue fever is a mosquito-borne, self-limiting viral disease. It is widespread in many tropical and subtropical regions, mainly in urban and semi-urban areas. It is caused by Dengue virus, which has four distinct serotypes (DENV-1 to DENV-4), and spreads among humans through the bites of female Aedes mosquitoes. The symptoms of dengue infection range from mild, flu-like signs to severe, potentially fatal conditions such as dengue shock syndrome (DSS) and dengue hemorrhagic fever (DHF). DHF and DSS are associated with antibody dependent enhancement of infection. The case fatality rate with DHF and DSS could be

as high as 44%.<sup>1</sup> Over the last five decades, the global incidence of dengue has risen significantly.

Approximately 3.9 billion people across 128 countries are at risk of dengue infection. The World Health Organization (WHO) estimates that 50–100 million dengue infections occur annually.<sup>2</sup> About 104.77 million dengue cases were recorded globally in 2017, compared to 23.28 million cases in 1990.<sup>3</sup> In India, the number of dengue cases has risen dramatically in the past decade, increasing more than five times from 28,066 cases in 2010 to 157,315 cases in 2019.<sup>4</sup> Tamil Nadu reported the highest number of cases, followed by Kerala, Karnataka, Punjab, and West Bengal.<sup>5</sup> Recent outbreaks of dengue have been observed in Tamil

\*Corresponding author: Pon Roja Murugaraj  
Email: [roja.murugaraj22@gmail.com](mailto:roja.murugaraj22@gmail.com)

Nadu and Puducherry, primarily due to rapid urbanization, increased travel, and ineffective water storage practices in the urban and rural areas, which have contributed to the spread of mosquito breeding sites.

Detection of NS1 antigen and IgM antibody to dengue virus are the basic methods to diagnose dengue virus infections. NS1 antigen can be detected from the 1<sup>st</sup> day of fever. Thrombocytopenia is another predictive marker for the diagnosis of dengue fever. This study was done to estimate the prevalence of dengue cases among patients with febrile illness admitted in a tertiary care hospital during a recent outbreak in Puducherry. The association of serological markers and platelet count with the severity of disease was measured. We also analyzed the clinical and laboratory profile of dengue fever which includes the seropositivity of NS1 antigen and IgM antibody.

## 2. Materials and Methods

This retrospective cross-sectional study was conducted in a tertiary care hospital in Puducherry after obtaining approval from the institute ethics committee. Data of patients with dengue fever during the period between January 2022 and December 2022 was analyzed. The total number of patients admitted with febrile illness during this period was 794, out of which 261 patients were serologically positive for dengue infection.

Sample size was determined using the formula:  $n = Z^2 P(1-P)/d^2$ , where n indicates sample size, Z stands for Z statistic for a level of confidence, P is the expected prevalence based on similar studies and D denotes precision. With an absolute precision of 5%, and confidence interval of 95%, the required sample size was 336.<sup>6</sup>

All serologically confirmed 261 cases admitted in our institution during the period from January 2022 to December 2022 were included in the study. Outpatients who were seropositive for dengue were excluded from the study.

Age, gender, duration of fever, risk factors, complications of dengue fever, platelet parameters, NS1 antigen and IgM antibody to dengue virus were taken into consideration.

NS1 antigen and IgM antibody to Dengue virus were detected by ELISA method (J.Mitra, Okhla Industrial area, New Delhi, India). ELISA test for NS1 antigen detection was based on direct sandwich principle and IgM antibody was by IgM capture ELISA. Data of all 261 patients serologically positive for NS1 antigen and/or anti-dengue IgM antibodies were analyzed for duration of fever, treatment modalities, risk factors, complications, morbidity, and mortality. Data were retrieved from the hospital information system. These findings along with laboratory parameters such as NS1 antigen and IgM antibody positivity and lowest platelet counts of the patients were correlated with the severity of the disease (DHF, DSS, etc.). Other platelet parameters (mean

platelet volume (MPV), platelet distribution width (PDW), platelet-large cell ratio (PLCR), plateletcrit (PCT)) were documented and analyzed. Statistical analysis was done using SPSS Version 20 software. The incidence of dengue was reported in percentage (%) with 95% confidence intervals. The Chi square test was used to determine the statistical significance of categorical variables. All p-values are reported as two-sided and statistical significance will be taken at the level of 5%.

