

Study on the Diagnostic Efficacy of Clinico-Laboratory Parameters in Serologically Diagnosed Cases of Dengue Fever

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Research Article

Abstract: Background: Dengue fever (DF), is the most common mosquito borne arbo-viral infection in humans. The high mortality associated with DF mandates early diagnosis and therapeutic interventions. **Aims:** To study the utility of clinico-laboratory parameters in serologically diagnosed cases of DF and to correlate with serological tests with the progression of disease. **Material and methods:** This is a prospective study done among patients, admitted to a tertiary care facility in South India between 1st March and 31st August 2013, with the diagnosis of Dengue fever (DF) either by dengue NS1 or Ig-M positive test. Their clinical and laboratory parameters were compared for the diagnostic utility with that of NS1 or Ig-M dengue tests in relation to the day of performing the tests. **Results:** 200 serologically diagnosed DF patients were evaluated with 145 positive for NS1 antigen and 55 for Ig-M antibody. 114 patients were male and 86 females. Clinical features of fever with headache, body ache and myalgia were present in 90%, whereas retro-orbital pain in only 40 % of cases. High grade fever and headache were the most specific clinical feature (92%). Almost 94% of NS1 positive cases presented within 4 days of onset of fever. Among NS1 positive cases leucopenia alone had a sensitivity of 55% and specificity 48% with PPV 73%. Thrombocytopenia had sensitivity 89% with PPV 70.45%. Anicteric hepatitis had sensitivity of 75% and specificity 40%. Combination of clinical features with 2 out of four laboratory parameters (high haematocrit, leucopenia, thrombocytopenia, anicteric hepatitis) increased sensitivity to 94%. The chances of developing complications were very less during the first 4 days (3%) and the sensitivity of NS1 was 0-40% after 5 days of symptom, but IGM was 70-100%. **Conclusion:** During the early stages of disease we observed the diagnostic efficacy of clinico-laboratory parameters is similar to NS1 antigen. In a developing country with limited resources, where DF is endemic the routine use of expensive serological investigations is questionable.

Keywords: Dengue Fever, clinico-laboratory parameters.

Introduction

Dengue fever (DF), is the most common mosquito borne arbo-viral infection in human. According to WHO, annually 50 million cases of DF occur world over with a mortality of 2.5%¹. Globally 2.5 – 3 billion individuals live in dengue endemic areas and the number of new cases every year is estimated in 50 million¹. Approximately 112 countries that experience dengue

transmission. Among the estimated 2.5 billion people at risk globally for dengue, about 1.8 billion (i.e., more than 70%), reside in Asia Pacific countries². Currently the disease is endemic in all continents except Europe. DENGUE viruses belong to flavi virus genus which is single stranded RNA viruses. They are of four serotypes which are named as DEN-1, DEN-2, DEN-3 and DEN-4^{3,4}. In spite of antigenic similarity they are different enough to elicit cross-protection only for a few months after infection by any one of them. DF is endemic in most parts of India. The first dengue fever case was reported from Vellore district of Tamil Nadu in 1956. All 4 serotypes have been isolated in India. The disease is most commonly seen in urban area. The reason probably due to the increased breeding places of aedes' aegypti in these areas. At present DEN1 and DEN2 serotypes are widespread in India. The clinical spectrum of DF varies from mild febrile illness to its grave form of dengue haemorrhagic fever (DHF)^{5,6,7}. Classical DF is characterised by the presence of fever, headache, myalgia, retroorbital pain, joint pain, vomiting, nausea and maculopapular rashes which last for 5-7 days⁶. The severity of pain led to the term break bone fever to describe DF. The clinical diagnosis of DF is often easy when it presents with classic symptoms and signs along with high haematocrit, neutropenia, thrombocytopenia and anicteric hepatitis. A delay in diagnosis is often due to its clinical similarity with common tropical infections such as malaria, leptospirosis and various viral infections. The high mortality associated with DF mandates early diagnosis and therapeutic interventions. There are many investigations available but the commonly used one are serological parameters. Though expensive, the specific serological markers like NS1 antigen facilitate early diagnosis of DF⁵. The inadvertent use of these tests, high cost and its non-availability in most centres questions its utility as a standard for diagnosis in a developing country

like ours were the disease in endemic. We intend to study the diagnostic utility of clinico-laboratory parameters in serologically diagnosed cases of DF. If the clinico-laboratory features are found to be significantly correlating with serological tests, use of such expensive tests could be limited to cases with a diagnostic dilemma.

