

Original Research Article

Utility of Haematological Parameters and Reactive Lymphocytes (Virocytes)/Plasmacyatoid Lymphocytes in Peripheral Smear in the Diagnosis of Dengue Fever

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Abstract

Introduction: Dengue fever is the most common Arboviral disease in the world. The four serotypes of dengue virus, 1,2,3 and 4 form an antigenic subgroup of flavivirus. The clinical manifestations of dengue fever range from in apparent to lethal. Various hematological parameters and reactive lymphocytes (Virocytes)/Plasmacyatoid lymphocytes in peripheral smear can help us in the diagnosis of dengue fever.

Aim: The aim of the study is to emphasize on the utility of hematological parameters and peripheral smear reactive lymphocytes (virocytes)/plasmacyatoid lymphocytes in the diagnosis of dengue fever.

Materials and Methods: A study was carried on a series of patients who attended various health care centres in and around Kurnool for a period of one month. Patients presented with complaints of fever, headache, rash and body pains. Then these patients were tested for dengue, then those positive cases hematological parameters and peripheral smear were analysed.

Results: Out of 100 dengue positive cases, 40 cases showed increased hematocrit, 60 cases had leucopenia, 100 cases had thrombocytopenia and 60 cases had relative lymphocytosis. On peripheral smear examination 100 cases showed reactive lymphocytes/plasmacyatoid lymphocytes.

Conclusion: Presence of raised hematocrit, leucopenia, relative lymphocytosis, thrombocytopenia and reactive lymphocytes in peripheral smear helps in the diagnosis of dengue fever.

Keywords: Dengue Fever; Hematological Parameters; Peripheral Smear; Reactive Lymphocyte; Thrombocytopenia.

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Introduction

Dengue fever is caused by virus which belongs to flaviviridae family. Dengue virus infection is transmitted by aedes family mosquitoes [1]. There are four clinical syndromes in dengue fever:

1. Undifferentiated fever
2. Classic Dengue fever

3. Dengue hemorrhagic fever

4. Dengue shock syndrome [2].

Dengue fever can be confused with other fevers like malaria, leptospirosis and typhoid fever [3]. We studied 100 cases of dengue fever to know the utility of hematological parameters and peripheral smear reactive lymphocytes in the diagnosis and differentiation of dengue fever. Increased hematocrit, leucopenia,

thrombocytopenia and reactive lymphocytes give us a diagnostic clue. So early and rapid diagnosis of dengue fever may help in the prevention of lethal complications.

the fully automated hematology analyzer with a five part differential. The peripheral smears were stained with Leishman stain.

Materials and Methods

Our study was prospective study. 100 cases of dengue fever were studied that attended the health care centres in and around Kurnool for a period of one month. Patients presenting with fever, headache, body pains, rash and who tested positive for dengue infection were included in the study. Patients with fever but were negative for dengue testing were excluded. The test for dengue was done using a rapid immunochromatographic test for the qualitative detection of NS1 antigen and differential detection of IgM and IgG antibodies to dengue virus in human serum. 5ml of blood was collected for hematological and serological investigations. 2ml of blood in EDTA vacutainers and 3ml of blood in plain vacutainers. The hematological investigations include complete blood counts and peripheral smear. The blood counts were performed on

Results

Among 100 dengue serology positive patients the majority were NS1 positive and very few showed positive for only antibodies or a combination of both antigen and antibodies (Table 1). Of the 100 patients studied 40% were showed increased hematocrit, 60% were showed leucopenia, 60% were showed relative lymphocytosis, 100% were showed thrombocytopenia and 100% have shown reactive lymphocytes (virocytes)/plasmacytoid lymphocytes in the peripheral smear examination (Table 2). Reactive lymphocyte (virocyte) in smear appear as increased cell size, increased amount of cytoplasm with increased basophilia and irregular cleaved nucleus with slightly open chromatin (Figure 1,3,4). Plasmacytoid lymphocyte in smear appear as with basophilic cytoplasm and peripherally placed nucleus resembling plasma cell (Figure 2).

Table 1: Pattern of antibody positivity to dengue virus in human serum

Antigen/antibody	No. of cases
NS1+	75
NS1+, IgM+	07
NS1+, IgG+	02
IgM+, Ig G+	02
IgM+	10
IgG+	04

Table 2 : Table showing no. of cases showing reactive lymphocytes

Reactive Lymphocytes	No. of cases
Absent	0 cases
<10%	12(12%)
>10%	88(88%)



Fig. 1: Reactive lymphocyte (virocyte)