## 3. Observations and Results

During the period of January 2022 to December 2022, 794 individuals came to our institution with a febrile illness; 261 of these patients (33%), tested positive for dengue infection. Seven hundred and ninety-four patients presented with febrile illness in our institution during the period from January 2022 to December 2022, out of which 261 patients were serologically positive for dengue infection (33%). Details about the demographic distribution and characteristics of the study population are given in **Table 1**.

Majority of our study population were males belonging to age group 21 to 30 years (32.2%). About 1.5% of the patients had past history of dengue fever. The number of dengue cases was significantly high during January, October, November, and December in the year 2022. The most persistent clinical symptom was fever, followed by vomiting and headache. Bleeding manifestations were seen in 8.4% of patients. **Figure 1** illustrates the prevalence of other clinical signs.

**Figure 2** depicts the seropositivity of NS1 antigen and IgM antibody within the study group. NS1 antigen was detected exclusively in 41% of the participants, while both NS1 antigen and IgM antibody were present in 38%.

**Figure 3** shows the association between duration of dengue fever and detection of NS1 antigen and IgM antibody. About 71% of the patients had fever lasting for 1 to 5 days, and detection of NS1 antigen and IgM antibody among them was found to be higher.

The platelet indices (PIs) include platelet count, MPV, PDW, PLCR, and PCT. MPV, PDW, PLCR and PCT records were not available for 28 patients, and this analysis was done only for the remaining 233 patients.

About 73.9% of the patients had thrombocytopenia. **Table 2** depicts the distribution of platelet count among dengue patients. Most of the patients with thrombocytopenia had platelet count in the range between 10,001 to 50,000 per microliter.

A statistically significant association was observed between seropositivity for dengue-specific parameters (NS1 antigen and IgM antibody) and platelet count (p-value = 0.036), PDW (p-value = 0.002), MPV (p-value = 0.014), and PLCR (p-value = 0.002), as shown in **Table 3**.

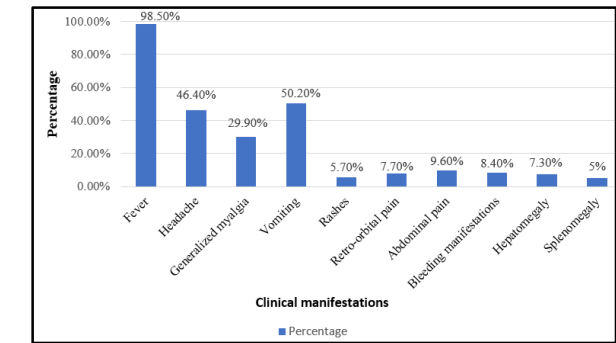


Figure 1: Clinical manifestations of dengue fever

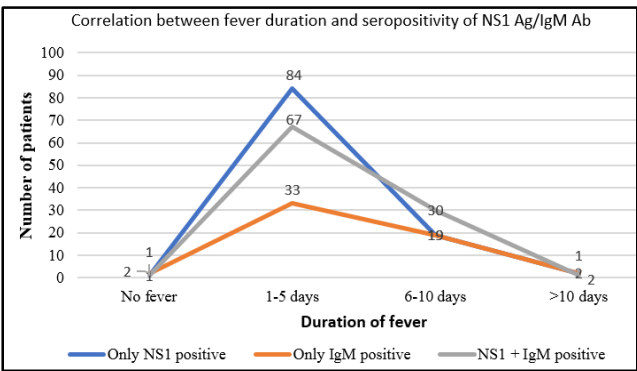


Figure 3: NS1 Ag and IgM antibody to Dengue virus positivity in relation to duration of fever.

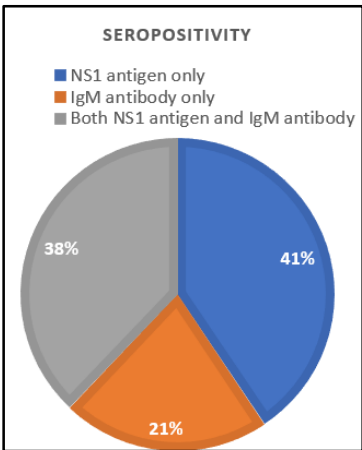


Figure 2: Comparison of various dengue specific parameters in the diagnosis of infection.

