



ROLE OF HS-CRP, ALT AND AST AS MARKER FOR DENGUE FEVER SEVERITY AT TERTIARY CARE HOSPITAL, RAJASTHAN

General Medicine

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KEYWORDS

INTRODUCTION:

Dengue is an emerging illness across globally, accounting for approximately 400 million cases annually in 129 countries [1], although only about one forth are symptomatic [2]. In India, dengue is endemic in various region like Bihar, Assam and Rajasthan. dengue outbreak is associated with high morbidity and mortality, resulting in a significant socioeconomic impact. According to the National Centre of Vector Borne Disease Control (NCVBDC), New Delhi, there were 233,251 reported cases and 303 deaths in 2022, with 13,491 cases and 10 deaths in Rajasthan [3].

Dengue can be divided into two categories by the World Health Organization [4]: one with clinical presentation of DF from mild febrile illness to dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS) [5].

Leukocyte count, prothrombin time, activated partial thromboplastin time, HsCRP, alanine aminotransferase (ALT) levels, aspartate aminotransferase levels (AST), and haematocrit levels are a few clinical parameters and laboratory investigations that have been studied internationally to predict the severity of the infection [6,7]. Elevated liver enzymes, specifically aspartate aminotransferase (AST) and alanine aminotransferase (ALT), are linked to increased liver-related mortality and occur in 65–97% of dengue cases [8,9,10]. Higher AST and ALT levels are also associated with a greater risk of bleeding or dengue haemorrhagic fever (DHF). Some studies have explored the role of C-reactive protein (CRP), an acute-phase reactant and marker of infection and inflammation, as an early indicator of severe dengue illness in both adults and children [11,12].

Most dengue cases are asymptomatic and often resolve without intervention. However, improper management of complications can raise the mortality rate to over 20%. Recent studies have identified a subgroup of severe dengue cases with higher rates of comorbidities and fatalities compared to non-severe cases [13].

Despite the low mortality rate resulting from severe cases, dengue requires constant examination and round-the-clock nursing care due to the unpredictable progression of complications, posing a burden on clinical triage and material resources. Accordingly, identifying markers that allow for predicting disease prognosis from the initial diagnosis is needed. Given the improved pathogenesis understanding, myriad candidates have been proposed to be associated with severe dengue progression.

Objective: To estimate the Hs-CRP, Alanine Transaminase (ALT) and Aspartate Transaminase (AST) level in patients with dengue fever and determine the association of these levels with the severity of Dengue fever.

Material and methods:

This hospital based descriptive type of observational study was conducted at department of general medicine, JLN medical college

and hospital, Ajmer. Study was conducted between the period of November 2022 to May 2023 or till sample size completed.

After the permission brought from the Research review board and ethics committee of the college, **all confirmed dengue fever cases aged 18 to 70 years who were willing to participate and admitted to the medicine ward, were included. Exclusions were made for patients with a known history of liver disease, those who had used hepatotoxic drugs in the last six months, and patients with significant medical comorbidities such as tuberculosis, diabetes, coronary heart disease (CHD), or neoplasms.**

Sample size: Sample of 150 cases was calculated at 95% confidence interval and power of 80% to verify expected difference of 96.48 (with SD of 300 U/L) for ALT level in between patients with DF and DHF (as per reference article) [14]. The sample was further increased up to 160 cases as final sample size.

Sampling: Allocation concealment was ensured using opaque sealed envelope method for allocation. Allocation was done by a person not involved directly in the research to avoid selection bias.

After a clinical history and physical examination, patients underwent various laboratory investigations, including a complete blood count, liver function tests, serum creatinine, blood urea, serum albumin, serum glucose, and urine analysis (checking for urine albumin and haematuria). Additionally, an abdominal ultrasound examination and chest x-ray (PA view) were performed. Dengue infection was confirmed via the ELISA method.

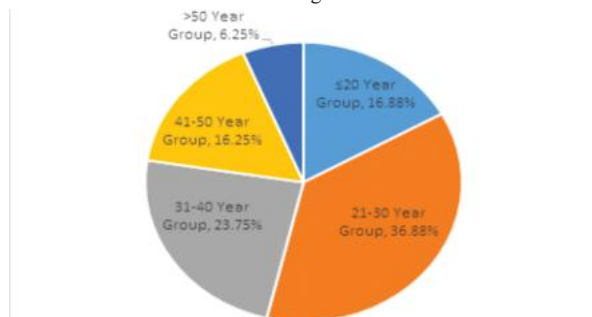
Under aseptic conditions, a 2 mL peripheral venous blood sample was collected from each dengue case in a plain vial within the first two days of hospital admission. Serum was separated by centrifugation at 3000 rpm and stored at 2-8°C. In this study, the presence of non-structural protein 1 (NS1) and IgM and IgG antibodies against the dengue virus were confirmed by ELISA.

Patients were divided into two groups (as per WHO classification, 2009): Group 1 comprised patients with non-severe classical dengue fever (DF), while Group 2 included those with severe dengue, including dengue haemorrhagic fever (DHF) and dengue shock syndrome (DSS). High-sensitivity C-reactive protein (hs-CRP) was used to assess severity, given its lower detection threshold compared to conventional CRP.

