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Original Research Article

NAVIGATING THE HAEMATOLOGICAL MAZE: UNRAVELING THE ROLE OF NLRA AND PLR AS PREDICTORS OF DENGUE SEVERITY - A CROSS-SECTIONAL STUDY

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ABSTRACT

Background: Dengue is a major mosquito-borne viral illness with clinical manifestations ranging from mild febrile illness to severe dengue associated with plasma leakage, hemorrhage, and organ dysfunction. Early prediction of disease severity remains essential for improving patient outcomes. Hematological indices such as neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) have emerged as potential biomarkers of systemic inflammation and disease progression.

Materials and Methods: This hospital-based cross-sectional study was conducted among 173 laboratory-confirmed dengue patients admitted to a tertiary care center between May 2024 and April 2026. Patients were categorized according to World Health Organization into non-severe and severe dengue groups. Clinical, hematological, and biochemical parameters were analyzed. NLR and PLR were calculated from complete blood counts. Receiver operating characteristic (ROC) curve analysis was performed to assess their predictive utility for severe dengue.

Results: Of the 173 patients, 52 (30.1%) had severe dengue. Severe dengue patients were significantly older and had longer symptom duration. Diabetes mellitus, vomiting, abdominal pain, bleeding manifestations, and hepatomegaly were significantly associated with severe dengue. Severe cases demonstrated significantly higher hematocrit, leukocyte count, AST, ALT, creatinine, and NLR, along with lower platelet count, albumin, and PLR (all $p<0.05$). NLR showed good predictive performance for severe dengue with an AUC of 0.84 (95% CI: 0.77–0.90), sensitivity of 82.7%, and specificity of 74.4% at a cut-off >2.7. PLR demonstrated moderate predictive utility with an AUC of 0.78 (95% CI: 0.71–0.85).

Conclusion: NLR and PLR are simple, cost-effective, and readily available biomarkers associated with dengue severity. Among them, NLR demonstrated superior predictive accuracy and may serve as a useful early indicator of severe dengue in routine clinical practice.

Keywords: Dengue; Neutrophil-to-lymphocyte ratio; Platelet-to-lymphocyte ratio; Severe dengue; Biomarkers.

INTRODUCTION

Dengue fever continues to be a major vector-borne viral disease affecting millions of individuals across tropical and subtropical countries. The infection is caused by the dengue virus (DENV), an RNA virus of the Flaviviridae family, and is primarily transmitted through the bite of infected Aedes aegypti mosquitoes.[1] In recent years, the incidence of dengue has increased dramatically due to rapid urban expansion, climate variability, inadequate sanitation, increased human mobility, and ineffective mosquito control strategies, particularly in densely populated developing nations such as India.[1] The World Health Organization estimates that approximately 390 million dengue infections occur annually worldwide, with nearly 96 million cases becoming clinically apparent.[2] India has witnessed repeated dengue

outbreaks with rising hospitalization rates and increasing occurrence of severe disease, placing substantial strain on healthcare infrastructure.[1]

The disease exhibits a broad clinical spectrum ranging from uncomplicated febrile illness to severe dengue associated with plasma leakage, hemorrhagic manifestations, shock, and organ impairment.[3] Although most patients recover with supportive care, severe dengue can rapidly progress to life-threatening complications if not recognized early. Timely identification of patients at risk for severe disease therefore remains a cornerstone in dengue management.[3] Current clinical warning signs recommended by the World Health Organization, including persistent vomiting, abdominal pain, mucosal bleeding, and fluid accumulation, are useful for monitoring progression; however, these manifestations often develop later during the critical phase of illness, limiting their role in early prognostication.[4]

Dengue infection produces characteristic hematological and immunological disturbances including thrombocytopenia, leukopenia, hemoconcentration, and exaggerated inflammatory responses.[5] Consequently, increasing interest has been directed toward inflammatory markers derived from routine hematological investigations as potential predictors of disease severity. Among these, neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) have emerged as promising biomarkers in various infectious and inflammatory conditions such as sepsis, COVID-19, and malaria.[6] These ratios are inexpensive, rapidly obtainable, and capable of reflecting the balance between systemic inflammation, immune regulation, and platelet consumption.

