NEUTROPHIL LYMPHOCYTE RATIO (NLR) AS DIAGNOSTIC MARKER IN CHILDREN WITH DENGUE FEVER

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Abstract

Objective: *To compare the sensitivity and specificity of the neutrophil lymphocyte ratio (NLR) in diagnosing dengue fever pediatric patients at a tertiary care center*

Study Design: *Analytical Cross sectional study*

Place and Duration of Study: *Department of Pediatrics, Fauji Foundation Hospital, Rawalpindi from Jun 2024-Dec 2024*

Methodology: *140 children fulfilling selection criteria were enrolled from pediatric emergency. Blood complete picture and dengue IgM serology was sent in all patients. Patients were labeled as dengue positive on NLR if calculated NLR was <1.63 and if IgM levels were >2.85. Primary variables observed were sensitivity and specificity of the neutrophil lymphocyte ratio in diagnosing dengue fever.*

Results: *According to defined cut-off values as per the study design, patients with NLR results suggestive of dengue fever were 98 (70.0%) while patients with dengue IgM results suggestive of dengue were 61 (43.6%) patients. Sensitivity and specificity of NLR when compared with gold standard dengue IgM results showed a sensitivity of 57.1%, with a specificity of 88.1%, with a false positive rate of 11.9% and a false negative rate of 42.9%*

Conclusion: *The study concludes that NLR has satisfactory sensitivity but good specificity as a diagnostic marker for dengue fever and can be used where IgM serology is not available.*

INTRODUCTION

The prevalence of dengue fever has been increasing exponentially since the past decade, and it is projected that 40 million cases of dengue fever are occurring each year worldwide. [1] Special endemic area for the disease include South America, Africa and Southeast Asia. Pakistan, India and Bangladesh report the highest number of cases each year. [2] Multiple risk factors including conducive vector environment, poor sanitation, crowded living and lack of prophylaxis have been reported to increase the disease prevalence in these countries. [3] In Pakistan, the number of cases each year since 2021 have been reported to range between 45000-57000. [4, 5] This staggering number is a major health burden on the healthcare system since dengue with complications require prolonged admission in hospitals.

Diagnostic options developed in the last decade for dengue fever diagnosis include virus isolation and antibody detection. The most reliable method currently in practice is Dengue IgM antibody

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detection both qualitatively and quantitatively. [6] It is considered the gold standard for diagnosis in endemic areas. While it has high sensitivity and specificity for the disease, it cost, availability and routine use in all cases poses issues in our resource constrained setups. [7] Recently, considerable work has been done to identify cost-effective and widely available markers for diagnosis in remote areas and where resource constraints do not allow expensive serological markers to become widely accessible.

Neutrophil lymphocyte ratio (NLR) has been reported to offer as a good diagnostic marker for dengue fever patients. Literature reports good sensitivity and specificity for dengue fever and requires no advance investigations. [8] With the only requirement of blood complete picture for assessment, our study aims to check sensitivity and specificity of NLR compared to gold standard IgM for dengue fever in dengue patients to add it to diagnostic protocols in our pediatric setups.

METHODOLOGY:

This analytical cross-sectional study was carried out at the Department of Pediatrics, Fauji Foundation Hospital Rawalpindi from Oct 2024-march 2024 after approval from the ethical review board vide letter no. 635/RC/FFH/RWP. The sample size was calculated keeping the confidence interval at 95%, margin of error at 5% with diagnostic accuracy of NLR in dengue fever patients at 90.93%.[9] Minimum sample size according to WHO calculator came out to be 127 patients. We included 140 patients in the final study protocol.

Inclusion criteria included pediatric patients ages 2 to 12 years, both genders, presenting with symptoms including fever (>100°F), body aches, flu, cough, chills on clinical examination

Exclusion criteria included patients with fever due to other causes including urinary tract infection, pneumonia, etc., Malignancies (leukemia's, acquired aplastic anemia), liver dysfunction (ALT & AST >40

IU), or renal failure (creatinine > 1.5 mg/dl) patients, immunodeficient states i.e. long-term corticosteroid therapy/organ transplantation or acquired immune deficiency, and other autoimmune and inflammatory diseases.

The study method included all patients as per the inclusion criteria furnished. 140 children fulfilling selection criteria were enrolled from pediatric emergency. Informed consent was taken from parents to use their child's information of research purpose only. Demographics like name, age, gender, weight, duration of symptoms, grade of fever, residence, number of people per room were be noted. Blood sample was taken in a 5cc disposable syringe and was split in 2 parts. Half of the blood sample was sent to the laboratory of the hospital for assessment of neutrophil and lymphocyte count. Reports were assessed and NLR will was noted by a resident pediatrics unaware of the study protocol. Patients were labeled as dengue positive on NLR if NLR was <1.63 according to guidelines. [10] The second half of the blood sample was sent for IgM for detection of dengue virus in blood and dengue was labelled as positive if IgM level was >2.85 according to guidelines. [11] The same data was recorded by a resident pediatrics blinded to the study protocol or its outcomes. At the end of the study, all complied data was sent to the statistical analysis team to assess defined study outcomes.

