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Serodiagnosis and Clinical Profile of Dengue Virus Infection in Patients Presenting to A Tertiary Care Hospital

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Dengue virus infection is transmitted by the *Aedes aegyptii* mosquito. It causes a spectrum of illnesses ranging from asymptomatic cases, mild dengue fever (DF) to severe complications like dengue hemorrhagic fever / dengue shock syndrome (DHF/ DSS). This viral infection is seen worldwide, with large number of cases in the South-East Asian countries, including, India. This study was conducted to find the distribution of dengue virus infection and to study the clinical profile among the hospitalized patients. The study was conducted in the department of Microbiology of a tertiary care center for a year. After history taking and clinical examination blood samples were collected from 250 suspected dengue cases. These samples were subjected to ELISA to detect Dengue IgM antibodies. Patient's platelet count and hematocrit were also determined. Out of the 250 cases 155 (62%) were positive for IgM dengue antibodies. The age group of affected patients ranged from 1-20 years. Disease was distributed more in males. The monsoon and post-monsoon seasons accounted for majority of cases. The disease manifested as DF- 79%, DHF - 17% and DSS - 4% cases. The various symptoms associated with disease were fever, headache, joint pain, retro-orbital pain, myalgia and backache. Decreased platelet count was seen only in 15% of the cases. Hematocrit was within normal range. Ours is an area endemic for dengue virus, with disease prevalent more in the young male children and adults. Severe forms of dengue are important causes of morbidity and mortality in the pediatric age group. Important public health measures must be taken during the monsoon and post-monsoon seasons to decrease this trend.

Key words: Dengue virus; IgM ELISA; Dengue Fever; Dengue Shock Syndrome**.**

The name 'Dengue' is derived from the Swahili word *'Ki denga pepo'*, which means 'sudden seizure by the demon'[1]. Following the Philadelphia epidemic in 1780, it was called as the 'break bone fever' by Benjamin Rush[2]. The dengue virus is an arthropod borne virus-*Arbovirus*, belonging to the family *Flaviviridae* and genus *Flavivirus*. It is a mosquito borne viral infection and is transmitted, primarily by *Aedes aegypti* and sometimes by *Aedes* albopictus[3].Dengue is caused by four distinct serotypes of viruses; *DEN-1, DEN-2, DEN-3 and DEN-4*[4].

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Dengue virus causes a spectrum of illnesses ranging from in-apparent, self- limiting classical dengue fever (DF) to life threatening dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS). Dengue fever is the most common emerging disease of Tropical and Sub tropical regions. It is the major cause of hospitalization and death especially among children in these regions. All the four serotypes of the virus are seen in India. During epidemics of dengue, the attack rates among susceptible are 40-90%[5].

An increased disease burden has been linked to the resurgence of the vector, overcrowding and urbanization. Despite its significant health and economic impacts, as of now there is no specific treatment and vaccine[6].

Diagnosis of dengue infection at the earliest is of utmost importance in order to prevent the life threatening complications like DHF/DSS. Diagnosis of infection by serology has high sensitivity (95-98%) and specificity (100%) [7].

A positive IgM Capture assay after five days of onset of symptoms was considered as the criteria for the diagnosis in the study. A rise in titre of IgG antibodies was not detected as most of patients were not available for second sample. Virus culture was not attempted as most of the patients were referred to the hospital more than a week after the onset of fever.

Therefore in the present study only IgM capture ELISA, using EUROIMMUNE ELISA kit was done to identify dengue positive cases of the clinically suspected cases and also to study the disease profile among the infected patients.

MATERIALS AND METHODS

The study was conducted in the Department of Microbiology, in a tertiary care center from February 2007 to February 2008.

Patients enrolled in the study were from both inpatient and outpatient departments of Pediatrics & Medicine.

Patients were selected keeping in mind the inclusion & exclusion criteria.

Inclusion criteria

Individuals with symptoms of fever of more than five days duration and with more than or equal to two of the following

1. Joint pain 2. Rash
3. Myalgia 4. Retro-orbital pain
5. Headache 6. Hemorrhagic manifestation.

Exclusion criteria

Patients with fever of less than five days duration. Complete clinical history was taken and a thorough clinical examination was done for all the 250 cases. Informed consent was taken from all the patients. Blood samples from suspected dengue cases were collected with all aseptic precautions. Each blood sample was divided into two halves. From one half serum was isolated and used for serology and the other half was used for complete blood count.

For all the 250 serum samples Enzyme linked immunosorbent assay was performed to detect the presence of IgM anti-dengue virus antibody, using EUROIMMUN ELISA kit.

The 250 blood samples were also analyzed for platelet count using an automated counter, Horiba, ABX diagnostics, MICROS- 60. The data obtained was statistically analyzed and the results were tabulated

RESULTS

A total of 250 blood samples were tested for dengue virus IgM antibody. One hundred and fifty five cases (62%) tested positive for dengue virus. Distribution of dengue positive cases was more among males i.e., 61% and less in females 39%.

