

# Evaluation of Thrombocytopenia in Dengue Infection Along with Seasonal Variation in Rural Melmaruvathur

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## ABSTRACT

**Introduction:** Dengue is one of the most prevalent mosquito-borne arboviral infection in India. Seventy percent of the 96 million apparent infections occur in Asia, in which India is making upto one third of the total. Despite considerable efforts to control the mosquito populations, dengue fever has emerged, spread and established itself rapidly. The most serious manifestations of the infection are Dengue Hemorrhagic Fever (DHF) and Dengue Shock Syndrome (DSS).

**Objectives:** Evaluation of thrombocytopenia in dengue infection along with seasonal variation.

**Material and Methods:** The present study was conducted retrospectively for a period of two years in Melmaruvathur from January 2011 to December 2012. Blood samples were collected from 1,464 patients experiencing febrile illness, clinically consistent with dengue infection. Serological confirmation of dengue infection and evaluation of platelet counts was done in all the serologically positive cases.

**Results:** A total of 1,464 fever cases were admitted during the study period and among them, 107 (7.3%) were found to be seropositive for dengue. A majority, 41 (38.3%) of the dengue cases was noted in the age group of 15-30 years, followed by less than 15 years of age and the mean age of the seropositive cases was  $29.78 \pm 15.3$  years. Among the dengue cases, 62.6% were males and 37.4 % were females. A statistically significant association ( $p < 0.05$ ) was found between the age groups and severity of thrombocytopenia, between severity of thrombocytopenia and clinical presentation of dengue infections. A significantly higher number of serologically positive cases were noted, when compared to serologically negative cases in the post monsoon period.

**Conclusion:** This retrospective study highlighted significant dengue infection during seasonal variation. The median age of dengue patients has decreased and younger patients may be more susceptible.

**Keywords:** Dengue infection, Dengue fever, Dengue hemorrhagic fever, Dengue shock syndrome  
Thrombocytopenia

## INTRODUCTION

Dengue fever is one of the most prevalent and fastest spreading mosquito-borne arboviral infection occurring in tropical and sub-tropical regions around the world and leads to explosive outbreaks in urban areas influenced strongly by rainfall and temperature [1,2]. According to WHO, About two-fifths of the world's population (2.5 billion people) are currently at risk of dengue fever. Dengue fever is endemic in more than 100 countries. About one in every 40 patients with DHF die from the disease if left untreated, approximately 20% of people with DHF die from the infection. Dengue infection is caused by any one of the four distinct but closely related dengue virus (DENV) serotypes (called DENV-1,-2,-3, and-4). These dengue viruses are single-stranded RNA viruses that belong to the family Flaviviridae and the genus Flavivirus. Dengue viruses are arboviruses (arthropod-borne virus) that are transmitted primarily to humans through the bite of an infected *Aedes* species mosquito [2]. Transmission may also occur through transfusion of infected blood or transplantation of infected organs or tissues. Human transmission of dengue is also known to occur after occupational exposure in health care settings (e.g., needle stick injuries) and cases of vertical transmission have been described in the literature (i.e., transmission from a dengue infected pregnant mother to her fetus in utero or to her infant during labor and delivery).

Dengue Infection is observed to be a seasonal disease. It leads to explosive outbreaks, mostly in urban areas strongly influenced by rainfall and temperature [2]. Interestingly, the El Nino phenomenon may also be related to the diseases transmitted by mosquitoes, such as malaria and dengue [3]. The regions where El Nino cycle has a strong effect on climatic conditions include South Africa, parts of South America and Southeast Asia.

## Objective

Evaluation of thrombocytopenia in dengue infection along with seasonal variation.

## MATERIAL AND METHODS

The present study was conducted retrospectively for a period of two years in Melmaruvathur Adhiparasakthi Institute of Medical Sciences, Melmaruvathur, Tamil Nadu, India, during January 2011 to December 2012. Blood samples were collected from 1,464 patients experiencing a febrile illness, clinically consistent with dengue infection. Serological confirmation of dengue infection and evaluation of platelet counts was done in all serologically positive cases. According to the intensity of rainfall, weather data was divided into three periods namely; Pre monsoon period [February-May], Monsoon period [June-September] and Post monsoon period [October-January].

The reference criteria for diagnosis DF/DHF/DSS [4] is given below:

### Clinical case definition of dengue fever

Classical dengue fever or break bone fever is an acute febrile viral disease frequently presenting with headaches, bone or joint pain, muscular pains, rash, and leucopenia caused by the aedes mosquito infected with a dengue virus.

### Clinical case definition DHF

#### Necessary Criteria

1. Fever or recent history of acute fever.
2. Hemorrhagic manifestations.
3. Low platelet count ( $100,000/\text{mm}^3$  or less).

