



A Study on Clinical Profile and Laboratory Diagnosis of Dengue Patients

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Abstract

Background: Dengue is a highly endemic infectious disease of tropical countries and is rapidly becoming a global burden. It is clinically similar to many other acute febrile illnesses, in which serological test plays an important role in early diagnosis and management. The spectrum of illness is broad and ranges in severity from mild symptoms to death. Changes in dengue epidemiology and increase in incidence rates (with and without co-morbidities) have lead WHO to propose a new dengue classification system according to disease severity.

Objective: The study was aimed at determining the seropositivity in clinically suspected dengue cases and also to study the clinical profile of dengue positive patients.

Materials and Methods: In this study, 2968 patients with Pyrexia of unknown origin (PUO) were included. Rapid Card test and Enzyme Linked Immunosorbent Assay (ELISA) were used for testing serum of patients.

Results: Of 2968, 521 were clinically suspected cases of dengue. 228 were confirmed to have dengue. Based on the 228 serological positive test, 126 (55%) were with dengue with warning signs followed by 74 (33%) dengue without warning signs and severe dengue 28 (12%)

Conclusion: Dengue fever continues to be a major public health problem. The study showed that 521 were clinically suspected cases of dengue and 228 were confirmed to have dengue. The study highlights that in most dengue endemic countries, access to diagnostic laboratories is limited and dengue diagnosis may rely solely on clinical recognition. Moreover, even where diagnostic laboratory services are available, virological tests are requested only upon a clinical suspicion of dengue, based on the presenting symptoms and signs.

Keywords: Dengue fever, new dengue classification, Pyrexia of unknown origin (PUO), Seropositivity.

Introduction

Dengue is a Spanish homonym of the Swahili phrase “ka dinga pepo” meaning cramp like seizure caused by an evil spirit that emerged during the Caribbean outbreak (1827-1828).¹

Dengue is caused by a positive stranded RNA (Ribo Nucleic Acid) virus of the Flaviviridae family with four distinct serotypes (Dengue Virus 1 to 4), that are closely related antigenically.² Dengue viruses are disseminated in nature simply

by a man-mosquito-man cycle. Female *Aedes aegypti* has been the most important epidemic vector in the tropical and subtropical region³ A dramatic worldwide expansion of the DENV has occurred due to rapid urbanization, increase in international travel, lack of effective mosquito control measures, and globalization.⁴ The spectrum of clinical illness may range from asymptomatic disease to broad range of syndromes with severe clinical manifestations. Most patients have a febrile phase lasting, for 2-7 days accompanied by generalized body ache, myalgia, arthralgia and headache. This is followed by critical phase of about 2-3 days duration. During this phase, there is risk of developing DHF/DSS which may prove fatal if prompt and appropriate treatment is not provided.⁵ These three conditions likely represent progressively severe stages of a continuous dengue disease spectrum. They are based on traditional WHO classification case definitions. The new classification based on single parameter allows better case capture.

WHO suggested dengue case classification and levels of severity (WHO, 2009) ⁵

I. Non severe dengue without warning signs :

(Probable dengue)

- a) Live in /travel to dengue endemic area.
- b) Fever and 2 of the following criteria:
 - 1) Nausea/ Vomiting,
 - 2) Rash,
 - 3) Body aches and pains,
 - 4) Tourniquet test positive,
 - 5) Leukopenia
- c) Laboratory-confirmed dengue (important when no sign of plasma leakage)

II. Dengue with warning signs

1. Abdominal pain or tenderness
2. Persistent vomiting
3. Clinical fluid accumulation
4. Mucosal bleed
5. Lethargy restlessness
6. Liver enlargement more than 2 cm
7. Laboratory: increase in Haematocrit concurrent with rapid decrease in platelet count

* requiring strict observation and medical intervention

III. Severe dengue should be considered if the patient is from an area of dengue risk presenting with fever of 2-7 days plus any of the following features:

Severe plasma leakage leading to shock

Fluid accumulation with respiratory distress

2. Severe bleeding as evaluated by clinician

3. Severe organ impairment

Liver: Liver enzymes AST or ALT \geq 1000 Units

CNS: impaired consciousness

Heart and other organs

During the early stages of the disease, virus isolation, nucleic acid or antigen detection can be used to diagnose the infection. The detection of IgM antibody to dengue virus by ELISA is the serological test of choice for most laboratories.⁵ Immediate bedside diagnosis would have an increasingly important role with the development of antiviral therapies, because the window of opportunity for antiviral therapy in dengue may be limited because of short-lived viremia.⁶ NS1Ag strips that can provide results within 15 minutes, could serve as a bedside diagnostic tool.⁷

Management of dengue is symptomatic and supportive. Encourage oral intake of fluids. Bed rest and cold sponging is used to keep temperature below 37⁰c.⁵ Patient should be closely monitored for the initial signs of shock. Serial haematocrit determinations are needed from the third day until the temperature has remained normal for one to two days.⁸

To reduce or prevent dengue virus transmission there is currently no alternative to vector control. *Aedes* mosquitoes breed in the stagnant water containers and discarded junk materials in and around houses. So *A. aegypti* requires the use of a combination of vector control methods, notably environmental management methods and chemical control methods based on the application of larvicides and adulticides space sprays.⁵

Dengue vaccine development has been a challenging task due to existence of four antigenically distinct dengue virus serotypes, each

capable of eliciting cross-reactive and disease-enhancing antibody response against the remaining three serotypes. Recently, Sanofi Pasteur's chimeric live-attenuated dengue vaccine (Dengvaxia) has been approved in Mexico, Brazil, and Phillipines for usage in adults between 9 and 45 years of age.⁹

Materials and Methods

The present study was carried out over a period of 16 months from February 2010 to June 2011. It included patients clinically suspected of dengue admitted in the wards of a tertiary care Hospital, Mumbai.

