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Neutrophil Lymphocyte Ratio as Screening Tool for Neonatal Sepsis in Developing Countries

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ABSTRACT

Objective: to recognize an alternate method for screening neonatal sepsis in patients while considering the sensitivity and accuracy of the diagnosis.

Study Design: Cross sectional study.

Place and Duration of Study: Combined Military Hospital, Rawalpindi Pakistan, from Jun to Dec 2021.

Methodology: Patients admitted to the Neonatal Intensive Care Unit of the Combined Military Hospital were included in the study. Demographic data, including age, weight, gender, and gestational age, were recorded. In addition, a statistical comparison of total leucocyte count, absolute neutrophil and lymphocyte counts, neutrophil-lymphocyte ratio, and C-reactive protein test was carried out for the cases and controls.

Results: It was evident from the results that a significant correlation between the neutrophil-lymphocyte ratio and neonatal sepsis exists. The mean NLR in non-septic individuals was (1.8±1.3) and in septic individuals, it was (2.9±1.9). The *p*-value was 0.001. Furthermore, ROC showed that at an NLR value of 1.6, sensitivity was 0.594 and 1-specificity 0.295.

Conclusion: Neutrophil lymphocyte ratio was an efficient screening tool for determining sepsis in neonates, with comparable results to the C-reactive protein test.

Keywords: Lymphocyte, Neonatal sepsis, Neutrophil, Neutrophil-lymphocyte ratio.

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INTRODUCTION

Neonatal sepsis is a leading cause of morbidity and mortality in this subgroup of the population.[1] The Global Burden of Disease Study carried out in 2016-2017 estimated an annual incidence of 1.3 million cases worldwide, associated with 203,000 neonatal deaths.[1] In Pakistan, sepsis is responsible for the deaths of 49 babies per 1000 live births.[2] A high index of suspicion, along with diagnostic tests, is required since features of sepsis in neonates are nonspecific and ill-defined.[3] Most babies admitted to the neonatal intensive care unit either require antibiotics or are administered antibiotics for one reason.[4,5]

Although blood culture is the gold standard for diagnosing neonatal sepsis, chances of getting a truly positive blood culture in neonates are minimal due to multiple reasons mentioned in the studies done in the past.[6] In addition, blood culture results take 4-7 days and are expensive, especially considering the burden of NNS in developing countries.[7]

Acute phase reactants, such as CRP and procalcitonin, are used to screen for sepsis. Procalcitonin has a sensitivity of 73.6%, specificity of 97% and positive predictive value of 91% in screening for NNS7.[8] However, procalcitonin is costly and unavai-lable at most centres in low-income countries. Multiple studies have shown CRP to be effective as a screening tool, with a sensitivity of 65.6 -95% and specificity of 82-92%8. CRP is faster than blood culture and cheaper than procalcitonin, which makes it a commonly used investigation to screen for NNS.[9] It has been stated in studies done previously that CRP starts to rise at 6 to 8 hours of life in neonates with infection and peaks at 24 hours, so if CRP remains persistently normal in an otherwise well baby, antibiotics can be safely stopped.[10]

This study aims to recognize an alternate method for screening neonatal sepsis in patients while considering the sensitivity and accuracy of the diagnosis. It was hypothesized that NLR is as accurate as CRP in detecting NNS.

METHODOLOGY

The cross sectional study was conducted at Neonatal Intensive Care Unit of the Combined Military Hospital Rawalpindi Pakistan, from June to December 2021 after IERB approval. A sample size of 186 was calculated using expected sensitivity of NLR of 80.8%,[11] expected specificity of 42.3%, the prevalence

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of 44%. Non-probability consecutive sampling technique was used.

Inclusion Criteria: Patients of either gender admitted to the NICU were included in the study. Laboratory diagnosis for this study was that any patient with positive blood culture, positive PCR, or procalcitonin>2.0 ng/ml was diagnosed with neonatal sepsis, regardless of signs and symptoms.

Exclusion Criteria: Those having deliveries with early rupture of membrane, preterm, diabetics and having other septic risk factors were included in the study, whereas patients with other diseases, such as haematological system diseases, major congenital malformation, and cyanotic congenital heart disease, were excluded from the study.

All patients admitted to NICU, who did not meet the exclusion criteria, were enrolled in the study. The age, sex, gestational age and weight of each patient was recorded in a proforma. CBC and CRP were taken at admission. Where possible, procalcitonin and blood culture was sent. Subjects were divided into two groups: those with evidence of NNS were labelled septic NNS. Subjects with no evidence of NNS were taken as controls.

Neonatal sepsis was diagnosed using IMCI and WHO criteria for severe infection in children i.e., 1) Convulsions, drowsy or unconscious, decreased activity, bulging fontanelle, 2) Respiratory rate >60 breaths /min, grunting, severe chest indrawing, central cyanosis, 3) Poor perfusion, rapid and weak pulse, 3) Jaundice, poor feeding, abdominal distention, 4) Skin pustules, periumbilical erythema, or purulence, 5) Edema or erythema overlying bones or joints, 6) Temperature >37.7°C (99.9°F) or less than 35.5°C (95.9°F).[11,12]

Data were analysed using Statistical Package for the Social Sciences (SPSS) version 23.00 and MS Excel 2016 software. Mean±SD were calculated for continuous variables. Frequency and percentage were calculated for categorical variables. The comparison of mean of total leucocyte count, absolute neutrophil, lymphocyte counts, NLR, and CRP between the sepsis and control groups was made by an independent sample t-test. The *p*-value lower than or up to 0.05 was considered as significant.