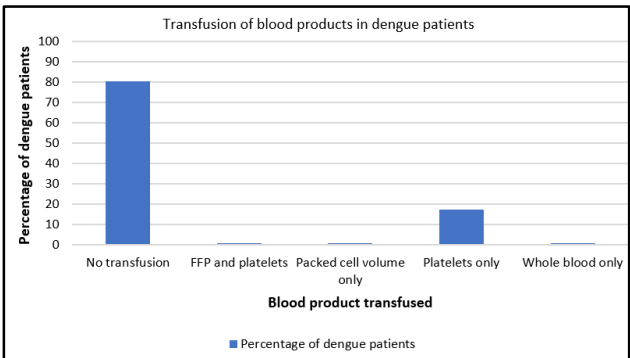


Figure 4: Transfusion of blood products in dengue patients.

Table 1: Demographic distribution and characteristics of the study population

Characteristics	Frequency	Percentage
Sex		
Male	167	64
Female	94	36
Age		
≤10 years	23	8.8
11-20 years	52	19.9
21-30 years	84	32.2
31-40 years	51	19.5
41-50 years	28	10.7
51-60 years	14	5.4
>60 years	9	3.4
Past history of dengue		
Yes	4	1.5
No	257	98.5
Family history of dengue		
Yes	6	2.3
No	255	97.7

**Table 2:** Platelet count among dengue patients

Platelet Count (per microliter)	Frequency	Percentage
<10000	35	13.4
10001-50000	136	52.1
50001 – 100000	47	18
100001-150000	22	8.4
>150000	21	8
Total	261	100

**Table 3:** Association between platelet indices and dengue specific parameters

Platelet indices	Seropositivity			Total
	NS1 +IgM positive	Only IgM positive	Only NS1 positive	
<b>Platelet Count (per microliter)</b>				
<10000	12 (34.3%)	3 (8.6%)	20 (57.1%)	35
10001-50000	56 (41.2%)	30 (22.1%)	50 (36.8%)	136
50001 – 100000	22 (46.8%)	11(23.4%)	14 (29.8%)	47
100001-150000	5 (22.7%)	4 (18.2%)	13 (59.1%)	22
>150000	4 (19.0%)	8 (38.1%)	9 (42.9%)	21
<b>Platelet Distribution Width</b>				
≤ 14	47 (29.0%)	37 (22.8%)	78 (48.1%)	162
> 14	38 (53.5%)	12 (16.9%)	21 (29.6%)	71
<b>Mean Platelet Volume (fL)</b>				
≤ 11.5	56 (31.5%)	39 (21.9%)	83 (46.6%)	178
>11.5	29 (52.7%)	10 (18.2%)	16 (29.1%)	55
<b>Platelet-Large Cell Ratio</b>				
≤ 30	25 (24.3%)	27 (26.2%)	51 (49.5%)	103
>30	60 (46.2%)	22 (16.9%)	48 (36.9%)	130

**Table 4:** Association between platelet indices and transfusion of blood products

Platelet indices	Transfusion of blood products		Total
	Yes	No	
<b>Platelet Count (per microliter)</b>			
<10000	30 (85.7%)	5 (14.3%)	35
10001-50000	19 (14.0%)	117 (86.0%)	136
50001 – 100000	0 (0.0%)	47 (100.0%)	47
100001-150000	1 (4.5%)	21 (95.5%)	22
>150000	1 (4.8%)	20 (95.2%)	21
<b>Mean Platelet Volume (fL)</b>			
≤ 11.5	27 (15.2%)	151 (84.8%)	178
>11.5	15 (27.3%)	40 (72.7%)	55
<b>Plateletcrit</b>			
<0.1	40 (21.9%)	143 (78.1%)	183
>0.22	1 (10.0%)	9 (90.0%)	10
0.1-0.22	1 (2.5%)	39 (97.5%)	40

