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Correlation between Neutrophil Lymphocyte Ratio (NLR) and the Severity of Dengue Hemorrhagic Fever at Candi Umbul Hospital, Magelang

Christine Notoningtiyas Santoso
General Practioner, Candi Umbul Hospital Magelang, Indonesia
Email: christine.notoningtiyas@gmail.com

KEYWORDS

Dengue hemorrhagic fever, neutrophil-to-lymphocyte ratio, platelet count, disease severity, hematological markers

ABSTRACT

Dengue hemorrhagic fever (DHF) remains a significant public health concern, especially in tropical and subtropical regions, where early identification of severe cases is crucial to prevent fatal complications. This study investigated the relationship between hematological parameters, namely neutrophil-to-lymphocyte ratio (NLR) and platelet count, and the severity of DHF among adult patients. A retrospective observational study was conducted at Candi Umbul Hospital, Magelang, from June 2024 to April 2025, involving 184 confirmed DHF patients classified into severity Grades I to III. NLR and platelet counts were calculated from complete blood counts taken upon admission. Spearman's rank correlation test assessed associations between these parameters and disease severity. Results showed a significant negative correlation between NLR and DHF severity ($\rho = -0.356$, $p < 0.001$), indicating lower NLR values in more severe cases. Similarly, platelet count negatively correlated with DHF severity ($\rho = -0.400$, $p < 0.001$), reflecting progressive thrombocytopenia with increased severity. Notably, no significant correlation was found between NLR and platelet count ($\rho = 0.004$, $p = 0.959$), suggesting that these parameters independently reflect different aspects of DHF pathophysiology. The findings highlight that combined NLR and platelet count can improve early detection of high-risk DHF patients by providing complementary insights into the disease's immunopathology. This underscores the importance of incorporating routine hematological markers in dengue severity assessments to guide timely clinical decisions.

DOI:

Corresponding Author: Christine Notoningtiyas Santoso*
Email: christine.notoningtiyas@gmail.com

INTRODUCTION

Dengue Hemorrhagic Fever (DHF) remains a significant public health challenge in tropical and subtropical regions, with its incidence steadily rising and placing considerable burdens on healthcare systems worldwide (Knerer, 2021; Santosa, 2020; Tsheten et al., 2021). The disease, caused by infection with one of the four dengue virus serotypes, manifests with a wide spectrum of clinical presentations, ranging from mild febrile illness to severe, life-threatening hemorrhagic and shock syndromes (Nasir et al., 2024). Early identification of patients at risk for severe disease is critical for effective management and reduction of morbidity and mortality, yet distinguishing severe cases based solely on clinical features remains challenging due to the nonspecific nature of initial symptoms (Bhati MR Meena H, 2024; Navya P Begum R, 2024; Sharma SK Kularia R, 2024).

Recent advances in laboratory medicine have highlighted the potential utility of hematological indices as accessible and cost-effective prognostic markers in dengue infection (ur Rahman et al., 2024). Among these, the neutrophil-lymphocyte ratio (NLR) has emerged as a promising indicator of systemic

inflammation and immune response dynamics in various infectious diseases, including dengue. NLR is calculated from routine complete blood count (CBC) parameters, making it readily available in most clinical settings. Several studies have demonstrated that changes in NLR values correlate with the severity of dengue infection, with lower NLR levels often associated with more severe clinical manifestations, such as DHF and dengue shock syndrome (DSS). This inverse relationship reflects the profound immune dysregulation and bone marrow suppression during the critical phase of dengue, leading to neutropenia and relative lymphocytosis (Bhati MR Meena H, 2024; Sharma SK Kularia R, 2024).

In parallel, thrombocytopenia, or a marked reduction in platelet count, is a well-recognized hallmark of DHF and serves as a key criterion in the World Health Organization's case classification for severe dengue. Platelet destruction and consumption are believed to result from immune-mediated mechanisms and direct viral effects on megakaryocytes, contributing to the risk of hemorrhagic complications. The interplay between NLR and platelet levels has garnered increasing research interest, as both parameters may provide complementary insights into disease progression and severity. Studies have reported that while platelet counts typically decrease during the acute and critical phases of DHF, NLR values may fluctuate in a manner that reflects the underlying immunopathology and the risk of severe outcomes (Das S Abreu C, 2022a; Singh A Bisht P, 2020).

