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 clinic-laboratory parameters in serologically diagnosed cases of DF and to correlate with serological tests with the progression of disease. **Material and methods:** This is are prospective study done among patients, admitted to a tertiary care facility in South India between 1rstMarch and 31rst 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 withthat 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 NSI 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 leucopoenia 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, were 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 bornearbo-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 million[1]. 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 countries[2]. 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 days[6]. 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 variousviral 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 DF[5].The inadvertent use of these tests, high cost and its non-availability in most centres questions it's 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 1rst March and 31rst 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 1rstMarch and 31rst 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 transamiases (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

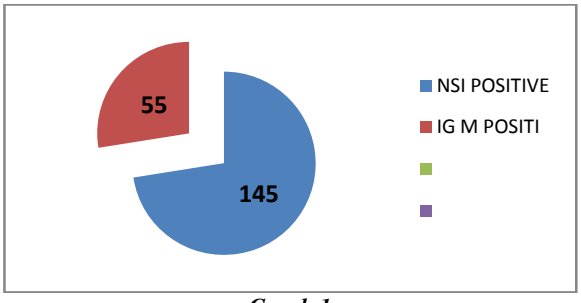
Clinical features	Total cases	Percentage
Fever	200	100%
Headache	184	92%
Myalgia	178	89%
Joint pain	172	86%
Nausea/vomiting	168	79%
Retroorbital pain	88	44%
Bleeding manifestations	12	6%

Table 5: Laboratory parameters

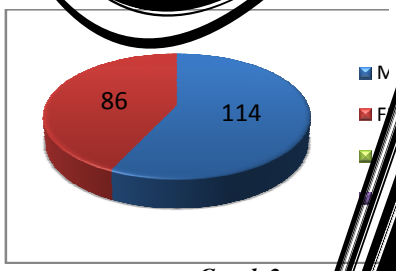
Laboratory parameters	NS1 (145)	IG M (55)
Leucopoenia	80	29
Thrombocytopenia	129	50
Anicteric hepatitis	109	109
Combination of clinical with laboratory parameters	137	52

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 145 cases were diagnosed through NS 1 cases through IG M antibody study. (TABLE 1).

Sex distribution

Among the 200 patients 114 were males 86 (43%) were females.

(TABLE 2, GRAPH 2)

Age distribution

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

Clinical features

Among the clinical manifestations, fever is common feature with 100% sensitivity

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 studies[10-12]. 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 bites[13]. 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 cases[14-17]. 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 symptomatic[19].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 studies[20].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/mm^3 were observed in 16 to 55 percent of patients[24-27].Leucopoenia is another major feature in dengue fever patients. Among the total NS1 antigen cases 80 were showed leucopoenia with a range of 2000-4000mm^3. The sensitivity of leucopeniain 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 feature[28-30].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 fever[28-30].Thecombination 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, leucopoenia 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 levelie. 94.5%.This sensitivity is almost equal to that of NS1antigen 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 fever[28-30].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.