Primary variables observed were sensitivity and specificity of the neutrophil lymphocyte ratio in diagnosing dengue fever. Demographic data were statistically described in terms of mean±SD, frequencies, and percentages when appropriate. Independent samples t-test was used to compare statistically significant means. Chi-square test was used to compare frequency variables. Sensitivity and specificity was calculated using a 2x2 contingency table with values of NLR and IgM results. A p value of ≤0.05 was considered statistically significant. All statistical calculations were performed using Statistical Package for Social Sciences 26.0

FIGURE-I: PHASES OF THE STUDY (n=140)
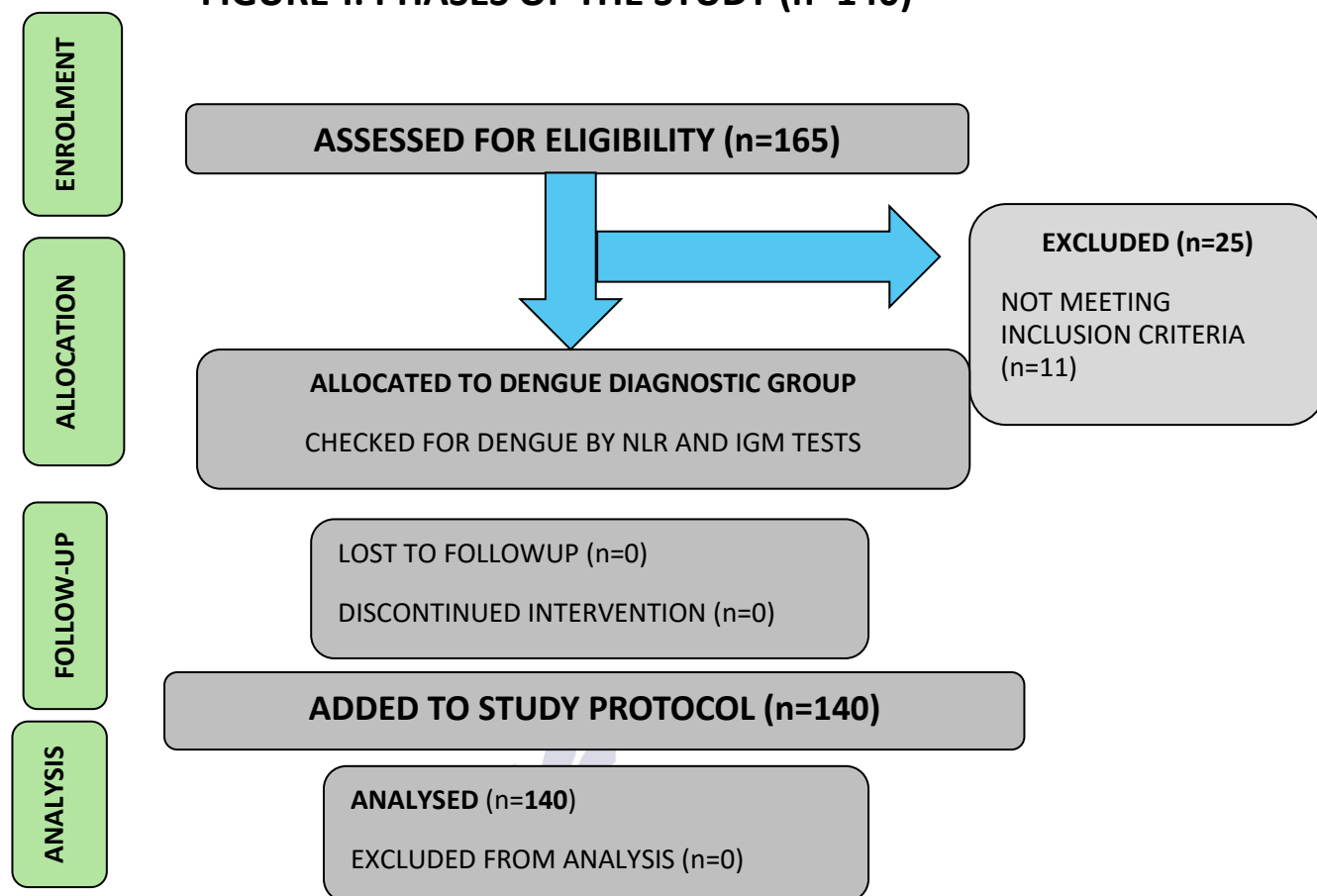
RESULTS:

A total of 140 patients were analyzed in the final study protocol. Mean age of patients in the study group was 6.83±1.30 years and mean weight was 15.36±1.00 kgs. Gender distribution showed 87 (62.1%) males and 53 (37.9%) females in the study group. Residential area of the study group showed 98 (70.0%) of patients lived in the rural while 42 (30.0%) patients lived in the urban areas. Sanitation and quality of living showed poor conditions in 78 (55.7%) of patients, satisfactory arrangements in 36 (25.7%) patients and adequate conditions in 26 (18.6%) patients of the study group (Table-I).