Blood samples of the clinically suspected cases of dengue were received in the laboratory. Serum was separated from the blood samples and transferred into labelled sterile plastic vials. These samples were used to confirm the diagnosis of dengue with serological tests and later stored at 4⁰C. All the serum samples were tested by 1.) Rapid card test: It is a rapid solid phase immunochromatography test for the qualitative detection of dengue NS1 Ag and differential detection of IgM and IgG antibodies to dengue virus from patient's serum samples. 2.) ELISA: All the serum samples were tested for I) IgM and II) IgG antibodies by "PanBio dengue capture ELISA test".

Results

The study carried out in a tertiary care hospital, included blood samples of clinically suspected cases of dengue admitted in the wards, during a 16 month period from February 2010 to June 2011. During the study period, a total of 2,968 patients with Pyrexia of unknown origin (PUO) were admitted to the wards of the hospital. Of these, 521 were clinically suspected cases of dengue. Serum samples from the 521 patients were tested for the presence of NS1 antigen and IgM, IgG antibodies by the rapid immunochromatography card test. They were also subjected to capture ELISA for IgM and IgG antibodies. 228 cases were confirmed to be dengue positive by these methods.

Table 1: Details of distribution of 228 serologically positive dengue cases into three categories: (WHO, 2009)

Categories of Dengue	Number (N)	Percentage (%)
Dengue without warning signs	74	33
Dengue with warning signs	126	55
Severe dengue	28	12
Total	228	100

A majority of cases (55 %) were having DF with warning signs.

Table 2: Details of distribution of symptoms in three categories of dengue infections (WHO, 2009)

Symptoms	Total		Dengue without warning signs		Dengue with warning signs		Severe dengue	
	N(=228)	%	N(=74)	%	N(=126)	%	N(=28)	%
Fever	228	100	74	100	126	100	28	100
Vomiting	111	48.7	33	44.6	59	46.8	19	67.9
Bodyache and Pain	186	81.6	66	89.2	106	84.1	14	50
Abdominal pain*	79	34.6	0	0	59	46.8	20	71.4
Bleeding*	40	17.5	0	0	29	23	11	39.3
Restlessness*	65	28.5	0	0	65	51.6	12	42.8

* Warning signs

Fever was present in 100 % cases in all three categories.

Body ache & pain was present in 89.2 % cases in dengue without warning signs and 84.1 % in dengue with warning signs.

Abdominal pain was the second common clinical presentation (71.4%) in cases with severe dengue

followed by vomiting (67.9 %) and bleeding (39.3 %) of which abdominal pain and bleeding were the warning signs.

Table 3: Details of signs in different categories of dengue infections (WHO, 2009)

Signs	Total (N=228)		Dengue without warning signs (N=74)		Dengue with warning signs (N=126)		Severe dengue (N=28)	
	N	%	N	%	N	%	N	%
Shock	16	7	0	0	0	0	16	57.1
Pleural effusion	30	13.2	0	0	22	17.5	8	28.6
Ascitis	18	7.9	0	0	12	9.5	6	21.4
Rash	118	51.8	24	32.4	71	56.3	23	82.1
Hepatomegaly	76	33.3	0	0	65	51.6	11	39.3
Jaundice	36	15.8	0	0	20	15.9	16	57.1
Altered sensorium	19	8.3	0	0	0	0	19	67.9

Hepatomegaly (51.6 %) was the commonest warning sign in cases of dengue with warning signs.

Rash (82.1%) was the commonest clinical finding in cases of severe dengue.

Table 4: Details of laboratory findings in different categories of dengue infections (WHO, 2009)

Laboratory findings	Total		Dengue without warning signs		Dengue with warning signs		Severe dengue	
	N(=228)	%	N(=74)	%	N(=126)	%	N(=28)	%
SGOT/SGPT ≥ 1000units	5	2.2	0	0	0	0	5	17.9
Serum Bilirubin > 3 mg/dl	36	15.8	0	0	20	15.9	16	57.1
Serum creatinine	10	4.4	0	0	0	0	10	35.7
Leucopenia ≤ 4000/cu.mm	46	20.1	30	40.5	9	7.1	7	25
Haematocrit ≥ 20%	79	34.6	0	0	55	43.7	24	85.7
Platelet count < 1.5 Lakhs/cu mm	162	71.1	40	54.1	94	74.6	28	100

Elevation of serum glutamate oxaloacetate transaminase (SGOT)/ serum glutamate pyruvate transaminase (SGPT) ≥ 1000 units in 17.9 % of severe dengue cases.

