

# DECIPHERING THE INTERRELATIONSHIP OF HEMATOLOGICAL AND SEROLOGICAL PARAMETERS IN DENGUE PATIENTS: AN IN-DEPTH INVESTIGATION

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## Abstract

Background. The World Health Organization has classified symptomatic dengue into two groups, dengue with or without warning signs and severe dengue. For the conclusive diagnosis of dengue virus detection, a variety of diagnostic techniques are employed, including molecular detection, virus isolation, and virus-specific serological testing. The immunochromatography test (ICT) for active dengue detection is simple to use, with a quick turnaround time. However, it has low sensitivity and specificity and also a higher cross-reactivity, which increases the number of false positive results. Therefore, in addition to quick dengue tests using ICT techniques, hematological and biochemical parameters can be useful as a supporting test for dengue diagnosis. The study aims to identify predictive hematological and biochemical parameters in each serological phase of dengue infection (NS1 and IgM) that can aid in dengue diagnosis, severity assessment, and patient management using rapid serological tests. Methodology: This was a retrospective study of seropositive dengue cases (176 cases) conducted at Saveetha Medical College and Hospital between November 2021 and March 2023. Hematological analysis was done using the SYSMEX XN1000 six part analyzer, and various parameters such as WBC count, RBC count, and platelet parameters were evaluated. The subjects were divided into various serogroups based on their seropositivity, and correlation between different hematological and serological parameters was done using SPSS software, and ANOVA test was used to calculate the p value. Results: Among our study population, 80 were NS1 positive, 140 were IgM positive, and 42 were both IgG positive. We analyzed the positivity of the serological test with biochemical parameters. There was a significant correlation between a fall in platelet count and the number of positive serological tests. When more than one serological test is positive, there was a greater fall in the platelet count and the same correlation was seen with WBC. Lymphocyte count was much lower among patients with NS1 positive alone. Other parameters don't have significant correlation with serological positivity. Conclusion: In summary, our research indicates that specific haematological and biochemical markers can forecast dengue infection outcome, which can help doctors diagnose and treat patients appropriately. Platelet count, WBC count, and lymphocyte count, if rightly and timely assessed, can be of value for better care of dengue patients.

**Keywords:** Dengue, Hematology, Serology.

## INTRODUCTION

Dengue fever is an arthropod borne viral disease caused by mosquito borne single stranded RNA positive Dengue virus of the family Flaviviridae, genus Flavivirus. The disease is transmitted by the female *Aedes aegypti* and *Aedes albopictus* mosquitoes, which transmit the virus during the blood meal.

Dengue virus is a 50 nm, single-stranded RNA virus with a genome approximately 11 kb in length [1]. The virus has three structural genes that encode the capsid protein (C), membrane protein (M), and envelope protein (E), as well as seven nonstructural (NS) genes that encode the NS proteins, including NS1, NS2A, NS2B, NS3, NS4A,

NS4B, and NS5 [2]. Dengue virus is primarily transmitted by the vectors *Aedes aegypti* and *Aedes albopictus*, and it is most common in tropical and subtropical regions. An infection with dengue is typically asymptomatic and self - curable [3]. The World Health Organization has classified symptomatic dengue into two groups, dengue with or without warning signs and severe dengue [4]. The incubation period of the virus ranges from 3 to 10 days, typically 5–7 days, and follows a clinical course as a biphasic febrile phase lasting 2–7 days, a critical phase lasting 24–48 hours, and a convalescent phase [5]. With an estimated 400 million people infected each year, the disease now affects over 100 countries, the majority of which are in Asia, accounting for 70% of the disease burden [3].

In recent years, dengue fever has become a major international health problem affecting tropical and sub-tropical regions around the world - especially urban and peri-urban areas. The global annual incidence is about 390 million dengue infections per year, of which 96 million manifest clinically. It presents a diverse clinical picture that ranges from asymptomatic illness to dengue fever to the severe illness of dengue hemorrhagic fever / dengue shock syndrome. The clinical symptoms serve as the basis for the preliminary diagnosis. Serological testing is a standard procedure for diagnosis confirmation.

To diagnose the acute infection, the preferred method used these days is the detection of Non Structural 1 (NS1) protein and IgM and IgG anti dengue virus antibodies using in vitro immunochromatographic assays. NS1 is a highly conserved glycoprotein that is present in high concentrations in the serum of dengue-infected patients from Day 1 to Day 9 after the onset of fever. When a person has a primary dengue infection, IgM is visible three to five days into the illness and lasts for two to three months, while IgG appears by day fourteen and lasts for life. Consequently, individuals suffering from secondary infections will typically exhibit a positive IgG result, but not necessarily a positive IgM result. There is an alteration in various hematological parameters during dengue fever. In dengue cases, leukopenia, thrombocytopenia, and the presence of atypical lymphocytes are frequently seen.

In dengue hemorrhagic fever, elevated hematocrit (Hct) is a known finding. The incidence and prevalence of seropositive dengue cases have been on a steady increase over the past decade. Therefore, monitoring these parameters can help reduce dengue related morbidity and mortality. The current study sought to assess the different hematological and serological parameters associated with dengue fever and investigate the relationship between them.