Statistical Analysis: Data was collected and stored in MS Office excel spread sheet, Statistical analyses were done using computer software Statistical Package for the Social Sciences (SPSS Trial Version 23 and Primer). The qualitative data were expressed in proportions and percentages, and the quantitative data (continuous data) were expressed as means and standard deviations. The difference in proportion was analysed using the chi-square test, and the difference in means among the groups was analysed using the student's t test. Significance level for tests were determined at 95% CI with $P < 0.05$.

Observations:

A total of 160 laboratory-confirmed dengue cases were observed. The proportion of male patients (68.75%) was significantly higher than that of female patients (31.25%). The residency distribution was nearly equal, with 51.88% from urban areas and 48.13% from rural areas. The mean age of the patients was 32.09 ± 12.08 years, and the age group distribution is illustrated in Table/ Figure 1.

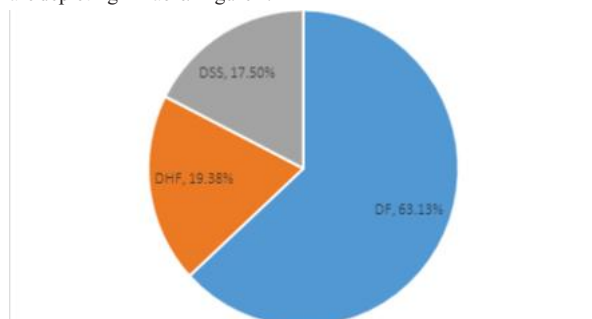


Table/ Figure 1.: The distribution patients as per age groups.

Regarding clinical features, all patients (100%) experienced fever. This was followed by headache (77.50%), rashes (44.38%), arthralgia (31.88%), retro-bulbar pain (30.63%), melena (26.25%), and myalgia (25%). Other symptoms, such as haematuria and epistaxis, were observed in only a few cases.

A significant portion of the patients (79.4%) did not exhibit organomegaly. Among those with organomegaly, hepatomegaly was the most common (81.82%), followed by splenomegaly (12.12%) and hepatosplenomegaly (6.06%). Pleural effusion was observed in 25% (40) of the cases, with more than half of these patients presenting mild pleural effusion.

NS1 positivity was observed in 85.63% of cases, while 19.38% (31) of patients showed isolated IgM positivity. Additionally, 4.38% (7) of patients tested positive for both IgM and IgG. Severity of dengue cases are depicting in Table/ Figure 2.



Table/ Figure 2. Distribution of patients as per severity of dengue cases

The severity of dengue was not found to be associated with the age of patients. However, platelet count significantly decreased as dengue severity increased ($p < 0.05$). Haematocrit, hs-CRP, ALT, and AST levels showed a significant increase with the severity of dengue, as illustrated in Table/ Figure 3.

Table/ Figure 3. Comparison of mean age, Platelet count, Haematocrit, Hs CRP, ALT and AST levels with the severity of the dengue.

Variables	Non severe	Severe	Level of significance
Age	30.8±11.808	34.31±12.315	0.07, (S)
Platelet count (PC)	57.07±30.749	30.56±24.126	<0.001, (S)
Hematocrit (HCT)	40.53±3.949	42.64±6.108	<0.009, (S)
hs-CRP	7±13.376	34.36±33.458	<0.001, (S)
ALT	86.75±65.63	161.62±112.938	<0.001, (S)
AST	124.15±63.322	220.3±123.121	<0.001, (S)

Among the 47 dengue cases requiring blood transfusions, Random Donor Platelets (RDP) were the most commonly (93.1%) administered product. The most frequent complication was serositis, including

pleural effusion (PE) and ascites, observed in 28.13% of cases. Other common complications included melena (26.25%), hypotension (15%), and bradycardia (13.75%). There were only two reported deaths. A comparison of complications according to dengue severity is shown in Table/ Figure 4.

Table/ Figure 4. Comparison of complication according to dengue severity.

	Non-severe (N=101)		Severe (N=59)		Total (N=160)		Level of significance
	No	%	No	%	No	%	
Serositis/PE/Ascites	7	6.93	38	64.41	45	28.13	<0.001S
Hepatitis	4	3.96	6	10.17	10	6.25	0.220NS
Melena	0	0.00	42	71.19	42	26.25	<0.001S
Hypotension	0	0.00	24	40.68	24	15.00	<0.001S
Bradycardia	3	2.97	19	32.20	22	13.75	<0.001S
Death	0	0.00	2	3.39	2	1.25	0.26NS

DISCUSSION:

In our study, the majority of patients (60.64%) were in the 21-40 age group, with a mean age of 32.09 ± 12.07 years. This age distribution aligns with findings from other studies, such as Mallhi TH et al. [15], which reported a mean age of 30.69 ± 16.13 years, and Agrawal VK et al. [16], reported a mean age of 30.3 ± 14.3 years. Similarly, Ayaz F et al [14]. Observed a mean age of $36.86 (\pm 13.9)$ years in their study.