Recent investigations have further explored the combined prognostic value of NLR and platelet counts in predicting the severity of dengue infection (Qian et al., 2023). For instance, research conducted in pediatric and adult populations has shown that higher NLR and lower platelet counts are significantly associated with severe dengue presentations, including DHF and DSS. These findings suggest that simultaneous assessment of NLR and platelet values could enhance early risk stratification and guide timely clinical interventions. Moreover, the use of such readily available hematological markers is particularly advantageous in resource-limited settings, where advanced diagnostic modalities may not be accessible (Maulida YR Amelia R, 2023; A. K. Prajapati, 2024).

Despite these promising observations, the correlation between NLR, platelet values, and the clinical severity of DHF remains an area of ongoing investigation, with some studies reporting variability in the predictive thresholds and temporal dynamics of these markers. Therefore, further research is warranted to elucidate the relationship between NLR and platelet counts across different populations and healthcare settings, and to establish standardized cut-off values for clinical application (Prijanto SA Suryawan IW, 2023).

Studies have examined the prognostic utility of hematological markers in dengue severity. A study by Chen et al. (2019) demonstrated that elevated NLR levels correlated with increased inflammatory responses and disease severity in dengue patients, suggesting NLR as a potential marker for early risk stratification. Similarly, Lee et al. (2021) found that platelet count was a reliable predictor of hemorrhagic complications in DHF, emphasizing the importance of thrombocytopenia monitoring during the acute phase. However, these studies independently assessed NLR and platelet count, with limited investigation into their combined prognostic value across adult populations in resource-constrained settings. The novelty of this research lies in its simultaneous evaluation of NLR and platelet counts in predicting DHF severity, emphasizing their complementary roles in reflecting the immunopathological and hematological changes during infection. This study further contributes by providing data from a tropical hospital setting, aiding the development of practical and cost-effective tools for the early identification of severe dengue cases (Buonacera A Stancanelli B, 2022).

This study aims to investigate the correlation between neutrophil-lymphocyte ratio (NLR) and platelet value with the severity of dengue hemorrhagic fever among patients admitted to Candi Umbul

Hospital Magelang, thereby contributing to the growing body of evidence supporting the use of simple hematological indices in the prognostic assessment of dengue infection (A. Prajapati, 2024). The findings of this study are expected to support clinicians in resource-limited settings by providing accessible, rapid, and cost-effective hematological indicators to stratify dengue severity. Early identification of high-risk patients can improve clinical decision-making, optimize resource allocation, and ultimately reduce morbidity and mortality associated with dengue hemorrhagic fever.

METHOD

Study Design

This retrospective observational cohort study evaluates the correlation between neutrophil-to-lymphocyte ratio (NLR), platelet count, and Dengue Hemorrhagic Fever (DHF) clinical severity. The study will be conducted at Candi Umbul Hospital Magelang, a secondary referral center located in a dengue-endemic region, over a defined period from June 2024 to April 2025. Ethical approval will be obtained from the institutional review board (IRB) of Candi Umbul Hospital, Magelang. Given the retrospective and non-interventional nature of the study, the requirement for individual informed consent will be waived in accordance with institutional and national guidelines.

Patient Cohort Selection

The study population will consist of adult patients hospitalized at Candi Umbul Hospital, Magelang, between June 2024 and April 2025 with a confirmed diagnosis of DHF. Diagnosis will be based on the 2011 World Health Organization (WHO) criteria, including clinical features such as hemorrhagic manifestations and plasma leakage, supported by positive NS1 antigen and/or dengue-specific IgM/IgG serology using rapid immunochromatographic tests.

Eligible patients must have complete blood count (CBC) results recorded at admission, including absolute neutrophil and lymphocyte counts, platelet count, and hematocrit. The NLR will be calculated as the absolute neutrophil to lymphocyte count ratio, and thrombocytopenia will be defined as a platelet count $<100 \times 10^3/\mu L$. Patients will be excluded if they have co-infections (e.g., bacterial sepsis), autoimmune diseases, hematologic malignancies, or incomplete medical records.

The minimum required sample size was calculated using Fisher's Z transformation method for correlation analysis, assuming a two-tailed significance level of 0.05, power of 80%, and an expected correlation coefficient of 0.3. Based on this approach, a minimum of 85 patients will be required to detect a statistically significant association.

Data Collection

Data will be collected retrospectively from anonymized medical records of patients meeting the inclusion criteria. Patients will be identified through the hospital's inpatient registry and laboratory information system. Trained personnel will extract demographic variables (age, sex), clinical data (DHF severity grade), and hematological parameters recorded at admission.

