



“ CLINICAL AND HEMATOLOGICAL STUDY OF DENGUE: A TERTIARY INSTITUTIONAL EXPERIENCE ”

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ABSTRACT

Aims and Objectives: This study was designed to investigate the prevalence and clinicopathological profile of Dengue in JA group of hospitals.

Material & Method: Serum samples were obtained from IgM positive dengue patients referred to the various departments during May 2015 to April 2016, and detailed history was taken on a pre-structured datasheet. All samples were tested for hematological and biochemical profile.

Result: Prevalence of dengue was 24.44%, with DF: DHF ratio of 3.34:1. The most affected age group was 11-20 years followed by 21-30 years. Fever (100%), backache and headache (45%) were the most common clinical features. Anemia, thrombocytopenia, bleeding manifestation were present in 78.28%, 22% and 5% dengue positive patients.

Conclusion: Dengue is common cause of illness in febrile patients coincide with post monsoon period. The study serves as the baseline data about circulation of dengue viruses in Gwalior region. Regular continuous surveillance is warranted to monitor the circulation of dengue virus in future.

KEYWORDS : Dengue Fever, Dengue haemorrhagic fever, Clinical

Dengue virus (DENV) infection is one of the mosquito-borne viral diseases with a major impact on public health, globally¹. About 3.5 billion people, 55% of the world's population living in tropical and subtropical regions is at risk, with about 50 million DENV infections occurring annually and approximately 500,000 requiring hospitalization annually².

Dengue is a mosquito-borne viral illness caused by one of the four serotypes of the dengue virus DENV (DENV-1 to DENV-4) belonging to the family Flaviviridae. They are transmitted mainly by the Aedes aegypti mosquito and also by Aedes albopictus³. Dengue has been an urban disease but now has spread to rural areas of India as well⁴. Dengue virus infection is a complex disease with symptoms being difficult to distinguish from other common febrile illnesses during acute phase and can progress from a mild, non-specific viral disease to severe cases characterized by thrombocytopenia, hemorrhage manifestations and hemo-concentration due to plasma leakage. Leucopenia is the most prominent hematological change, sometimes with counts of less than 2×10^7 /microL. However, there are reports of mild leukocytosis at the onset of the disease, with neutrophilia. Lymphocytosis is a common finding, with the presence of atypical lymphocytes. Of biochemical variables, the most frequent changes occur in the liver function tests such as in serum aspartate aminotransferase (AST), serum alanine aminotransferase (ALT), Gamma-glutamyl transpeptidase and alkaline phosphatase levels, and serum albumin concentrations.

In this context, the present study aimed to assess the biochemical and hematological dynamics of patients with dengue in order to increase the sensitivity of screening by healthcare in the most serious cases.

MATERIAL AND METHODS:

Study design:

This study was a hospital-based prospective study conducted for a period of 1 year (May 2015 to April 2016) in Department of Pathology, Gajra Raja Medical College (GRMC) and associated Jayarogya group of hospital (JAH) Gwalior in association with Defense Research and Development Establishment (DRDE) Gwalior. The institutional ethical committee approved the study protocol.

Participants:

The study group consists of 100 patients admitted in hospital with dengue Immunoglobulin M (IgM) positive. Patient suffering from any

infection, viral hepatitis during this period were excluded from study.

Case Definition⁵:

Dengue fever: Acute onset of high fever 3–14 days after the bite of an infected mosquito. Symptoms include frontal headache, retro-orbital pain, myalgias, arthralgias, rash, and low white blood cell count.

Dengue haemorrhagic fever: World Health Organization (WHO) criteria:

1. Fever or recent history of fever lasting 2–7 days.
2. Any hemorrhagic manifestation.
3. Thrombocytopenia: Platelet counts less than $100 \times 10^9/L$
4. Evidence of increased vascular permeability.