**Table 5:** Association between platelet indices and bleeding manifestations

Platelet indices	Bleeding manifestations	Total	
	Yes	No	
<b>Platelet Distribution Width</b>			
≤ 14	9 (5.6%)	153 (94.4%)	162
> 14	11 (15.5%)	60 (84.5%)	71
<b>Mean Platelet Volume (fL)</b>			
≤ 11.5	9 (5.1%)	169 (94.9%)	178
>11.5	11 (20.0%)	44 (80.0%)	55

**Table 4** demonstrates a statistically significant correlation between blood transfusion and platelet count (p-value = 0.000), MPV (p-value = 0.041), and PCT (p-value = 0.012).

PDW (p-value = 0.020) and MPV (p-value = 0.001) had a statistically significant association with bleeding manifestations (**Table 5**).

SGOT, SGPT, GGT and ALP levels were deranged in around 185 (70.9%) patients. Urea and creatinine values were elevated in 11 (4.2%) patients.

About 10(3.8%) patients developed DHF, 9(3.4%) patients developed DSS, and 3(1.1%) patients expired. Figure 4 shows transfusion of blood products in dengue patients.

Transfusion of blood products was not required for majority of dengue patients (80.5%). Only platelet transfusion was done in 17.2% patients.

#### 4. Discussion

Dengue fever is a significant public health issue. The complications associated with dengue fever and DHF arise from multiple factors, including coagulopathy, thrombopathy and vasculopathy. Several elements can contribute to the development of thrombocytopenia in dengue fever, ranging from an immune reaction against platelets to a reduction in platelet production.<sup>7</sup> Platelet indices provide insight into whether platelet destruction is actively occurring or if the bone marrow is responding to the condition. This helps us determine if the patient would require blood transfusion.<sup>8</sup> It includes platelet count, MPV, PDW, PLCR and PCT.

This study analyzes the seroepidemiology of dengue fever during a recent outbreak in Puducherry, conducted in a tertiary care hospital, during the period from January 2022 to December 2022. A total 794 of patients presented with febrile illness in our institution during this period, out of which 261 patients were serologically positive for dengue NS1 antigen or anti-dengue IgM antibody. Seroprevalence of dengue infection was observed to be 33%.

We found that the incidence of dengue was more among patients aged between 21 and 30 years (32.2%). The second most affected age group was between 11 and 20 years (19.9%). The least affected age group was more than 60 years (3.4%). These results are similar to a study conducted by Anish Laul et al during a dengue outbreak in Northern India in 2016.<sup>9</sup> A study conducted in central India by Ukey and Bondade found that the age group most affected by dengue was between 15 and 30 years, with a higher incidence in males compared to females.<sup>10</sup>

Males were more frequently affected than females in this study, showing a male-to-female ratio of 1.8:1. Similar results were found in a few other studies conducted by Kaur, Laul and Bondade.<sup>7,10,11</sup> The number of dengue cases in our

study was significantly higher during the colder months (October, November, December, and January). In a study conducted by Murhekar et al, dengue positivity was higher during September and October months.<sup>11</sup> Studies conducted by Bondade and Murhekar showed higher incidence of dengue fever during the monsoon season.<sup>11,12</sup> Temperature and rainfall have a potential role on the vector, which aids in the large-scale transmission of dengue virus. Although there was a distinct seasonal peak, cases were recorded throughout the year, indicating ongoing endemic transmission.

In our study, the most frequently observed clinical symptom was fever, present in 98.5% of cases, followed by vomiting in 50.2% and headache in 46.4%. The least frequently observed symptom was splenomegaly, occurring in 5% of patients. Fever and headache were also identified as the primary complaints in a few other studies conducted elsewhere.<sup>13,14</sup> Bleeding manifestations were seen in 8.4% of the patients, which was congruent to the studies conducted by Srivastava et al and Aggarwal.<sup>15,16</sup> Statistically significant association was found between certain platelet indices (MPV and PDW) and bleeding manifestations.