Laboratory parameters will include total leukocyte count, absolute neutrophil count, absolute lymphocyte count, platelet count, and hematocrit percentage. NLR will be calculated by dividing the absolute neutrophil count ($\times 10^3/\mu L$) by the absolute lymphocyte count

(×10³/µL). Platelet count will be analyzed as a continuous and categorical variable based on WHO-defined thresholds for thrombocytopenia.

DHF severity will be classified into four grades (I–IV) using WHO criteria, considering bleeding, plasma leakage, and shock. Only the first recorded laboratory results during hospital admission will be used to minimize bias from disease progression or treatment effects. Data integrity checks and periodic validation will be conducted to ensure reliability. All data will be de-identified before analysis to protect patient confidentiality.

Operational and Variable Definition

This study involves several key variables to investigate the association between laboratory parameters and the clinical severity of DHF. The primary independent variables are the NLR and platelet count derived from routine complete blood test (CBC) testing. The dependent variable is the DHF severity grade. DHF is an acute viral illness caused by the dengue virus and transmitted by the Aedes aegypti mosquitoes. This study uses rapid tests to diagnose DHF based on clinical symptoms and confirmation through Dengue NS1 antigen and/or IgM/IgG serology. DHF severity is classified into three grades: Grade I, Grade II, and Grade III. DHF Grade I patients experience fever, nonspecific infection symptoms, a positive tourniquet test, and thrombocytopenia. Meanwhile, spontaneous skin or mucosal bleeding is found in Grade II, in addition to the Grade I symptoms. In Grade III DHF, there are signs of circulatory failure such as hypotension, cold extremities, and rapid pulse. For this study, patients were grouped based on available clinical data into the appropriate DHF grade to assess the correlation between NLR, thrombocytopenia, and disease severity. NLR is calculated by dividing the absolute neutrophil count (×10³/µL) by the absolute lymphocyte count (×10³/µL) obtained from a complete blood count (CBC). NLR serves as a biomarker for systemic inflammation. A value of 1–3 is considered normal, >3 indicates elevated inflammation, and <1 may indicate immune suppression. Platelet count is measured as part of a CBC and is expressed in ×10³/µL. Thrombocytopenia is defined as a platelet count <150,000/µL, and in the context of DHF, a value <100,000/µL is considered a diagnostic criterion according to WHO guidelines.

Laboratory Measurements and Procedures

Dengue infection was confirmed through serological testing using the NS1 antigen and/or dengue-specific IgM/IgG antibodies using rapid immunochromatographic diagnostic kits from FastclearQ. Peripheral venous blood samples were collected using a standardized phlebotomy procedure and transferred into tubes containing K3EDTA as an anticoagulant. Subsequently, these samples were processed for complete blood count (CBC) analysis using an automated hematology analyzer available at Candi Umbul Hospital, Magelang clinical pathology laboratory, to identify a differential enumeration of blood cell populations. Key hematological parameters extracted for this study included the absolute neutrophil count, absolute lymphocyte count, platelet count, and hematocrit level. The NLR was calculated manually by dividing the absolute neutrophil count by the absolute lymphocyte count. To ensure the validity of the hematological data, routine quality control methods were followed in accordance with the laboratory's standard.

Statistical Analysis Methods

All statistical analyses were performed using the IBM SPSS Statistics Ver 25. Firstly, using the Kolmogorov-Smirnov and Shapiro-Wilk Tests, a normality test was conducted on the continuous variables (NLR and platelet count). Both variables were not normally distributed. Thus, non-parametric methods were conducted for the subsequent analysis. Descriptive statistics were used to summarize the baseline characteristics of the participants, including sex, age group, DHF severity, average platelet count, and NLR. Categorical data were presented as frequencies and percentages. Spearman's rank correlation test assessed the relationship between NLR value and DHF severity grade, platelet count and DHF severity grade, and the correlation between NLR value and platelet count. A significance level of $p < 0.05$ was considered statistically significant.

RESULT AND DISCUSSION

Demographic and Baseline Clinical Characteristics of the Study Cohort

Table 1. Baseline characteristics of patients

Patient Characteristics	Value
Sample size (n)	184
Sex, n (%)	
Male	73 (39.7)
Female	111 (60.3)
Age (Mean ± SD)	30.55 ± 15.27
Age group, n (%)	
<18 years old	33 (17.9)
18–25 years old	46 (25.0)
26–45 years old	67 (36.4)
46–65 years old	32 (17.4)
>65 years old	6 (3.3)
DHF Severity, n (%)	
DHF 1	101 (54.9)
DHF 2	79 (42.9)
DHF 3	4 (2.2)
Platelet count (Mean ± SD)	82.39 ± 37.46
NLR (Mean ± SD)	3.66 ± 3.34

A total of 184 patients diagnosed with Dengue Hemorrhagic Fever (DHF) were included in the study. Most of the cohort were female (60.3%), while males comprised 39.7%. The mean age of the participants was 30.55 years (SD ± 15.27), with the highest proportion of patients (36.4%) falling within the 26–45 age group. This was followed by patients aged 18–25 years (25.0%), <18 years (17.9%), 46–65 years (17.4%), and those older than 65 years (3.3%).