Dengue Shock Syndrome: defined as any case that meets the four criteria for DHF and has evidence of circulatory failure manifested by (1) rapid, weak pulse and narrow pulse pressure (≤ 20 mmHg [2.7 kPa]) or (2) hypotension for age, restlessness, and cold, clammy skin.

Method:

Demographic and clinical characteristics of the participants were recorded on a self-designed semi structured per-forma in all these patients. Just after admission, 5ml. of blood was collected aseptically from each patient. 1 ml of clotted blood was used for Immunoglobulin M (IgM) antibody by mac ELISA; remaining 2 ml of collected blood sample was kept in EDTA vial and was used for haematological investigations by manually as well as by 5 part analyser. Rest 3 ml of clotted blood after separating serum was used for biochemical test and NS1 Antigen.

Statistical analyses:

Data were analyzed using SPSS version 20, with a 2-tailed α level of 5%. Statistical analyses were performed by Chi-square tests analyses. The criterion for statistical significance was set at $p < 0.05$, and for statistical trend at $p < 0.10$. Given the small sample size, our hypothesis testing must be considered preliminary rather than definitive.

RESULTS:

1. Epidemiological profile of DENV IgM Positive patients:

Total of 409 cases of clinical fever was admitted in various departments during May 2015 to April 2016 in JA group of Hospitals. 100 cases were positive for IgM; prevalence of dengue seropositive was 24.44%. 80 percent were males (n=80) and 20 percent (n=20) were

female. Male: Female ratio was 4:1. Dengue fever was present in 77% while 23% have dengue haemorrhagic fever. We did not found any case of DSS. The age of all patients ranged from 6 month to 69 years. The mean age was 16 years (SD 10.69) and 24.09 years (SD 12.38) for DHF and DF respectively (Table 1).

| Variable | N | Dengue Fever | Dengue Haemorrhagic Fever | X ² test |
|------------------------|----------|--------------|---------------------------|---------------------|
| Dengue severity | 100 | 77 | 23 | |
| Sex | | | | p <. 722,df 1 |
| Male | 80 | 61(79.22%) | 19(82.60%) | |
| Female | 20 | 16(20.77%) | 4(17.39%) | |
| Age (Years) | | | | p <. 331df 6 |
| 0-10 | 21(21%) | 12(15.58%) | 9(39.13%) | |
| 0-1 | 2(2) | 2(2.59%) | 0(0%) | |
| 1-10 | 19(19%) | 10 (12.98%) | 9(39.13%) | |
| 11-20 | 29 (29%) | 23(29.87%) | 6 (26.08%) | |
| 21-30 | 27(27%) | 22(28.57%) | 5(21.73%) | |
| 31-40 | 17(17%) | 15(19.48%) | 2(8.69%) | |
| 41-50 | 4(4%) | 3(3.89%) | 1(4.34%) | |
| 51-60 | 1(1%) | 1(1.29%) | 0 | |
| 61-70 | 1(1%) | 1(1.29%) | 0 | |

2. Clinical Profile of Dengue Positive patients:

Most common clinical presentation was fever followed by headache /backache/myalgia. The relatively fewer common clinical features were hemorrhagic manifestation, rash, and retro-orbital pain. Bleeding manifestations were more common in DHF and these finding were statistically significant (Table 2).