Age groups	Sex		Total n (%)
	Male n (%)	Female n (%)	
< 15 years	18 (26.9)	11 (27.5)	29 (27.1)
15 – 30 years	24 (35.8)	17 (42.5)	41 (38.3)
31 – 50 years	19 (28.4)	9 (22.5)	28 (26.2)
> 50 years	6 (9)	3 (7.5)	9 (8.4)
Total	67 (62.6)	40 (37.4)	107 (100)

[Table/Fig-1]: Age and sex distribution of dengue fever cases

Age group	Platelet count				Total	p-value
	< 20,000	21,000 - 50,000	51,000 – 100,000	> 1 LAKH		
<15 years	5 17.2%	7 24.1%	13 48.8%	4 13.8%	29 100.0%	0.04*
15-30 years	1 2.4%	9 22.0%	15 36.6%	16 39.0%	41 100.0%	
31-50 years	2 7.1%	10 35.7%	7 25.0%	9 32.1%	28 100.0%	
Above 50 years	2 22.2%	2 22.2%	3 33.3%	2 22.2%	9 100.0%	
Total	10 9.3%	28 26.1%	38 35.6%	31 29.0%	107 100.0%	

[Table/Fig-2]: Age group wise distribution and platelet count  
\*p<0.05 (significant)

Platelet count	Category of dengue			Total	p-value
	DF	DHF	DSS		
<20,000	0 0.0%	7 70.0%	3 30.0%	10 100.0%	.003*
21,000-50,000	16 57.1%	10 35.7%	2 7.1%	28 100.0%	
51,000-100,000	20 52.6%	12 31.5%	6 15.7%	38 100.0%	
>100,000	22 71.0%	9 29.0%	0 0.00%	31 100.0%	
Total	58 54.2%	38 35.5%	11 10.2%	107 100.0%	

[Table/Fig-3]: Relationship between platelet count and category of dengue infection  
\*p<0.05 (significant)

	Total no of suspected cases of Dengue	Total no of suspected adult cases	Total no of serologically positive adult cases	Total no of suspected pediatric cases	Total no of serologically positive pediatric cases	Total no of serologically positive cases
January	79	45	2	34	1	3
February	84	62	2	22	0	2
March	69	44	3	25	2	5
April	44	33	1	11	0	1
May	58	44	4	14	3	7
June	98	67	3	31	2	5
July	92	76	3	26	1	4
August	101	78	6	23	2	8
September	177	133	11	44	3	14
October	224	168	13	56	5	18
November	211	192	14	54	6	20
December	227	177	16	50	4	20
Total	1,464	1,119	78	390	29	107

[Table/Fig-4]: Seasonal variation of dengue infection

### Objective evidence of “leaky capillaries:”

Elevated hematocrit (20% or more over baseline).

Low albumin.

Pleural or other effusions.

### Clinical case definition of DSS

Four criteria for DHF.

Evidence of circulatory failure manifested indirectly by all of the following.

Rapid and weak pulse.

Narrow pulse pressure (B20 mm Hg) OR.

Hypotension for age.

Cold, clammy skin and altered mental status.

Frank shock is direct evidence of circulatory failure.

## RESULTS

### Distribution of Dengue fever cases

A total of 1,464 fever cases was admitted during the study period and among them, 107 (7.3%) were found to be seropositive for dengue. Of the seropositive cases, 76 (71%) had thrombocytopenia (platelet count <1 lakh) while the remaining, 31 (29%) had platelet counts >1 lakh. A Majority, 41 (38.3%) of the dengue cases was noted in the age group of 15 – 30 years, followed by less than 15 years of age (27.1 %). The mean age of the seropositive cases was 29.78 ± 15.3 years. Among the dengue cases, 62.6 % were males and 37.4 % were females. The age and sex distribution is given in [Table/Fig-1].

Out of 76 patients with thrombocytopenia (platelet count <1 lakh), 38 (50.0%) had mild thrombocytopenia (platelet count 51,000 – 1 lakh), 28 (36.8%) had moderate thrombocytopenia (platelet count 20,000 – 50,000) and remaining 10 (13.2%) had severe thrombocytopenia (platelet count <20,000). Thrombocytopenia was found to be more severe in age group <15 years, when compared to other age groups. A statistically significant association (p<0.05) was found between the age groups and severity of thrombocytopenia [Table/Fig-2].

At the time of admission to hospital, of the 107 seropositive cases, 58 (54.2%) patients clinically presented with dengue fever, followed by 38 (35.5%) patients with DHF and remaining 11 (10.2%) in DSS. Among the thrombocytopenia cases, it was observed that, 47.4% (18/38 cases) of patients with mild thrombocytopenia, 42.3% (12/28 cases) of patients with moderate thrombocytopenia and 100% (10/10) of patients with severe thrombocytopenia presented with

clinical features of DHF/DSS. A statistically significant association ( $p < 0.05$ ) was seen between severity of thrombocytopenia and clinical presentation of dengue infection cases. The relationship between severity of thrombocytopenia and clinical category of dengue infection is shown in the [Table/Fig-3].

On follow up of patients after necessary treatment, all cases of dengue fever and 31 cases of DHF, showed clinical improvement with an elevation of platelet counts and complete recovery, irrespective of their varying platelet counts. Remaining 18 cases (7 DHF and 11 DSS) were referred to a higher specialized centre for appropriate management.