Majority of the patients had fever for 1-5 days (70.5%), and most of these patients were only NS1 antigen positive, or both NS1 antigen and IgM antibody positive. This indicates that NS1 antigen serves as an early diagnostic marker. Among the 261 seropositive dengue cases, 41% tested positive for only NS1 antigen, 21% for only IgM antibody, and 38% for both NS1 antigen and IgM antibody. NS1 antigen appears from day-1 of fever and can be detected till 9 to 14 days, whereas IgM antibody appears by day-5, and remains till 90 days.

Platelet count less than 1,50,000 per microliter was considered as thrombocytopenia. Majority of the patients in this study developed thrombocytopenia (73.9%). A statistically significant correlation was observed between platelet count and the need for blood transfusion. A similar finding was reported in a study by Asha et al.<sup>8</sup> These patients also had a statistically significant association with seropositivity for dengue specific parameters (NS1 antigen and IgM antibody). About 13.4% of the patients had platelet count <10,000 per microliter, among which 85.7% of the patients required blood transfusion, and 91.4% of the patients were seropositive for NS1 antigen, exclusively, or along with IgM antibody. The pathophysiology behind thrombocytopenia in dengue is not fully understood. It may result from direct suppression of thrombopoiesis in the bone marrow, potentially caused by the dengue virus's effect on endothelial cells or the destruction of platelets by anti-NS1 antibodies. Rapid intervention is required to avoid associated risk of thrombocytopenia.

PDW in most of the patients (62.1%) was less than or equal to 14 fL, and 77.1% of these patients were seropositive for NS1 antigen, exclusively (48.1%) or along with IgM antibody (29%). PDW was found to have a statistically

significant association with seropositivity for dengue specific parameters (NS1 antigen and IgM antibody).

Most patients (68.2%) had a MPV of  $\leq 11.5$  fL. Among them, 46.6% were positive for only NS1 antigen, while 31.5% were positive for both NS1 antigen and IgM antibody. A significant statistical correlation was observed between MPV and seropositivity for dengue-specific markers (NS1 antigen and IgM antibody). MPV was found to have a statistically significant association with blood transfusion. A study by Kantharaj suggested that elevated MPV could serve as a marker for platelet production rate and activation.<sup>9</sup> A high MPV with persistent thrombocytopenia points to peripheral platelet destruction, while a low MPV suggests a reduced production or suppression of the bone marrow.<sup>16</sup>

A large portion of the patients (49.8%) had PLCR more than 30%. Among these patients, 46.2% tested positive for both NS1 antigen and IgM antibody, while 36.9% were positive for only NS1 antigen. A statistically significant association was observed between PLCR and seropositivity for dengue-specific markers (NS1 antigen and IgM antibody). An elevated PLCR was highly sensitive and specific for diagnosing dengue fever in a study conducted by Ram and Meena, which is in accordance with our study.<sup>17</sup>

Majority of the patients in our study (70.1%) had PCT less than 0.1%, out of which 21.9% patients required transfusion of blood products. PCT was found to have a statistically significant association with blood transfusion. Similar findings were present in a few other studies.<sup>8,17</sup> Platelet indices did not have any significant association with the development of dengue complications (DHF/DSS). Liver function tests were found to be deranged in about 70.9% of the patients. Similar findings were present in another study.<sup>18</sup>

Out of 261 patients, 51(19.5%) patients required blood transfusion, mostly platelet transfusion (17.2%). About 10 (3.8%) patients developed DHF, 9 (3.4%) patients developed DSS, and 3 (1.1%) patients expired. About 91.5% of the patients did not develop any complications, which says that appropriate diagnostic methods were used, and the treatment was effective.

## 5. Conclusion

Dengue fever poses a significant health concern in Puducherry. Our study estimated the prevalence of dengue infection in our institution to be 33% for the period from January 2022 to December 2022. We found a notable association between seropositivity for dengue-specific markers (NS1 antigen and IgM antibody) and platelet count. Additionally, MPV and PDW showed a statistically significant correlation with bleeding manifestations. Platelet indices did not have any significant association with the development of other dengue complications. A longer period of study and a larger sample size would have helped us achieve better results and probably, a significant association

between the parameters. Since this was a retrospective analysis, we relied solely on the hospital information system for data. However, the unavailability of certain patient details posed a challenge to our study.

## 6. Conflict of Interest

None.

## 7. Source of Funding

None.

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