Thrombocytopenia was seen in 74.6 % cases of dengue with warning signs and 100 % cases of severe dengue.

Table 5: Total no. of serologically positive cases by rapid, ELISA and by both the tests

Rapid test	ELISA test	Rapid +ELISA
222	171	228

Discussion

Dengue is the most rapidly spreading mosquito-borne viral disease in the world. In the last 50 years (1960-2010), incidence of dengue has increased 30-fold with an expanded geographic

distribution of both the viruses and the mosquito vector to new countries and from urban to rural settings.⁹

All the 521 samples were tested in the laboratory by rapid card test and ELISA test. Of the 521 samples, 228 tested positive for dengue by these tests. Based on the 228 serological positive test, 126 (55%) were with dengue with warning signs followed by 74 (33%) dengue without warning signs and severe dengue 28 (12%) as shown in table 1.

The most frequent symptoms observed in dengue positive cases in the present study (Table 2) was fever 100% in all the three categories (WHO, 2009) and Bodyache 81.6%, as observed in other studies [Padabidri et al, (1993)¹⁰ fever 100% & arthralgia 76% in 1993; Neeraja et al, (2006)¹¹ fever 100% & myalgia 53%]. The other symptoms in decreasing order of frequency were vomiting in 48.7%, abdominal pain in 34.6%, restlessness/lethargy in 28.5%, hemorrhagic manifestations in 17.5%.

In an outbreak of DHF in Indonesia studied by Richards et al (1993)¹² predominant symptoms were fever (100%), vomiting 48%, abdominal pain 39% and body ache 39%.

In the present study hepatomegaly was observed in 33.3% of the patients. Rash was present in 51.8%. Pleural effusion and ascitis were found in 13.2% and 7.9% patients respectively, which was similar to other Indian studies [Sharma et al (1998)¹³ and Narayan et al (2002).¹⁴]. In the present study altered sensorium was present in 8.3% followed by shock in 7% as shown in table 3.

In a study of Dengue in Andhra Pradesh 2004 (Neeraja et al, in 2006)¹¹ skin rash 41% and altered sensorium 10.3%.

Sharma et al (1998)¹³ reported altered sensorium in 5.1% of DHF/DSS patients.

Jaundice in the present study was noted in 15.8% patients. A study by Chhinaa et al (2008)¹⁵ reported incidence of jaundice in dengue to be 19.5%. While a study by Mohan et al (2000)¹⁵ reported 25%.

In the present study the Biochemistry and pathological profiles were taken from the reports of the patients.

In the present study deranged Liver function test (LFTs) in the form of increased SGOT/SGPT \geq 1000 units was found in 17.9% of severe dengue patients (table 4) which is almost similar to a study in Singapore by Low et al (2011) i.e. 15%.¹⁶ In the present study thrombocytopenia, that is, platelet count less than 150,000/mm³ were found in 71.1% of the total cases and Haematocrit \geq 20% in 34.6% (table 4). Chandrakanta et al (2008)¹⁷ reported the incidence of thrombocytopenia to be 60%.

In the present study leucopenia was observed in 20.1% of patients (table 4). In a study by Agarwal et al (1999)¹⁸ and a study by Padbidri et al (1995)¹⁰ leucopenia was observed in 15% and 25% of the patients respectively.

Knowledge of symptoms associated with severe disease and age group susceptible for severe infection will greatly help in recognizing such cases early and their prompt management. Early recognition of the warning signs (intense continuous abdominal pain, persistent vomiting, and restlessness or lethargy) and early treatment are of utmost importance to reduce case fatality rate (PAHO, 1994).¹⁹

Conclusion

India is endemic for dengue infections and various states / union territories are reporting outbreaks with maximum reported cases in Delhi that is 6,221 in 2010.¹⁵

In the present study a total of 2,968 patients with PUO were admitted to the wards of the hospital. 521 were of clinically suspected dengue fever cases, out of these 228 samples were found to be serologically positive for dengue virus. Majority of cases were of DF with warning signs (55.3%). Among the warning signs, Hepatomegaly (51.6%) and restlessness (51.6%) was the commonest sign in cases of dengue with warning signs.

Among severe dengue cases, altered sensorium (67.9%) was the commonest clinical finding and

liver failure was seen in 57.1% of cases. Thrombocytopenia was seen in 74.6 % cases of dengue with warning signs and 100 % cases of severe dengue. Thus decreased platelet count was directly related to the severity of the disease.

Dengue is an infection that is present in urban, semi urban and rural areas. Our healthcare system is extremely resource poor. The speed, ease and dependability of Immunochromatographic card test (ICT) make it an excellent tool in resource poor peripheral areas where laboratory has to function without great technological backup and still is expected to provide reasonable opinion to the clinician in the management of infections like dengue. BUT in most dengue endemic countries, access to diagnostic laboratories is limited and dengue diagnosis may rely solely on clinical recognition. Moreover, even where diagnostic laboratory services are available, virological tests are requested only upon a clinical suspicion of dengue, based on the presenting symptoms and signs.

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