Table 2: Clinical Profile of Dengue Positive patients

| Clinical feature | DF | DHF | Total | X ² test |
|-------------------------------|-------------|-------------|-----------|---------------------|
| 1.Fever | 77 (77%) | 23 (23%) | 100(100%) | P<. 001 Df 1 |
| 2.Headache/ Backache/ Myalgia | 33 (73.33%) | 12 (26.66%) | 45(45%) | P<. 431, Df 1 |
| 3.Nausea/ Vomiting | 25 (71.42%) | 10 (28.57%) | 35(35%) | P <. 331, Df 1, |
| 4.Retro_ orbital pain | 4 (66.66%) | 2 (33.33%) | 6 (6%) | P <. 535, df 1 |
| 5.Abdominal Pain | 14 (38.88%) | 22 (61.11%) | 36 (36%) | P<. 001 Df 1 |
| 6.Ecchymosis | 0 | 5 (100%) | 5 (5%) | P<. 001 Df 1 |
| 7.Rash | 4 (22.22%) | 14 (77.77%) | 18 (18%) | P<. 002 Df 1 |
| 8.Bleeding Manifestations | 0 | 5 (100%) | 5 (5%) | P<. 001 Df 1 |
| · Epistaxis | 0 | 3 | | |
| · Melena | 0 | 1 | | |
| · Gum | 0 | 1 | | |

3. Biochemical Profile of Dengue Positive Patients:

Study revealed anemia (Hb>13gm%) was present in 93 % of seropositive cases. 70 % dengue seropositive patients were having hemoglobin level between 11-13gm/dl, while 23 % dengue seropositive patients were having hemoglobin level below 11gm/dl. Anemia was more common in DF (96%) than DHF (78%). These findings were statistically significant (p<. 005). 88% of dengue seropositive patients were having hematocrit between 31-50 while 9% dengue seropositive patient was having hematocrit below 30. Only 3 % cases have hematocrit above 50. Hemoconcentration was more common in DHF as compare to DF cases. These findings were statistically significant (p<. 005). Leucopenia was present in 76 % of dengue seropositive cases. 42 % dengue seropositive patients were having TLC between 3000-4000, while 34 % dengue seropositive patients were having TLC below 3000. Leucopenia was more common in DHF (82%) than DF (73%). These findings were statistically not significant (p<. 328). Study revealed 22% of dengue seropositive patients were having platelet count below 100000/cc while 78% dengue seropositive patients were having platelet count above 1,00,000/cc. These findings were statistically significant (p<. 002).

AST was elevated in 36% dengue seropositive patients. AST was more elevated (95.66%) in DHF than DF (18%). ALT was elevated in 32% dengue seropositive patients. ALT was more elevated (91.29%) in DHF than DF (14.27%) (Table 3).

| Parameter | DF | DHF | Total | X ² test |
|------------------------------|-------------|-------------|----------|---------------------|
| 1.Hemoglobin (gm./dl) | | | | |
| <11 | 22 (28.57%) | 1 (4.34%) | 23 | P <.001 Df 2 |
| 11-13 | 53 (68.83%) | 17 (73.91%) | 70 | |
| 13-16 | 2 (2.59%) | 5 (21.73%) | 7 | |
| Total | 77 | 23 | 100 | |
| 2.Hematocrit | | | | |
| >50 | 0 | 03(13.04%) | 3 (3%) | P<.001 Df 2 |
| 31-50 | 68 (88.31%) | 20 (86.95%) | 88 (88%) | |
| <30 | 09 (11.69%) | 0 | 9 (9%) | |
| Total | 77 | 23 | 100 | |
| 3.TLC | | | | |
| 1000-3000 | 28 (36.36%) | 6 (26.08%) | 34(34%) | P<. 328 Df 2 |
| 3000-4000 | 29(37.66%) | 13(56.52%) | 42 (42%) | |
| >4000 | 19 (24.67%) | 4 (17.39%) | 23 (23%) | |
| Total | 77 | 23 | 100 | |
| 4.Platelet Count | | | | |
| <20000/cc | 0 | 1(4.34%) | 1 | P<. 002 Df 3 |
| 21000-30000/cc | 0 | 2(8.6%) | 2 | |
| 31000-40000/cc | 0 | 3(13.04%) | 3 | |
| 41000-50000/cc | 0 | 5 (21.73%) | 5 | |
| 51000-100000/cc | 0 | 11 (47.82%) | 11 | |
| 101000-150000/cc | 77 (100%) | 1 (4.34%) | 78 | |
| Total | 77 | 23 | 100 | |
| 5.AST | | | | |
| Normal Level | 63(81.81%) | 1(4.34%) | 64 | P<. 002 Df 3 |
| 1-2x | 10(12.98%) | 13(56.52%) | 23 | |
| 3-4x | 4(5.19%) | 6(26.08%) | 10 | |
| >4x | 0 | 3(13.06%) | 3 | |
| Total | 77 | 23 | 100 | |
| 6. ALT | | | | |
| Normal Level | 66(85.71%) | 2(8.69%) | 68 | P<. 001 Df 3 |
| 1-2x | 10(12.98%) | 11(47.82%) | 21 | |
| 3-4x | 1(1.29%) | 8(34.78%) | 9 | |
| >4x | 0 | 2(8.69%) | 2 | |
| Total | 77 | 23 | 100 | |