Aims and objectives

1. To study the utility of clinic-laboratory parameters in serologically diagnosed cases of DF.
2. To study the correlation of clinic-laboratory parameters with serological tests with the progression of illness/fever

Material and methods

Source of data Patients admitted in Father Muller Medical College Hospital between 1st March and 31st August 2013 with diagnosis of DF either by dengue NS1 or IG M positive test will be studied. Study design: This is a retrospective study among patients with DF.

Methods This is a retrospective study conducted in patients admitted in Father Muller Medical College Hospital between 1st March and 31st August 2013 with diagnosis of DF either by dengue NS1 or IG M positive test. Case records of patients admitted with diagnosis of DF either by dengue NS1 or IG M positive test will be collected from medical records department. The clinical details and laboratory parameters of all patients will be obtained from the records and captured to the preformatted data sheet. The clinical details include fever, headache, myalgia, joint pain, retro orbital pain, rashes over body and bleeding manifestations were documented. The day of onset of fever and other clinical manifestations also documented. The detailed evaluation of laboratory parameters also documented which includes haemoglobin, packed cell volume, total count, platelet count, serum albumin, serum bilirubin, alanine transaminases (SGOT), aspartate transaminases(SGPT). The details of NS1 or Ig M dengue tests including the day of performing the tests from onset of fever will be documented.

The preformatted data sheet is enclosed.

Clinical features	fever, headache, myalgia, retro-orbital pain, rashes over body, bleeding manifestations
Laboratory parameters	haemoglobin, packed cell volume, total count, platelet count, serum albumin, serum bilirubin, SGOT, SGPT
Serological test	NS1 test / Ig M dengue test

Inclusion criteria

1. Patients above the age of 18.
2. Cases of DF diagnosed either by NS1 antigen or IG M dengue tests.

Exclusion criteria

Patients with coexisting infections, chemotherapy, radiotherapy, renal failure will be excluded.

Data analysis

The data obtained will be analysed by percentage, mean, frequency, chi-square test.

Results

Table 1: Total case

Total cases	NS 1 positive	IG M dengue positive
200	145	55

Table 2: Sex distribution

Total cases	Male	Female
200	114	86

Table 3: Age distribution

Age range	Male	Female
20-29	33	16
30-39	27	24
40-49	26	20
50-59	16	15
60-69	10	10
>70	2	1
Total	114	86

Table 4: Clinical features

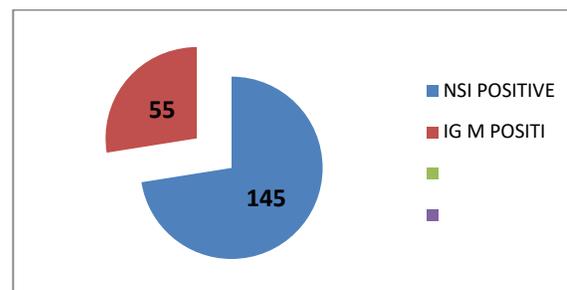
Clinical features	Total cases	Percentage
Fever	200	100%
Headache	184	92%
Myalgia	178	89%
Joint pain	172	86%
Nausea/vomiting	168	79%
Retroorbital pain	88	44%
Bleeding manifestations	12	6%

Table 5: Laboratory parameters

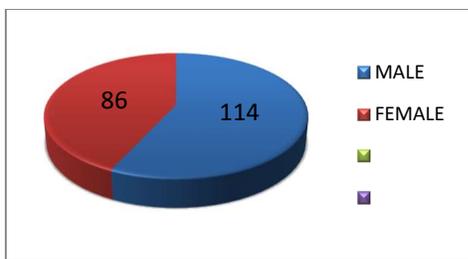
Laboratory parameters	NS1 (145)	IG M (55)
Leucopenia	80	29
Thrombocytopenia	129	50
Anicteric hepatitis	109	109
Combination of clinical with laboratory parameters	137	52

Table 6: Parameters

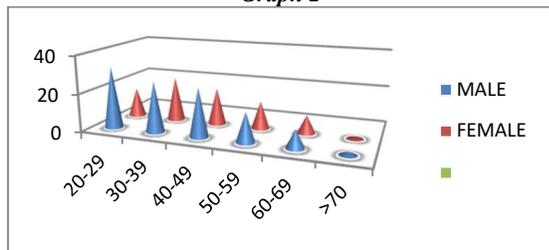
	<4 Days of Symptom Onset	>4 Days
NS1(145)	136	9
IG M(55)	4	51
	140	60