Dengue specific clinical variables showed mean duration of fever in the study group at 5.55±1.02 days. A total of 95 (67.9%) of patients had moderate grade of fever (100-102°F) while 45 (32.1%) patients had high grade of fever (103-106°F) on initial presentation. According to defined cut-off values as per the study design, patients with NLR results suggestive of dengue fever were 98 (70.0%) while patients with dengue IgM results suggestive of dengue were 61 (43.6%) patients (Table-II).

Sensitivity and specificity of NLR when compared with gold standard dengue IgM results showed a sensitivity of 57.1%, with a specificity of 88.1%, with a false positive rate of 11.9% and a false negative rate of 42.9% (Table-III).

TABLES

TABLE-I DEMOGRAPHIC AND CLINICAL CHARACTERISTICS (n=140)

VARIABLE	STUDY GROUP (n=140)
MEAN AGE (YEARS)	6.83±1.30
MEAN WEIGHT (KGS)	15.36±1.00
GENDER	
• MALE	87 (62.1%)
• FEMALE	53 (37.9%)
RESIDENCE	

• RURAL	98 (70.0%)
• URBAN	42 (30.0%)
SANITATION AND QUALITY OF LIVING	
• POOR	78 (55.7%)
• SATISFACTORY	36 (25.7%)
• ADEQUATE	26 (18.6%)

TABLE-II DENGUE SPECIFIC CLINICAL VARIABLES (n=140)

VARIABLE	STUDY GROUP (n=140)
MEAN DURATION OF FEVER (DAYS)	5.55±1.02
GRADE OF FEVER	
• MODERATE GRADE (100-102 °F)	95 (67.9%)
• HIGH GRADE (103-106 °F)	45 (32.1%)
PATIENTS WITH NLR SUGGESTIVE OF DENGUE	98 (70.0%)
PATIENTS WITH IGM SUGGESTIVE OF DENGUE	61 (43.6%)

NLR*= NEUTROPHIL TO LYMPHOCYTE RATIO

TABLE-III SENSITIVITY AND SPECIFICITY OF NLR WHEN COMPARED TO DENGUE IGM RESULTS (n=140)

		IGM SEROLOGY RESULTS (GOLD STANDARD)	
		POSITIVE	NEGATIVE
NLR RESULTS (DIAGNOSTIC TEST)	POSITIVE	56 (57.1%) (A)	05 (11.9%) (B)
	NEGATIVE	42 (42.9%) (C)	37 (88.1%) (D)

A= SENSITIVITY
B= FALSE POSITIVES
C= FALSE NEGATIVES
D= SPECIFICITY

DISCUSSION:

The study concluded that NLR has a very good specificity in pediatric patients with dengue fever. While the sensitivity is satisfactory for dengue fever, the test can be effectively used in areas where dengue serology in expensive or unavailable and patient burden is high to suggest a high clinical suspicion and tailor treatment guidelines accordingly. Comparison of our study and critical analysis with local and international literature shows NLR is being increasingly used as a diagnostic marker in a number of diseases in the neonatal and pediatric age group

including neonatal sepsis, malaria and dengue especially in low resource and modest medical setups which cannot afford advanced diagnostic investigations. [12, 13]

In a study carried out by Moteiro et al, the authors concluded that while NLR proved to be a good diagnostic marker, its assessment of dengue severity was not reliable in differentiation between dengue with and without warning signs proposing that NLR should be co-related with clinical picture and other markers till serology is awaited. [14] This is in contrast to a study carried out by Koundinya et al which concluded that NLR was effective in assessing dengue severity while also predicting the course of the disease on serial measurements. They concluded that lower the NLR the higher the disease severity and decreasing values of NLR <2 corresponded to a poorer prognosis. [15]

When comparing out study with available literature, poor sanitation conditions and rural area localities are associated with increased frequency of dengue infections. These risk factor especially in endemic countries like ours need to be addressed. In a local study done by Mehmood et al, the authors concluded that not only rural areas but urbans slums are also high risk areas for dengue infections. [16] Other local literature on the subject shows that in a study carried out by Sajjad et al, a strong co-relation was found between NLR as well platelet count. The authors reported that decreasing levels of both NLR as well as platelet counts were associated with a worsening prognosis in patients admitted to the hospital. [17] Another study by Tabassum et al concluded that among the hematological parameters used for dengue severity, along with NLR, PLR (platelet to lymphocyte ratio) is also a good marker for dengue diagnosis and assessment of severity. [18] While these markers cannot completely substitute the diagnostic power of dengue IgM serology due to low sensitivity, their high specificity can aid in treatment of patients in peripheral and resource constrained setups when co-related with history and clinical picture.

CONCLUSION:
The study concludes that NLR has satisfactory sensitivity but good specificity as a diagnostic marker for dengue fever and can be used where IgM serology is not available

LIMITATIONS OF THE STUDY:
The limitations are that the study is single center only. Our results are restricted to the pediatric population and studies in adult patients need to be done for more comprehensive results.

CONFLICT OF INTEREST:
None.

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