Discussion:

Prevalence of dengue seropositive was 24.44% in our cases, whereas ratio of DF to DHF ratio was 3.34: 1. Our study revealed the most susceptible age group for dengue fever was 11-20 years and followed by 21-30 years suggesting that the individuals in these age groups were actively involved in outdoor activities that increased their chances of exposure to the infective DENV vector bite, however DHF were more in 0-10yrs of age in compare to higher age groups, probable explanation for these finding were passive transfer of antibody from the mother or previous infection.

Fever (100%), backache and headache (45%) were the most common clinical features followed by vomiting (35%) and abdominal pain (36%) in our study; the relatively fewer common clinical features were hemorrhagic manifestation (5%), retro-orbital pain. In study done by Mittal et al 'showed that fever (100%), headache (63%), abdominal pain (71%) and petechial (35.5%) of patients. Fever rash was present in 18 % of cases, it was typical macular or maculopapular, often becoming confluent and sparing small islands of normal skin. The rash was not associated with scaling or pruritus. Pervin et al', reported occurrence of rash in 33 % of patients. Headache, backache and Myalgia were observed in 35 % of patients. Pervin et al, reported myalgia in 84.5% of patients.

In our study, Anemia (Hb <13) was observed in 78.25% of DHF patients and 97.4% of DF. Similarly Hematocrit >50 was observed in 3% of DHF patients whereas, hematocrit >30-50 was observed in 86.95% of DHF patients and 88.95% of DF patients. Thrombocytopenia (platelets < 1,00,000/cmm) was seen in 22 % of patients in our study. The platelet counts in these patients ranged between 10000-1,00,000/cmm. Bleeding manifestations were present in 5 % patients. Ratagiri et al,⁸ reported thrombocytopenia in 82 %, DHF in 60 %, DSS in 22% and DF in 18 % of patients. Leucopenia (TLC <4000) was observed in 82.6 % of DHF patients and 74.02% of DF patient in our study. Ratagiri et al reported leucopenia in 26 % of patients and Benerjee et al⁹ demonstrated no evidence of leucopenia in their studies. This thrombocytopenia may be due to decreased production in bone marrow, temporary bone marrow suppression, virus-antibody complex mediated immune destruction of platelet or increased consumption of platelet induced by secondary infection associated with release of high level of platelet activating factors or increased adhesiveness of platelet to the vascular endothelial cells¹⁰. One important laboratory finding is elevation of liver enzymes, which was reported in various studies. Similarly, in our study, the rise in AST and ALT in was observed in 36% and 32% of patients respectively.

CONCLUSION:

Dengue is the most important arboviral infection of public health significance. Dengue is reported at regular interval from different parts of India. In the present study investigation of patients suspected of having Dengue like illness was carried out during May 15 to April 16. A total of 409 samples were screened and 100 samples are found positive for Dengue IgM antibody indicating a prevalence of 24.44%. The study serves as the baseline data about circulation of Dengue viruses in Gwalior region. Regular continuous surveillance is warranted to monitor the circulation of Dengue virus in future.

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