The biological rationale behind the use of NLR and PLR in dengue is supported by the underlying disease pathogenesis. Elevated neutrophil counts may indicate enhanced cytokine-driven inflammatory activity, while lymphopenia can occur due to apoptosis and redistribution of lymphocytes during acute viral infection.[7] Likewise, progressive thrombocytopenia resulting from bone marrow suppression, immune-mediated destruction, and peripheral platelet consumption contributes to altered PLR values. Therefore, increased NLR and decreased PLR may correlate with worsening inflammation, vascular permeability, and severe clinical outcomes in dengue infection.[7]

In view of the increasing dengue burden and the need for simple prognostic tools, the present study was undertaken to evaluate the association of NLR and PLR with dengue severity and to determine their usefulness as accessible biomarkers for early risk stratification in routine clinical practice.

MATERIALS AND METHODS

Study design and study setting: A hospital-based analytical cross-sectional study was carried out in the Department of Pathology in association with the Department of General Medicine at a tertiary care teaching institution over a duration of two years, from May 2024 to April 2026. Written informed consent was obtained from each participant before enrollment, and confidentiality of patient-related information was strictly maintained throughout the study period.

Study participants and eligibility criteria: Adult patients aged 18 years and above presenting with clinical suspicion of dengue fever and subsequently confirmed by laboratory investigations were considered eligible for inclusion. Confirmation of dengue infection was performed using NS1 antigen detection and/or dengue IgM antibody testing by ELISA method. Patients with evidence of other concurrent infectious illnesses such as malaria, leptospirosis, typhoid fever, or bacterial sepsis were excluded. Individuals with hematological malignancies, chronic inflammatory or autoimmune disorders, pre-existing blood dyscrasias, or those receiving steroid or immunosuppressive therapy were also excluded. Cases with incomplete laboratory or clinical records were omitted from the final analysis to ensure data accuracy.

Sample size determination and sampling method: Sample size estimation was based on findings from a previously published cross-sectional study by Nandhini et al., which reported a significant association between NLR values and dengue severity.[8] Using the formula for estimation of proportions in cross-sectional studies: $n=Z2\times p\times(1-p)/d2$; assuming a 95% confidence level ($Z = 1.96$), expected proportion (p) derived from prior study (~10%), and an absolute precision (d) of 5%, minimum sample size calculated was 173 participants.[8] Eligible patients fulfilling the inclusion criteria were enrolled consecutively throughout the study period until the required sample size was attained.

Clinical evaluation and data collection: After obtaining informed written consent, detailed demographic and clinical information including duration of fever, presenting symptoms, comorbid conditions, and warning signs were documented in a structured case record form. Thorough clinical examination was performed at the time of admission. Patients were additionally categorized into febrile, critical, or recovery phases according to standard dengue clinical staging to account for temporal variations in hematological parameters.

Classification of dengue severity: Severity assessment was performed according to the World Health Organization. Patients were grouped into non-severe dengue (with or without warning signs) and severe dengue. Severe dengue was diagnosed in the presence of severe plasma leakage causing shock or respiratory compromise, severe hemorrhagic manifestations, or significant organ dysfunction including hepatic, neurological, or cardiac involvement.

Laboratory investigations and calculation of hematological indices: Blood samples were collected within 24 hours of hospital admission before initiation of definitive treatment. Complete blood count parameters including hemoglobin, hematocrit, total leukocyte count, absolute neutrophil count, absolute lymphocyte count, and platelet count were measured using an automated hematology analyzer under standardized laboratory conditions. Biochemical investigations such as serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), serum creatinine, and serum albumin were also evaluated. NLR was derived by dividing the absolute neutrophil count by the absolute lymphocyte count, whereas PLR was calculated by dividing platelet count by absolute lymphocyte count.

Outcome variables: The primary outcome of the study was the severity of dengue infection. The relationship of NLR and PLR with dengue severity was assessed. Secondary objectives included evaluation of their association with warning signs, biochemical abnormalities, and their diagnostic performance in predicting severe dengue.

Statistical analysis: All collected data were entered into Microsoft Excel and statistically analyzed using SPSS software version 25.0 (IBM Corp., Armonk, NY). Continuous variables were summarized as mean ± standard deviation (SD), while categorical variables were expressed as numbers and percentages. Differences between non-severe and severe dengue groups were analyzed using the independent t-test for continuous variables and chi-square test or Fisher's exact test for categorical variables, as appropriate. Receiver operating characteristic (ROC) curve analysis was performed to evaluate the diagnostic performance of neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) in predicting severe dengue. Area under the curve (AUC), sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were determined, and optimal cut-off values were obtained using the Youden index. A p-value of <0.05 was considered statistically significant.