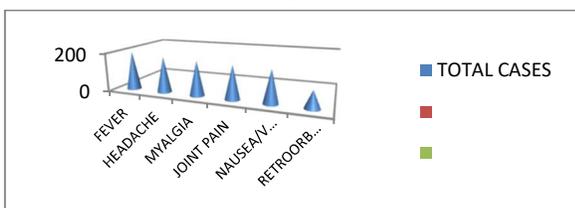
Graph 1



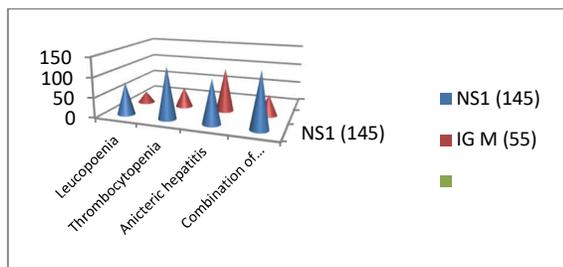
Graph 2



Graph 3



Graph 4



Graph 5

Total cases

Out of 200 serologically diagnosed dengue fever patients, 145 cases were diagnosed through NS 1 antigen and 55 cases through IG M antibody study. (TABLE 1, GRAPH 1).

Sex distribution

Among the 200 patients 114 were males (57%) and rest 86 (43%) were females. (TABLE 2, GRAPH 2)

Age distribution

Among the male population maximum number patients were in the age group of 20-30 and among females maximum population group were between 30-40.(TABLE 3, GRAPH 3)

Clinical features

Among the clinical manifestations, fever was the most common feature with 100% sensitivity followed by

headache which was present in 184 out of 200 (92%),then myalgia/body ache 178(89%) followed by joint pain present in 172(86%). Retro orbital pain was the presenting symptom only in 88 cases out of 200 with sensitivity of 44%. (TABLE 4, GRAPH 4)

Laboratory parameters

The laboratory parameters also significantly contributed to the diagnosis of dengue fever. Among that thrombocytopenia was the most characteristic feature associated with dengue fever. Out of 145 NS 1 ANTIGEN positive cases 129 had thrombocytopenia with a sensitivity of 89% and positive predictive value of 70.45%.Among IGM dengue cases thrombocytopenia was present in 50 out of 55 with a sensitivity of 91%.Leucopenia was another major feature in dengue fever patients. Among the total NS1 antigen positive cases 80 were showed leucopenia with aleucocyte range of 2000-4000mm3. The sensitivity of leucopenia in NS 1 antigen positive cases was 55% and specificity was 48% with PPV of 73%.Anicteric hepatitis was noticed in 109 cases out of 145 NS 1 antigen positives cases (sensitivity 75.17% PPV 91.6%) and 45 cases out of 55 IG M Dengue positive cases(sensitivity 81.82%). The combination of clinical features with 2 out of four laboratory parameters (high haematocrit, leucopenia, thrombocytopenia, anicteric hepatitis) drastically improved the sensitivity of dengue fever diagnosis to 94.48%(137 cases came as positive among total 145 cases)(TABLE 5, GRAPH 5)Total 140 patients were reached the hospital in the first five days. Out of these 140 patients 4 developed complications (3%). Two patients developed hematuria, one patient had ascitis and the other had hypotension. Among the remaining 60 patients i.e. those who presented 5 days after the onset fever 9 were positive for dengue NS1 Antigen and remaining were positive for IGM dengue.(TABLE 6)

Discussion

The high mortality associated with DF mandates early diagnosis and therapeutic interventions. Serological tests like NS1 antigen or IG M/IG G antibody tests are commercially available and very much successful in detecting dengue virus infection in the early stages^{8,9}. Nowadays these serological tests are the commonest diagnostic tool in dengue fever patients. But the question comes when we consider the cost of these serological tests. In a developing country with limited resources, were DF is endemic the routine use of such expensive serological investigations should be reevaluated. There are many studies conducted in the field of DF to demonstrate the sensitivity and importance of serological markers. And most of them found out that NS1 antigen is a highly sensitive tool for detection of dengue fever in the first 3 days of symptom onset. The present study also revealed