Fig. 2: Plasmacytoid lymphocyte

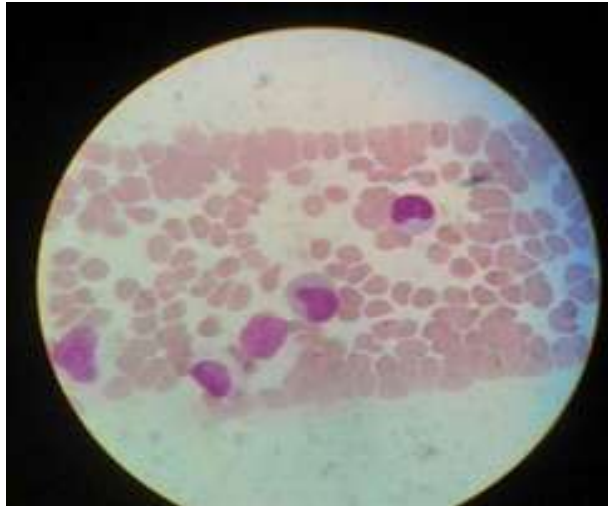


Fig. 3: Reactive lymphocyte (virocyte)

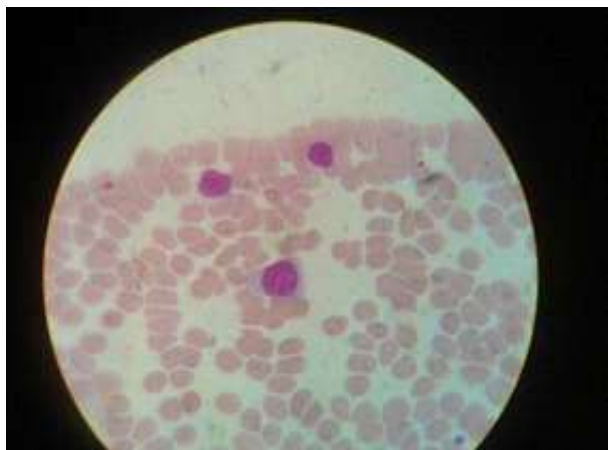


Fig. 4: Reactive lymphocyte (virocyte)

Discussion

Dengue fever can be confused with other fevers like malaria, typhoid and leptospirosis [4]. In such scenario along with the patient hematological parameters like increased hematocrit, leucopenia, relative lymphocytosis and thrombocytopenia and reactive lymphocytes (virocytes)/plasmacytoid lymphocytes in the peripheral smear examination helps us in early and rapid diagnosis of dengue fever.

In our study 60% of cases demonstrated leucopenia correlating with study done by mehta et al.[2] and chakravarthi et al.[5] whereas Archana shetty et al.[6], Arshad et al.[7], Kinjal et al.[8], Malathesha et al. [9], and Banerjee et al.[10] studies have reported lower proportion of leucopenic cases. Leucopenia in dengue patients was due to virus induced bone marrow suppression.

In our study 100% cases were showed platelet count <1,50,000. Rasmavathi et al.[11] and Dutta et al.[12], reported thrombocytopenia in 77.8% and 71.3%

respectively. Decrease in platelet count was due to depression of bone marrow, platelet destruction caused by autoantibodies released by plasma cells and reactive lymphocytes which are activated by infected B cells and cytokines like IL-6 and due to direct infection of the bone marrow megakaryocytic precursors [13]. According to Khan et al thrombocytopenia was a persistent finding in dengue, however an absence should not rule out the possibility of dengue infection [14]. Hematocrit levels were raised in 40% of cases in our study. Jameel et al.[15] and Rashmi MV et al.[14] reported a lower incidence of raised hematocrit levels of 26% and 23% respectively in their study. Main cause of hemoconcentration may be due to plasma leakage because of cytokine release [16]. The discrepancy may be due to difference in clinical course.

We have studied peripheral smears in all our dengue positive cases. Reactive lymphocytes(virocytes) and plasmacytoid lymphocytes were seen in 100% of our cases. Rasmavathi et al reported atypical / activated lymphocytes in 85% of cases and jameel et al reported in 93% of cases. Various terminology were used for these reactive lymphocytes [17]. However in the literature there is no data about lymphocyte morphology on peripheral smear in the dengue fever [6]. The importance of these lymphocytes is that higher the counts the severe the dengue [18]. The immunopathogenesis of the dengue fever may be due to aberrant immune activation causing inversion of CD4/8 counts, cytokine overproduction, monocytosis and reactive lymphocytosis [13,15]. According to study done by Archana et al. [6] morphological study of reactive lymphocyte proves to be a highly useful and cost effective adjunct in prognosticating patients with dengue fever. In their study they concluded that reactive lymphocytes slightly larger than mature lymphocyte and with skirting and scalloped cytoplasmic edges are associated with higher platelet counts and lymphopenia, while larger lymphocytes with rounded edges and intense basophilia of the cytoplasm are significantly associated with lower platelet counts and absolute lymphocytosis [6].

Conclusion

From the above study it was concluded that hematological parameters like increased hematocrit, leucopenia, relative lymphocytosis, thrombocytopenia and reactive lymphocytes (virocytes)/plasmacytoid lymphocytes in the peripheral smear are very important and cost effective in rapid diagnosis of dengue fever. These reactive lymphocytes along with other hematological parameters helps us in differentiating from other fevers like malaria, typhoid and leptospirosis, this can also prevent unnecessary antimalarial exposure and resistance.

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