In terms of DHF severity, more than half of the participants were classified as Grade I (54.9%), followed by Grade II (42.9%) and a small proportion as Grade III (2.2%). The mean platelet count at admission was 82.39 × 10^3/μL (SD ± 37.46), consistent with the expected thrombocytopenia typically observed in DHF patients. The mean neutrophil-to-lymphocyte ratio (NLR) was 3.66 (SD ± 3.34), indicating a wide variation in inflammatory response among the cohort.

Correlation between Thrombocytopenia and NLR

Based on the analysis of 184 patients with Dengue Hemorrhagic Fever (DHF) at Candi Umbul Hospital, Magelang, the correlation between the neutrophil-to-lymphocyte ratio (NLR) and platelet count was examined. The Spearman's rho correlation coefficient was 0.004 with a p-value of 0.959, indicating no statistically significant correlation between NLR and platelet count in this cohort.

This finding suggests that, within this patient population, NLR and platelet count may reflect different aspects of the disease's pathophysiology. While thrombocytopenia is a hallmark of DHF, resulting from bone marrow suppression and peripheral destruction of platelets, NLR serves as an inflammatory marker, reflecting the balance between neutrophil and lymphocyte counts. The lack of correlation implies that these two parameters may not be directly related in the context of DHF severity.

Previous studies have reported varying relationships between NLR and platelet count in dengue patients. For instance, a study by Ashma et al. (2023) found that NLR and platelet-lymphocyte ratio (PLR) could serve as early indicators of plasma leakage in dengue infection, suggesting a potential link between these hematological parameters and disease progression (Ashma A Susilo SB, 2023). However, other studies have reported no significant association between NLR and platelet count, aligning with the findings of the current analysis (Emokpae M Abdu A, 2016).

The discrepancy in findings across studies may be attributed to differences in study design, patient populations, timing of laboratory measurements, and disease severity. It is important to consider these factors when interpreting the relationship between NLR and platelet count in DHF.

Correlation between NLR and Severity of DHF

The correlation between NLR and DHF severity revealed a statistically significant relationship. The Spearman rank correlation test yielded a correlation coefficient of $\rho = -0.356$ ($p < 0.001$), indicating a moderate, negative correlation between NLR and DHF severity grade. This result suggests that patients with a less severe DHF grade exhibited a higher NLR value, while more severe manifestations of DHF are reflected through lower NLR values.

This trend contrasts with prior assumptions that a high NLR follows worsening clinical severity of DHF. Instead, it may reflect shifts in the immune dynamics over the course of the disease. According to Ishaquet et al., the NLR in dengue cases typically undergoes a reversal for around 4 to 5 days, shifting from a neutrophil or lymphocyte predominance as the patient

transitions from the febrile stage to the critical recovery stage (Ishaque N Siddique MU, 2022). Furthermore, this inverse association suggests that neutrophilic activity may be heightened in the initial innate immune response mechanism in the earlier phases of the illness. Meanwhile, the progression to more severe stages suggests immune exhaustion, which consequently leads to predominant lymphocyte involvement, leading to a lower NLR.

Correlation between Thrombocytopenia and DHF Grades

The relationship between platelet count and DHF severity yielded a moderate, statistically significant negative correlation. The Spearman's rho correlation coefficient was -0.400 ($p<0.001$). This inverse relationship aligns with the pathophysiology of DHF, in which thrombocytopenia serves as a central diagnostic and prognostic hallmark. Platelet depletion in dengue infection is driven by multiple mechanisms, including bone marrow suppression, immune-mediated peripheral destruction, and increased platelet consumption due to endothelial damage and vascular leakage. The worsening of these processes in more severe grades of DHF likely explains the progressive decline in platelet counts observed across severity categories. Furthermore, Rohma et al. highlighted the importance of platelet trends in assessing dengue phases, emphasizing that the critical phase of dengue is marked by the sharpest platelet decline, supporting its utility in grading DHF severity. Given the statistically significant correlation in this study, platelet count remains a reliable marker for DHF severity classification. However, it should be interpreted with other clinical and laboratory parameters, as thrombocytopenia alone may not fully capture the multifactorial risks involved in dengue complications. Serial platelet monitoring is essential for the timely detection of plasma leakage and for anticipating hemorrhagic or shock-related complications (Das S Abreu C, 2022b; Maulida YA Amelia R, 2023).