Dengue positive cases were more in the age group range of 1-20 years, i.e. children & young adults. However the complications of DHF & DSS were more associated with children between 1-10 years (Table 1).

Seasonally the dengue cases were more clustered during the months of August to November. Among the patients found positive for dengue IgM antibody, majority of the patients had dengue fever (79%), others had DHF (17%) & DSS (4%).

Table 1. Age wise distribution of dengue positive cases

Age	Dengue positive	Distribution %	DF	DHF	DSS
Less than 11 months	10	6.4	8	2	0
1-10 y	95	61.2	71	19	5
11-20 y	37	23.8	31	5	1
21-30 y	6	3.8	6	0	0
31-40 y	1	0.6	1	0	0
41- 50 y	6	3.8	5	1	0
More than 51 y	0	0	0	0	0
Total	155	100	122	27	6

Table 2. Clinical spectrum of cases

Diagnosis	Number	Distribution %
df	122	79
dhf	27	17
dss	6	4
Total	155	100

The most common symptoms that the patients came with were fever (100%), headache (70%) and joint-pain (66%). The less common symptoms were; retro-orbital pain (46%), myalgia (37%), backache (25%), hemorrhagic manifestations (15%), splenomegaly (13%), hepatomegaly (10%) and CNS symptoms (2%)

In our study 23/155 (15%) had thrombocytopenia, rest of the cases 132/155 (75%) had normal platelet count. Thrombocytopenia was more seen in DHF & DSS (>50%).

Table 3. Relation of platelet count with DF, DHF and DSS

	Thrombocytopenia	Normal count	Total
DF	0 (0%)	122 (100%)	122
DHF	18 (67%)	9 (33%)	27
DSS	5 (83%)	1 (17%)	6
TOTAL	23	132	155

Table 4. Hematocrit levels in dengue positive cases

	No. of cases	Hematocrit	
		Range	Mean +/- SD
DF	122	20.3-44.0	35.1 +/- 3.7
DHF	27	21.0-36.2	28.8 +/- 4.6
DSS	6	24.3- 49.4	39.4 +/- 10.8
Dengue positive	155	20.3- 49.4	34.2 +/- 5.0
Dengue Negative	95	22.2-49.0	36.3 +/- 3.9

The mean hematocrit values of dengue positive cases were 34.2+/-5 and dengue negative cases were 36.3+/-3.9.

DISCUSSION

Dengue virus infection has emerged as one of the most important emerging arboviral infections world-wide[8]. Around 2.5- 3 billion people live in areas where dengue virus can be transmitted. Each year approximately 50 million infections occur, with 0.5 million cases of DHF and at least 12,000 deaths, mainly among children[9].

In our study sixty two percent of the suspected cases with fever were dengue positive. Hence in every case of fever with myalgia one of the important differential diagnoses to be kept in mind is dengue virus infection. Other studies by Huber *et al* also show similar association [10].

Dengue infection in India was first reported in 1780 at Chennai, India. Today dengue virus and all its clinical forms are documented in almost all parts of India [11].In an area where dengue virus is endemic, cases are more seen in children and young adults as compared to the older population, who may have become immune to the virus[3]. The complications of DHF/DSS contribute significantly to the morbidity and mortality among the pediatric age group. In our study most of the dengue positive cases were in the age group of 1-20 years. This was similar to other studies conducted by Gore MM *et al* [12] .

Positive cases were more among males than in females; this was comparable to other studies by Vijaykumar *et al* [13] . Male predominance may be due to their increased exposure to the bite of *A. aegypti*, due to their clothing habits and outdoor activities.

The disease also shows seasonal distribution with cases more during the months of August- November, the monsoon and post-

monsoon season. This can be attributed to the presence of conducive environment for the breeding of *A. aegyptii* mosquito. Similar seasonal distribution was also seen in other studies as by Rao CVRM *et al* [14].

In our study the majority of the patients had dengue fever (79%), the remaining had the more severe forms i.e., DHF (17%) and DSS (4%). This corresponds to a study by Neeraj *et al* [3].

Among the patients examined all of them had fever. The other common symptoms noted were, headache, joint pain, retro-orbital pain, myalgia, backache, hemorrhagic manifestations, splenomegaly, hepatomegaly and CNS symptoms. This was similar to study conducted by Khan *et al*[15].

One of the important markers for dengue infection is decrease in the platelet count. However in our study thrombocytopenia was more seen in DHF/DSS, as compared to dengue fever cases. Other studies, i.e. Cherian T *et al* [16] have shown high association between dengue illness and thrombocytopenia. This variation may be due to the fact that ours is a tertiary care center. Cases which come here are usually referred by other hospitals in the district, and some of the patients undergo some form of treatment.

Even in the mean hematocrit values of dengue positive cases not much variation was noted. This may be due to the non-availability of pre-illness hematocrit to compare, modification of hematocrit values by treatment (IV fluids, blood transfusion, colloid transfusion) and non-availability of serial hematocrit values

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