the high sensitivity of NS1 in first 4 days of fever/symptom onset (almost 80-100%) with declining sensitivity after 4 days (0-20%). Our study also demonstrated the increasing sensitivity of IG M Antibody test during the course of illness and its sensitivity reaches almost 80-100% after 5 days of fever. These findings in our study are similar to the observations in other studies¹⁰⁻¹². In this study we also noticed that the chance of getting complications were extremely low in the early period of acute febrile illness (3%)^{10,11,13}. The present study conducted in 200 patients demonstrated the male predominance in the occurrence of dengue fever. Out of 200 patients 114 were males (57%) and 86 were females (43%). This is similar to few other studies from Asia, done in DF patients, have tended to find greater male incidence. The reason for this increased incidence in males has been attributed to the prolonged outdoor activities and thereby increased chance of exposure to mosquito bites¹³. These reports from Asia are in contrast to studies in South America, which have found either equal proportions of male and female dengue cases or a greater proportion of female cases¹⁴⁻¹⁷. The commonly affected age group in the present study are younger population. The maximum cases among males were occurred in the age group of 20-29 years and in females between 30-39 years of age. These results also agree with other studies.¹⁸ Based upon experimentally induced infection in adult human volunteers, it was concluded that almost all dengue virus infections are symptomatic¹⁹. Classic dengue fever (DF) presentation is an acute febrile illness which is associated with headache, retro orbital pain, myalgia and joint pain which evoked the term "break-bone fever"²⁰. The febrile period may also be followed by a period of marked fatigue that can last for days to weeks, especially in adults. In the present study revealed that dengue fever invariably associated with high grade fever (100% cases) which last for 5-7 days. This pattern is similar to many other studies²⁰. Following this febrile period there was a period of marked fatigue that can last for days to weeks, especially in adults. The next common symptom was headache (92%) followed by myalgia (89%) and joint pain (86%). Nausea and vomiting were present in about 79% cases. Rashes were present in 50-60% cases. These results are concordance with many other similar studies^{21,22,23}. The incidence of retro orbital pain was only in 44% cases. The laboratory parameters also significantly contributed to the diagnosis of dengue fever. Among this thrombocytopenia was the most characteristic feature associated with dengue fever. Out of 145 NS 1 ANTIGEN positive cases 129 had thrombocytopenia with a sensitivity of 89% and positive predictive value of 70.45%. In IGM dengue cases thrombocytopenia was

present in 50 out of 55 with a sensitivity of 91%. These results are similar to many other studies conducted in the same field. In several studies platelet counts $<100,000$ cells/mm³ were observed in 16 to 55 percent of patients²⁴⁻²⁷. Leucopenia is another major feature in dengue fever patients. Among the total NS1 antigen cases 80 were showed leucopenia with a range of 2000-4000mm³. The sensitivity of leucopenia in NS 1 antigen positive cases was 55% and specificity was 48% with PPV of 73%. Other studies also showed that leukopenia is common in both adults and children with DF and is a useful diagnostic feature²⁸⁻³⁰. Anicteric hepatitis noticed in 109 out of 145 NS 1 antigen positives cases (sensitivity 75.17% PPV 91.6%) and 45 out of 55 IG M Dengue positive cases (sensitivity 81.82%). This study also showed the importance of AST which was affected more than ALT in dengue fever. Similar reports are available from various other studies which also concluded that AST is more elevated than ALT in acute infections of dengue fever²⁸⁻³⁰. The combination of clinical features with 2 out of four laboratory parameters (high haematocrit, leucopenia, thrombocytopenia, anicteric hepatitis) drastically improved the sensitivity of dengue fever diagnosis to 94.48% (137 cases came as positive among total 145 cases) So in this study we demonstrated the classical presentation of dengue fever that is thrombocytopenia, leucopenia and anicteric hepatitis, all has got high sensitivity and positive predictive value. Thrombocytopenia is the commonest laboratory abnormality in DF. By combining the clinical and laboratory parameters we would be able to get an absolute increase in the sensitivity level. 94.5%. This sensitivity is almost equal to that of NS1 antigen in the first 4 days of symptom onset. The classical differentiating feature between dengue with other similar infections like malaria and leptospirosis are the severity of thrombocytopenia, presence of leukopenia and anicteric hepatitis. AST elevation in DF is much more compared to ALT level. Similar reports are available from various other studies which also concluded that AST is more elevated than ALT in acute infections of dengue fever²⁸⁻³⁰. At the end of this study we revealed that the combination of clinical and laboratory parameters are equally sensitive to NS1 antigen during the early period of infection. The usage of these expensive serological tests like NS1 antigen is validated only in the first four days of illness. After 4 days, it is interesting to notice that its sensitivity dropped less than 20%. Not only that the possibility of developing complications is very minimal during the first 4-5 days and the treatment in this period is mainly symptomatic. So a thorough clinical evaluation combined with laboratory parameters can replace NS1 ANTIGEN estimation.

Conclusion

The diagnostic utility of clinico-laboratory parameters is almost equal to serologically diagnosed cases of DF. So we would like to suggest restricting the use of such expensive tests only in those cases with a diagnostic dilemma.

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