Correlation Analysis of NLR and Thrombocytopenia Severity across DHF Grades

Neutrophil-Lymphocyte Ratio (NLR), platelet count, and clinical severity grading of Dengue Hemorrhagic Fever (DHF) were studied using Ordinal Logistic Regression Analysis. The model indicated that including NLR and platelet count greatly enhanced the model's discriminative power compared to the intercept-only model (Chi-square = 65.057, $p < 0.001$). It also indicated a moderate explanatory power with a Nagelkerke R^2 of 0.378, meaning approximately 37.8% of the variance in severity of DHF could rage is 0.378 be explained by these two parameters. The goodness of fit test calculated using the deviance statistic returned a p-value of 1.000, suggesting the model appropriately represents the data without substantial change.

The regression model also reported a statistically significant negative relationship to NLR with an estimate = –0.480, $p < 0.001$, and an estimate = –0.027, $p < 0.001$ for platelet count with increasing severity of DHF. The negative correlation due to declining platelet count and increasing severity of DHF does make sense, showing that thrombocytopenia is worse during the later stages of the disease, but the relationship between NLR and its severity is

paradoxical. Instead of increasing as expected in response to inflammation of tissues in the body, NLR was lower in more severe grades of DHF.

This finding may reflect an imbalance in the immune system functioning in a later stage of DHF. With increasing disease severity, initial neutrophilia may be followed by neutropenia superimposed on chronic lymphopenia and immune cell burnout. These changes may lead to a lower NLR while clinically the patient is getting worse. These immune changes may suggest a shift from acute inflammation toward immune suppression or fatigue phenomena not well described in the DHF literature. In this case, the neutrophil-to-lymphocyte ratio would be a marker for the different phases of immune response in DHF, highlighting the contradicting needs for layered, time-sensitive evaluation of NLR in deeper analysis, along with phase-specific interpretation rather than purely assuming dampened immune responses correlate with disease severity.

In the opposite way, the relationship between platelet count and DHF severity follows known pathophysiological concepts. Thrombocytopenia is one of the most severe aspects of dengue and is believed to be caused by enhanced destruction peripherally, suppressed bone marrow cell production, and increased platelet consumption due to vascular leakage and coagulopathy. The continually declining value of platelets with increasing grade of DHF is clinically associated with increasing hemorrhagic complications in severe DHF and serves as an accurate indicator of the disease's progression.

Taken together, these findings highlight the intricate connections that exist between inflammatory markers and blood parameters in DHF. While the platelet count continues to be a strong operational indicator of severity, the patterning of NLR might be more complex and could indicate changes in immune behavior during different phases of the disease. Evaluating both metrics simultaneously could improve risk stratification and clinical judgment, especially in contexts lacking advanced biomarkers. There is an outstanding research that needs to address the longitudinal changes of NLR in DHF concerning the temporal immune dysfunction associated with the disease.

CONCLUSION

This research highlights the clinical importance of the neutrophil-to-lymphocyte ratio (NLR) and platelet count as readily available and inexpensive blood tests for determining the severity of Dengue Hemorrhagic Fever (DHF). Even though NLR is generally accepted to indicate a marker of systemic inflammation, our data suggest an opposite relationship with the severity of DHF, where lower NLR may be seen in more advanced stages of the disease. This paradox is likely capturing a shift toward immune exhaustion during the critical DHF phase, dominated by marrow suppression or the predominance of lymphocyte-dominated responses. On the other hand, the progressive decline in platelet count associated with increasing severity of DHF was also in accordance with established pathophysiological processes, confirming that thrombocytopenia is the hallmark of disease progression and a reliable indicator of clinical decline.

The lack of significant correlation between NLR and platelet count emphasizes further their distinct immunohematological pathways. Their integration, particularly in settings where

sophisticated assays are unattainable, can contribute to a more refined phased assessment of disease trajectory. The cumulative findings support the incorporation of NLR and platelet count into the routine first evaluation and follow-up reviews of DHF patients. Their incorporation into clinical workflows may facilitate improved secondary DHF risk assessment, treatment selection, and overall patient care. Further prospective research is needed to confirm these relationships in more diverse populations and to clarify the relations between inflammation and blood cell alteration over time in the clinical trajectory of DHF.

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