RESULTS

The study included 173 confirmed dengue patients, of whom 121 (69.9%) had non-severe dengue and 52 (30.1%) had severe dengue. Patients with severe dengue were significantly older compared to those with non-severe disease (39.9 ± 15.0 vs 33.8 ± 13.2 years; p = 0.032). The mean duration of symptoms was also significantly longer in severe dengue (5.6 ± 1.9 vs 4.4 ± 1.5 days; p = 0.008). Male predominance was observed in both groups without significant association (p = 0.364). Among comorbidities, type 2 diabetes mellitus was significantly more common in severe dengue (32.7% vs 14.9%; p = 0.011). Clinically, vomiting, abdominal pain, bleeding manifestations, and hepatomegaly were significantly more frequent in severe dengue patients (all p ≤ 0.004), indicating greater systemic involvement in severe disease [Table 1].

Table 1: Baseline Demographic Characteristics, Comorbidities, and Clinical Manifestations Among Dengue Patients (n = 173).

Characteristics	Total Cases (n=173)	Non-severe Dengue (n=121)	Severe Dengue (n=52)	p-value
Age (years)	35.6 ± 14.1	33.8 ± 13.2	39.9 ± 15.0	0.032
Male gender	111 (64.2%)	75 (62.0%)	36 (69.2%)	0.364
BMI (kg/m²)	24.1 ± 3.9	23.7 ± 3.7	24.9 ± 4.2	0.086
Duration of symptoms (days)	4.8 ± 1.8	4.4 ± 1.5	5.6 ± 1.9	0.008
Associated comorbidities				
Type 2 diabetes mellitus	35 (20.2%)	18 (14.9%)	17 (32.7%)	0.011
Systemic hypertension	38 (22.0%)	23 (19.0%)	15 (28.8%)	0.149
Chronic hepatic disease	11 (6.4%)	5 (4.1%)	6 (11.5%)	0.067
Presenting clinical manifestations				
Vomiting episodes	73 (42.2%)	42 (34.7%)	31 (59.6%)	0.004
Abdominal discomfort/pain	65 (37.6%)	33 (27.3%)	32 (61.5%)	<0.001
Bleeding manifestations	37 (21.4%)	15 (12.4%)	22 (42.3%)	<0.001
Enlarged liver (hepatomegaly)	51 (29.5%)	25 (20.7%)	26 (50.0%)	<0.001

BMI: body mass index.

WHO-defined warning signs were significantly more prevalent among severe dengue patients. Persistent vomiting was observed in 59.6% of severe cases compared to 34.7% of non-severe cases (p = 0.004). Evidence of fluid accumulation (42.3% vs 9.9%), lethargy/restlessness (46.2% vs 14.9%), and hemoconcentration with thrombocytopenia (65.4% vs 19.8%) were also markedly higher in severe dengue (all p <0.001). Features exclusively seen in severe dengue included dengue shock syndrome (48.1%), major bleeding episodes (38.5%), and significant organ dysfunction (42.3%), confirming appropriate clinical stratification according to WHO severity criteria [Table 2].

Table 2: Distribution of WHO Warning Signs and Severe Dengue Features Among Study Participants.

Clinical Parameter	Non-severe Dengue (n=121)	Severe Dengue (n=52)	p-value
Persistent vomiting	42 (34.7%)	31 (59.6%)	0.004
Evidence of fluid accumulation	12 (9.9%)	22 (42.3%)	<0.001
Lethargy or restlessness	18 (14.9%)	24 (46.2%)	<0.001

Hemoconcentration with thrombocytopenia	24 (19.8%)	34 (65.4%)	<0.001
Dengue shock syndrome	—	25 (48.1%)	—
Major bleeding episodes	—	20 (38.5%)	—
Significant organ dysfunction	—	22 (42.3%)	—

Classification based on World Health Organization.