## **MATERIALS AND METHODS**

This was a retrospective study of seropositive dengue cases (176 cases) conducted at Saveetha Medical College and Hospital between November 2021 and March 2023. Hematological analysis was done using SYSMEX XN1000 and various parameters (WBC, RBC and Platelet parameters) were evaluated. The subjects were divided into various serogroups based on their seropositivity, and correlation between different hematological and serological parameters was done using SPSS software, and ANOVA test was used to calculate the p value.

## RESULTS

In our study of 179 patients, the most common incidence was in the age group of 21-30 years (n = 69), followed by 11-20 years (n = 34), and 31-40 years (n = 22). Only 10 patients were above 60 years. 118 patients were male, and the rest, 61 were female, in our study.

In our study, 17 patients had hemoglobin less than 10 g/dL, 95 patients had a normal WBC count, leucopenia was seen in 66 patients, and the rest had leukocytosis. All patients had thrombocytopenia, while 101 patients had platelet count less than 1 lakh, among which 57 had count less than 50,000.

Among our study population, 80 were NS1 positive, 140 were IgM positive, and 42 were both IgG positive. We analyzed the positivity of serological test with biochemical parameters. The results are shown in Table 1.

**Table 1: Correlation between biochemical and serological parameters**

Mean	IGM Positive	IGG Positive	NS1 Positive	IGG+IM Positive	IGG+NS1 Positive	IGM +NS1 Positive	TRIPLE Positive	P Value
HB	13.06	13.64	13.88	13.64	14.3	14.46	14.3	0.216
HCT	39.33	41.11	42.35	41.32	42.37	42.85	42.31	0.521
Platlet	0.95	0.7	0.84	0.7	0.57	0.56	0.51	0.021*
WBC	6264	6229	4496	6242	5478	5353	5478	0.043*
Neut	50.3	44.93	54.9	44.92	48.66	50.1	48.6	0.124
Lymp	41.22	46.4	37.19	46.4	41.24	40	41.43	0.04*
Mono	6.15	6.58	6.57	6.58	7.34	7.14	7.34	0.141
Eos	1.87	1.47	1.5	1.47	1.27	1.21	1.274	0.214
Baso	0.35	0.37	0.32	0.37	0.36	0.37	0.37	0.856

There was a significant correlation between a fall in platelet count and the number of positive serological tests. The fall in platelets is greater when more than one serological test is positive, and the same correlation was seen with the WBC count. Lymphocyte count was much lower among patients having NS1 positive alone. Other parameters don't have significant correlation with serological positivity.

## DISCUSSION

Our study demonstrated the strong correlation between dengue infection and age, which has also been found in other studies [6, 7]. Moreover, this study showed people in the age group of 21-30 years are more susceptible to infection, which is supported by other studies [8, 9]. Contrary to what Pun et al. [7] reported, our study also revealed the negligible finding of dengue virus infection with sex.

Regular blood and biochemical markers that could be connected to dengue cases were examined in our study. Our study showed a significant relationship between specific serological courses of disease and the parameters analyzed, including lymphocyte count, leucopenia, and thrombocytopenia.

According to the World Health Organization, the most significant laboratory parameters measured during dengue infection are hematocrit and thrombocytopenia [10]. However, hematocrit did not significantly correlate with IgM or NS1 during the serological course of our investigation. Similar but insignificant results have been reported by other studies; this could be because our study only included patients with mild primary active infections due to dengue fever. Because of this, there is a lower

possibility of plasma leakage, which does not suggest abnormal hematocrit readings [11, 12]. Our study found that thrombocytopenia, which has been shown in numerous studies to be positively correlated with dengue severity, is still significant [13–16]. This decrease in platelets could be attributed to low production or increased platelet destruction via activation of the complement factor C3 and subsequent binding of the C5b-9 complex to the platelet surface [17].

In another study, leukopenia with monocytosis (in NS1 only) and leukopenia with lymphopenia (in both NS1 + IgM) were also observed in certain serological durations of illness. Other studies have also noted leukopenia with lymphopenia and leukopenia with monocytosis. Leukopenia is caused by virus-induced destruction of WBC and inhibition of myeloid progenitor cells; leukopenia with monocytosis is explained by monocytes phagocytosing and presenting the antigen to T-helper cells, which results in monocytosis.

Platelets are involved in hemostasis, tissue repair, and infection. They are the natural sources of various growth factors like Platelet Derived Growth Factor (PDGF), vascular Endothelial Growth Factor (VEGF), Insulin like Growth Factor-1 (IGF-1) and Transforming Growth Factor  $\beta$  (TGF- $\beta$ ). These growth factors have an important role in the inflammation, angiogenesis, repair, and regeneration of tissues [26]. The activation of platelets caused morphological alterations in the mean value of platelet count, which was also reduced in this study. The fall in platelet count is the result of varied mechanisms such as the virus suppressing the bone marrow, causing reduced production, and platelet destruction caused by the anti-dengue antibodies, peripheral consumption, and viral replication.

## CONCLUSION

This study has revealed that certain hematological and serological markers can potentially predict how a dengue infection will progress in a patient. This can in turn aid the physicians in the diagnosis and proper patient management. Platelet count, WBC count, and lymphocyte count, if rightly and timely assessed, can be of value for better care of dengue patients. Future studies to assess the clinical significance and utility of the correlation between hematological and serological parameters in prognosticating and managing dengue patients are recommended.

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