References

1. WHO, World Health Organization (2000).Scientific working group ondengue. Meeting report, Geneva, Switzerland, 3–5 April 2000. WHO: Geneva.
2. Kyle JL, Harris E. Global spread and persistence of dengue. *Annu Rev Microbiol* 2008;62:71-92.
3. Kalayanarooj S, Vaughn DW, Nimmannitya S, Green S, Suntayakorn S, Kunentrasai N, Viramitrachai W, Ratanachu-eke S, Kiatpolpoj S, Innis BL, Rothman AL, Nisalak A, Ennis FA. Early clinical and laboratory indicators of acute dengue illness. J Infect Dis 1997;176:313-21.
4. Potts JA, Rothman AL. Clinical and laboratory features that distinguish dengue from other febrile illnesses in endemic populations. Trop Med Int Health 2008;13:1328-40.
5. Lee MS, Hwang KP, Chen TC, Lu PL, Chen TP. Clinical characteristics of dengue and dengue hemorrhagic fever in a medical centre of southern Taiwan during the2002 epidemic. J Microbiol Immunol Infect 2006;39:121-9.
6. Teixeira MG, Barreto ML. Diagnosis and management of dengue. BMJ 2009; 339:b4338.
7. WHO Regional Office for Southeast Asia. Comprehensive guidelines for prevention and control of dengue and dengue haemorrhagic fever. Revised and expanded version. SEARO Technical Publication Series, New Delhi, India 2011.
8. Rigau-Pérez JG, Gubler DJ, Vorndam AV, Clark GG. Dengue surveillance--United States, 1986-1992. MMWR CDC Surveill Summ 1994; 43:7.
9. Blacksell SD, Mammen MP Jr, Thongpaseuth S, et al. Evaluation of the Panbio dengue virus nonstructural 1 antigen detection and immunoglobulin M antibody enzyme-linked immunosorbent assays for the diagnosis of acute dengue infections in Laos. DiagnMicrobiol Infect Dis 2008; 60:43.
10. Fry SR, Meyer M, Semple MG, Simmons CP, Sekaran SD, Huang JX, et al. The diagnostic sensitivity of dengue rapid testassays is significantly enhanced by using a combined antigen and antibody testingapproach. PLoSNegl Trop Dis 2011;5:e1199.
11. Halstead SB. The XXth century dengue pandemic: need for surveillance and research. World Health Stat Q 1992;45:292–8.
12. Chaterji S, Allen JC Jr, Chow A, Leo YS, Ooi EE. Evaluation of the NS1 rapid test and the WHO dengue classification schemes for use as bedside diagnosis of acute dengue fever in adults. *Am J Trop Med Hyg*. Feb 2011;84:224-8.
13. Monath TP. Dengue: the risk to developed and developing countries. Procular, a normal plasma AST level was a strong negative predictor NatlAcadSci USA 1994;91:2395–400.
14. *Global Health Atlas.* Geneva, World Health Organization (http://apps.who.int/globalatlas/default.asp, accessed 26 December2010).
15. Günther J et al. Distribution of dengue cases in the state of Oaxaca, Mexico, during the period 2004–2006. J*ournal of ClinicalVirology*, 2009;45:218–222.
16. Amélia PA et al. Dengue epidemic in Belém, Pará, Brazil, 1996–1997. *Emerging Infectious Diseases*, 2000, 6(3).
17. García-Rivera EJ, Rigau-Pérez JG. Dengue severity in the elderly in Puerto Rico. *Pan American Journal of Public Health*, 2003;13:362–368
18. Singh MP, Majumdar M, Singh G, Goyal K, Preet K, Sarwal A, Mishra B, Ratho RK.
19. NS1 antigen as an early diagnostic marker in dengue: report from India. Diagn Microbiol Infect Dis. 2010;68:50-4.
20. Sabin AB. Research on dengue during World War II. Am J Trop Med Hyg 1952; 1:30.
21. Rigau-Pérez JG. The early use of break-bone fever (Quebrantahuesos, 1771) and dengue (1801) in Spanish. Am J Trop Med Hyg1998; 59:272.
22. Simmons CP, Farrar JJ, Nguyen vV, Wills B (April 2012). "Dengue". *N Engl J Med*366 (15): 1423–32.
23. Chen LH, Wilson ME (October 2010). "Dengue and chikungunya infections in travelers". *Current Opinion in Infectious Diseases* (5): 438–44.
24. Wolfff K, Johnson RA 0(eds.) (2009). "Viral infections of skin and mucosa". *Fitzpatrick's color atlas and synopsis of clinical dermatology* (6th ed.). New York: McGraw-Hill Medical. pp. 810–2.
25. Schwartz E, Mendelson E, Sidi Y. Dengue fever among travelers. Am J Med 1996; 101:516.
26. Kalayanarooj S, Vaughn DW, Nimmannitya S, et al. Early clinical and laboratory indicators of acute dengue illness. J Infect Dis 1997; 176:313.
27. Potts JA, Rothman AL. Clinical and laboratory features that distinguish dengue from other febrile illnesses in endemic populations. Trop Med Int Health 2008; 13:1328.
28. Trofa AF, DeFraites RF, Smoak BL, et al. Dengue fever in US military personnel in Haiti. JAMA 1997; 277:1546.
29. Souza LJ, Alves JG, Nogueira RM, GicovateNeto C, Bastos DA, Siqueira EW, et al, Aminotransferase changes and acute hepatitis in patients with dengue fever: analysis of 1,585 cases. Braz J Infect Dis 2004; 8:156-163.
30. Kuo CH, Tai DI, Chang-Chien CS, Lan CK, Chiou SS, Liaw YF. Liver biochemical tests and dengue fever. Am J Trop Med Hyg 1992; 47:265-270
31. Trung DT, Thao le TT, Hien TT, Hung NT, Vinh NN, Hien PT, et al. Liver involvement associated with dengue infection in adults in Vietnam. Am J Trop Med Hyg 2010; 83:774-780.