Severe dengue patients demonstrated significantly higher hemoglobin levels (13.9 ± 1.8 vs 13.1 ± 1.4 g/dL; p = 0.021), packed cell volume (43.8 ± 5.3 vs 39.2 ± 4.4%; p <0.001), and total leukocyte count (4860 ± 1380 vs 4180 ± 1160/mm³; p = 0.009). Severe thrombocytopenia was observed in severe dengue, with markedly lower platelet counts compared to non-severe cases (58 ± 22 vs 116 ± 41 ×10³/µL; p <0.001). Biochemical abnormalities including elevated AST, ALT, and serum creatinine along with reduced serum albumin were significantly associated with severe dengue (all p <0.001). Furthermore, NLR was significantly increased in severe dengue (3.84 ± 1.52 vs 2.01 ± 0.92; p <0.001), whereas PLR was significantly reduced (91.2 ± 38.4 vs 136.8 ± 57.1; p <0.001) [Table 3].

Table 3: Comparison of Hematological and Biochemical Parameters Between Non-severe and Severe Dengue Groups.

Laboratory Parameter	Non-severe Dengue (n=121)	Severe Dengue (n=52)	p-value
Hemoglobin (g/dL)	13.1 ± 1.4	13.9 ± 1.8	0.021
Packed cell volume (%)	39.2 ± 4.4	43.8 ± 5.3	<0.001
Total leukocyte count (/mm³)	4180 ± 1160	4860 ± 1380	0.009
Platelet count (×10³/µL)	116 ± 41	58 ± 22	<0.001
Serum AST (IU/L)	86 ± 39	176 ± 78	<0.001
Serum ALT (IU/L)	68 ± 31	149 ± 66	<0.001
Serum creatinine (mg/dL)	0.94 ± 0.24	1.34 ± 0.51	<0.001
Serum albumin (g/dL)	3.7 ± 0.5	3.0 ± 0.6	<0.001
Neutrophil-to-lymphocyte ratio (NLR)	2.01 ± 0.92	3.84 ± 1.52	<0.001
Platelet-to-lymphocyte ratio (PLR)	136.8 ± 57.1	91.2 ± 38.4	<0.001

AST: aspartate aminotransferase; ALT: alanine aminotransferase; NLR: neutrophil-to-lymphocyte ratio; PLR: platelet-to-lymphocyte ratio.

Receiver operating characteristic (ROC) analysis demonstrated good predictive accuracy of NLR for severe dengue with an AUC of 0.84 (95% CI: 0.77–0.90; p <0.001). An optimal NLR cut-off value of >2.7 yielded 82.7% sensitivity and 74.4% specificity, with a negative predictive value (NPV) of 88.7%. PLR also showed moderate predictive utility with an AUC of 0.78 (95% CI: 0.71–0.85; p <0.001). A PLR cut-off value of <108 demonstrated 76.9% sensitivity and 69.4% specificity. Overall, NLR exhibited superior discriminatory performance compared to PLR for predicting severe dengue [Table 4].

Table 4: Diagnostic Performance of Neutrophil-to-Lymphocyte Ratio and Platelet-to-Lymphocyte Ratio in Predicting Severe Dengue.

Diagnostic Indicator	NLR	PLR
Area under ROC curve (95% CI)	0.84 (0.77–0.90)	0.78 (0.71–0.85)
Optimal threshold	>2.7	<108
Sensitivity (%)	82.7	76.9
Specificity (%)	74.4	69.4
Positive predictive value (%)	64.2	58.8
Negative predictive value (%)	88.7	84.6
Statistical significance	<0.001	<0.001

ROC: receiver operating characteristic; AUC: area under the curve; CI: confidence interval; NLR: neutrophil-to-lymphocyte ratio; PLR: platelet-to-lymphocyte ratio.

DISCUSSION

The present study evaluated the role of neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) as predictors of dengue severity among 173 laboratory-confirmed dengue patients and demonstrated that both indices are significantly associated with severe dengue. Severe dengue accounted for 30.1% of cases, which is comparable to observations from Indian tertiary care studies in Dinkar et al., and Vikhe et al., where severe disease prevalence has ranged between 20–35%, particularly in referral centers managing complicated cases.[9,10]

The significantly higher age among severe dengue patients in our study (39.9 ± 15.0 vs 33.8 ± 13.2 years; p=0.032) suggests that advancing age may predispose individuals to poorer clinical outcomes due to altered immune response, endothelial dysfunction, and greater prevalence of comorbid conditions. Similar findings have been reported in studies from India, Thailand, and Brazil, by Vikhe et al., Talukdar et al., and Cerqueira-Silva et al., where older adults exhibited increased risk of plasma leakage and organ dysfunction.[10-12]