In our study, male predominance was observed in 68.75% patients. Similar male predominance was observed in the study of Mallhi TH et al. [15], Agrawal VK et al. [16] and Ayaz F et al. [14] reported male predominance as 56.7%, 55.6% and 69.2%, respectively.

In this study, equal proportions of patients were from rural and urban areas, likely due to the catchment area of the healthcare facility serving both demographics. This distribution may reflect the widespread presence of *Aedes aegypti* mosquitoes in varied environments, making dengue fever equally prevalent in rural and urban settings.

This study observed that the fever, headache, rashes, retro bulbar pain, melena and arthralgia were the most common clinical feature. These findings were concordant with the study undertaken by Mallhi TH et al. [15], which found that typical manifestations of dengue-like fever, myalgia, arthralgia, headache, vomiting, abdominal pain, and skin rash were observed in more than 40% of patients. Kittittrakul C et al. [17] found that the common presenting symptoms and signs were myalgia (95.9%), nausea/vomiting (87.7%), positive tourniquet test (77.2%), abdominal pain (42.7%), hepatomegaly (34.6%), and bleeding (20.5%).

The Agrawal VK et al. [16] study established an independent association of rashes with severe dengue (OR: 1.99), which is similar to our study. while Sirivichayakul C et al. [18] showed no association. Dengue virus can incite the production of cytokines with stimulation of vascular endothelial changes, infiltration of mononuclear cells, and perivascular oedema, consequently leading to a skin rash [19].

In current study, hepatomegaly was the common organomegaly presentation while splenomegaly and hepato-splenomegaly were less common. The study of Dhanwada S et al [20] observed the association of hepatomegaly, with the severity of dengue also the study of Samanta J et al. [21] reported that organomegaly was common in dengue patients, with hepatomegaly being the most prevalent form.

In our study, out of 63.13% of cases were presented as non-severe dengue while DHF was present in 19.38% and DSS were in 17.50% of cases and the pleural effusion observed in 25 % cases. Which was increased with severity of dengue. NS1 positivity was observed in 85.63% of cases, while 19.38% (31) of patients showed isolated IgM positivity. Additionally, 4.38% (7) of patients tested positive for both IgM and IgG. Suggesting secondary dengue virus infection. **Agrawal VK et al.** [16] observed that the IgM positivity was seen in 60.7% cases and IgG positivity in 21.8% cases, while 8.9% had both IgM and IgG antibodies positivity and 140 (41.9%) were NS1 antigen test positivity. Thrombocytopenia was more common among the severe dengue patients, and this finding is consistent with the previous reports (Boo et al.) [22]. This is predominantly due to the increased destruction of platelets as a result of the complement activation, peripheral sequestration, and marrow dysfunction, and, of late, oxidative stress

(Soundravally et al) [23]. The platelets are also shown to adhere to the endothelial cells due to stimulation by the dengue virus (Krishnamurti et al.) [24]. Thrombocytopenia is a classical of dengue infection, and this can be caused due to binding of platelets to the activated endothelial cells (Teixeira et al.) [25].

Our study found a positive association between increased hs-CRP, AST, and ALT levels and dengue severity. Elevated hs-CRP levels, consistent with the findings of Atukuri SR et al. [26], were significantly higher in severe dengue cases. Chen et al. reported increasing CRP levels with severity in adult dengue patients, showing mean CRP values of 8.5 in DF, 15.2 in DHF I, and 124.5 in DHF III, which was statistically significant ($p < 0.0001$). Elevated CRP levels suggest more severe tissue damage from increased inflammation and often correlate with poorer outcomes, highlighting the importance of closely monitoring and promptly treating dengue patients with high CRP levels.

Additionally, Kittittrakul C et al [17]. Observed that elevated AST levels in febrile phase correlated with bleeding, with high AST and ALT levels noted in 11.0% and 7.0% of cases, respectively. Furthermore, high ALT levels during the febrile stage were associated with shock, supporting the link between elevated liver enzymes and more severe complications in dengue patients.

Zubair A et al [27]. found that AST and ALT levels were significantly elevated in patients with poorer outcomes, aiding in the early identification of high-risk cases. Similarly, in our study, cases with unfavorable outcomes exhibited significantly elevated transaminase levels, aligning with findings from other research. Rafi A et al. [28] reported that raised serum ALT or AST was present in 14.1% of cases, while elevated serum creatinine occurred in 6.6%. Ayaz F et al [14] suggested that higher ALT levels at presentation might signal a greater risk of progression to severe form of. Consequently, patients with elevated ALT should be prioritized for close monitoring.

CONCLUSION

Dengue tends to affect the young adult male (21-40 year). Hepatomegaly is common in dengue patients and the hs-CRP levels associated with the severity of dengue, suggesting a heightened inflammatory response. Higher serum levels of ALT and AST at presentation in dengue may indicate liver damage, can give a clue about possibility of progression to severe forms of DF (DHF and DSS) or may be at higher risk of developing complications such as severe dengue or multi-organ dysfunction. Severe dengue is strongly associated with complications such as ascites, melena, hypotension, and bradycardia, all of which were much more common in severe cases than in non-severe cases. Thus, these patients prioritized and monitored more rigorously.

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