According to the study during the monsoon period, only 31 cases (29%), post monsoon 61 cases (57%) and pre-monsoon period 15 cases (14%) were confirmed serologically positive [Table/Fig-4]. The difference between numbers of serologically positive cases as compared to serologically negative ones in the post-monsoon period was significantly higher.

## DISCUSSION

A total of 107 seropositive cases was noted among 1,464 patients experiencing a febrile illness, clinically consistent with Dengue infection admitted during the study period. In the present study, most patients with serological and virological confirmation were in the age group of 15-30 years and this is in accordance with the study by Pruthvi et al., [5]. In Pakistan, study by Erum et al., shows median age group affected by dengue infections are young patients and males were affected predominantly [6]. In Indonesia, data from 1975 to 1984 showed an increase in incidence rates among young adults in Jakarta as well as in the provincial areas [7]. In our study DI was predominant among males (62.6%) than females (37.4%). This is in accordance with the study by Jayashree et al., [8]. Although DF is a self limited febrile illness, DHF is characterized by prominent hemorrhagic manifestations associated with thrombocytopenia and an increased vascular permeability [9]. The clinical diagnosis of DHF especially in the early phase of illness is not easy. Laboratory diagnosis depends on virus isolation and identification of virus-specific antibodies. Dengue viruses can be isolated from serum, plasma or leukocytes during the febrile phase of the disease (within 6 days). IgM antibodies are the first to appear and are detectable in 50% of patients by days 3 -5 after onset of illness, increasing to 80% by day 5. IgG antibody is detectable at low titers at the end of the first week, increasing thereafter and still detectable after several months. Molecular methods (nucleic acid detection assays) may identify the virus within 24-48 hours.

Laboratory findings such as thrombocytopenia and a rising hematocrit in DHF cases are usually observed by day 3 or 4 of the illness. Thrombocytopenia may be occasionally observed in DF but is a constant feature and one of the diagnostic criteria of DHF and DSS [10]. Though the dengue virus induced bone marrow suppression decreased platelet synthesis, an immune mechanism of thrombocytopenia caused by increased platelet destruction appears to be operative in patients with DHF [11]. The present study showed DHF/DSS was more common in patients of DI with thrombocytopenia and this is supported by Mourao MP who has observed that patients with DHF/DSS had lower platelet counts than patients with only DF [12].

In the study at Belgium, thrombocytopenia was found in 83% of cases during hospitalization [13]. In the present study 71% of cases of DI had thrombocytopenia. The diagnosis of DF/DHF/DSS was based on the criteria stated in the [Table/Fig-1], whereas in the study of De Castro et al., [11] The diagnosis of DHF was confirmed by The presence of hemorrhagic signs such as petechiae with purpura, epistaxis, menorrhagia or a positive tourniquet test. DSS was confirmed with a pulse pressure B20 mm Hg. Recently, Premaratna et al., demonstrated a rash in 95% of the patients using a hand impression technique in a study conducted at a teaching hospital

in Sri Lanka [14]. In view of this, it is suggested that evaluation of rash in patients presenting with short duration fever may help in the diagnosis of dengue fever.

The major pathophysiological hallmark that determines disease severity and distinguish DHF from DF and other viral hemorrhagic fevers are plasma leakage due to increased vascular permeability and abnormal hemostasis. Hypovolemic shock occurs as a consequence of and subsequent to, critical plasma volume loss. Abnormal hemostasis including increased capillary fragility, thrombocytopenia, impaired platelet function, consumptive coagulopathy and in the most severe fever form disseminated intravascular coagulation (DIC) contribute to varying degree of hemorrhagic manifestations [15].

This study coincided mainly with the post monsoon period of subnormal rainfall and was followed by relatively heavy rainfall during the monsoon period from June to September 2011-12. The difference in the rainfall and temperature between three seasonal periods was found to be significant. The outbreak coincided mainly with the post monsoon. Unusual heavy rainfall subsequently led to decrease in temperature during the later part of monsoon period.

The temperature showed a decline and remained almost constant during the months of July and August (30.2°C), continuous heavy rainfall subsequently led to a further decrease in the temperature during the month of September to 29°C. Relative humidity increased during the rainy season and remained high for several weeks. An in-depth analysis of these three factors thus led to a proposal that optimum temperature with high relative humidity and abundant stocks of fresh water reservoirs generated due to rain developed optimum conditions conducive for mass breeding and propagation of vector and transmission of the virus [16].

## CONCLUSION

According to this study, platelet count is a predictive as well as a recovery parameter of DF/DHF/DSS. This retrospective study highlighted significant dengue infection during seasonal variations. Median age group of dengue patients has decreased and younger patients may have become more susceptible. The only way to prevent from becoming infected and developing dengue fever is to avoid being bitten by the mosquito. However, if caught early on, it is easier to prevent complications. Further research needed about the significant dengue epidemic during climatic conditions.

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