Among associated comorbidities, diabetes mellitus showed a significant association with severe dengue (32.7% vs 14.9%; p=0.011). This observation is supported by previous literature indicating that diabetes may aggravate dengue severity through chronic endothelial injury, exaggerated inflammatory

cytokine release, and impaired innate immunity.[13] Although hypertension and chronic liver disease were more frequent among severe cases, statistical significance was not achieved, possibly due to limited subgroup size.[13]

Clinically, vomiting, abdominal pain, mucosal bleeding, and hepatomegaly were significantly more prevalent in severe dengue, consistent with the warning signs described in the World Health Organization.[14] Furthermore, severe dengue patients demonstrated markedly higher frequencies of fluid accumulation, lethargy, and hemoconcentration with thrombocytopenia (all $p<0.001$), reflecting increased capillary permeability and plasma leakage, which represent key pathogenic mechanisms in severe dengue. The exclusive occurrence of dengue shock syndrome, major bleeding, and organ dysfunction among severe cases validates the reliability of the WHO severity classification applied in this study.[14]

The hematological findings of the present study were characteristic of progressive dengue severity. Severe dengue patients demonstrated significantly elevated hemoglobin and packed cell volume levels, indicating hemoconcentration secondary to plasma leakage. Similar findings have been consistently reported in studies by Sharma et al. and Sadgir et al., where rising hematocrit correlated strongly with vascular permeability and disease progression.[15,16] Profound thrombocytopenia observed in severe dengue ($58 \pm 22 \times 10^3/\mu L$ vs $116 \pm 41 \times 10^3/\mu L$; $p<0.001$) can be explained by a combination of bone marrow suppression, peripheral platelet destruction, immune-mediated clearance, and platelet consumption within damaged endothelium.[15] Elevated AST and ALT levels among severe dengue patients further suggest hepatic involvement due to direct viral injury, hypoxic damage, and cytokine-mediated hepatocellular inflammation.[16] Concurrent hypoalbuminemia and elevated creatinine levels additionally indicate capillary leak syndrome and early renal impairment.[16]

A major finding of this study was the significantly elevated NLR among severe dengue patients (3.84 ± 1.52 vs 2.01 ± 0.92; $p<0.001$). This finding is biologically plausible because severe dengue is characterized by intense systemic inflammation, neutrophil activation, cytokine storm, and lymphocyte depletion.[17] Neutrophilia may reflect heightened innate immune activation and endothelial inflammation, while lymphopenia is thought to occur due to apoptosis and redistribution of lymphocytes during acute viral infection.[17] Similar elevations in NLR among severe dengue patients have been documented in studies by Kareliya et al., Sami et al., and Srisuphanunt et al., from India, Bangladesh, and Thailand where NLR emerged as a reliable marker of disease progression.[18-20] The ROC analysis in our study demonstrated good predictive performance of NLR with an AUC of 0.84 and an optimal cut-off value >2.7, yielding 82.7% sensitivity and 74.4% specificity. These values are comparable to previously published studies by Kareliya et al., and Sami et al., reporting AUCs ranging from 0.75–0.87 for NLR in severe dengue prediction.[18,19]

PLR was significantly reduced in severe dengue patients (91.2 ± 38.4 vs 136.8 ± 57.1; $p<0.001$), highlighting the impact of severe thrombocytopenia relative to lymphocyte count. Although PLR demonstrated slightly lower predictive accuracy compared to NLR (AUC 0.78), it still showed acceptable sensitivity and specificity. Similar reductions in PLR have been reported in dengue cohorts from studies by Agrawal et al., and Ashma from Southeast Asia, suggesting that PLR may serve as an adjunctive marker of severity.[21,22] However, the superior performance of NLR observed in our study suggests that inflammatory imbalance may have greater prognostic significance than platelet reduction alone.

CONCLUSION

The present study demonstrates that neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) are valuable hematological markers for predicting dengue severity. Severe dengue was significantly associated with elevated NLR, reduced PLR, hemoconcentration, thrombocytopenia, and deranged biochemical parameters. Among the two indices, NLR showed superior diagnostic performance with good sensitivity and specificity for identifying severe dengue. These findings highlight the role of inflammatory and immune dysregulation in dengue pathogenesis. As NLR and PLR are inexpensive, easily obtainable, and derived from routine blood investigations, they may serve as practical tools for early risk stratification and timely clinical decision-making, particularly in resource-constrained settings.

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