RESULTS

A total of 186 neonates were included in our study, of which 10(59.1%) were male and 76(40.9%) were female. The mean gestational age of participants was 32.6±5.1 weeks. Based on the CRP, 64(34.4%) neonates were diagnosed as having neonatal sepsis, whereas 122(65.6%) neonates did not have neonatal sepsis (Table-I). Blood complete profile was compared between septic and non-septic neonates. Mean TLC in septic neonates was found to be 15.1±8.4x10^3/µl, as opposed to 14.2±6.2 x10^3/µl in non-septic neonates. Platelet cell volume was 0.42±0.1fl in septic neonates and 0.57±0.8 fl in non-septic neonates.

Table-I: Baseline profile of neonates included in study (n=186)

Gestational Age	**32.6±5.1 Weeks**
TLC	14.5±7.0
RBC	4.4±0.9
Hb	15.0±3.4
PCV	0.52±0.6
MCV	101.9075±8.2
MCH	33.5±3.1
MCHC	33.0±2.6
PLATELETS	231.3±137.2
CRP	15.8±29.4
NLR	2.2±1.6
NNS	
Sepsis No sepsis	64(34.4%) 122(65.6%)

Haemoglobin count was lower in septic neonates, 14.3±3.6 g/dl, compared to 15.4±3.3 g/dl in non-septic neonates. The difference in haemoglobin levels was statistically significant (*p*=0.043). CRP was the main test used to categorize neonates as septic and non-septic. There was a marked difference in CRP in septic neonates (40.1±39.8mg/l) and non-septic individuals (2.6±2.2mg/l), which was significant with a *p*-value of 0.001 (Table-II).

Table-II: Comparison of septic versus non-septic neonates (n=186)

Variables	**Sepsis (n=64)**	**No Sepsis (n=122)**	***p*-value**
Age (Days)	4.0±5.4	3.0±3.3	0.113
Sex			
Male Female	36(56.2%) 28(43.8%)	74(60.7%) 48(39.3%)	0.561
Gestational age			
TLC	15.1±8.4	14.2±6.2	0.412
RBC	4.3±0.9	4.5±0.9	0.072
Hb	14.3±3.6	15.4±3.3	0.043
PCV (fl)	0.42±0.1	0.57±0.8	0.151
MCV	98.7±9.8	103.6±6.8	0.001
MCH	32.5±3.2	34.0±3.0	0.002
MCHC	33.2±2.3	32.9±2.8	0.475
PLATELETS	216.4±164.0	239.1±122.2	0.283
CRP	40.1±39.5	2.6±2.2	0.001
NLR	2.9±1.9	1.8±1.3	0.001

There was a significant correlation between NLR and neonatal sepsis. The mean NLR in non-septic individuals was (1.8±1.3) and in septic individuals, it was (2.9±1.9). The *p*-value was 0.001. Furthermore, ROC showed that at an NLR value of 1.6, sensitivity was 0.594 and 1-specificity 0.295 (Figure).

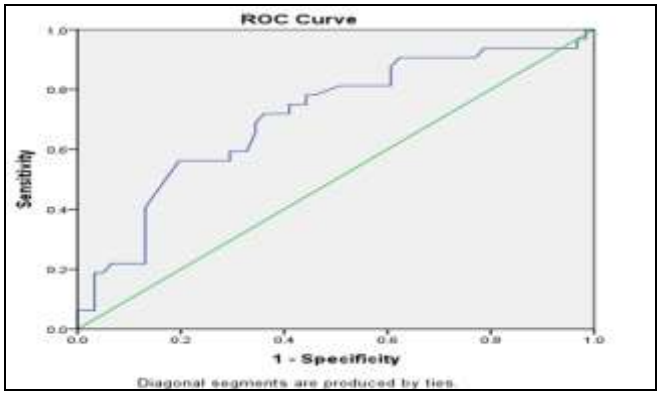

Figure: ROC curves of NLR to predict the presence of neonatal sepsis (n=186)

DISCUSSION

Neonatal sepsis remains a leading cause of death in newborns in developing countries despite improvements in the health care system. It accounts for 30-50% of neonatal mortality in developing countries.[12] Our study has demonstrated the importance of the NLR ratio in neonatal sepsis, where its sensitivity was 59% and specificity of 29%. A similar result was reported by Li *et al*. in China, where the sensitivity of NLR was found to be 51% and the specificity of 75%.[13] Neutrophil to Lymphocyte ratio (NLR) is a cost-effective and sensitive test in diagnosing neonatal sepsis. NLR has emerged as a new effective diagnostic tool in diagnosing neonatal sepsis at an earlier stage. This view was supported by Santosh K Panda and colleagues, who found the sensitivity of NLR to be 68.3% and specificity of 46.2% using a cut-off value of more than 1.7.[14] Omran *et al*. found a sensitivity of NLR to be 80% and a specificity of 57% using a cut-off value of 2.7% in newborns in Egypt.[15] In another study carried out in Indonesia sensitivity of NLR was found to be 81.8% and specificity of 66.1% using a cut-off value of 2.3%.[16] In another study in China, the sensitivity of NLR was 77%, and specificity was 78%, using a cut-off value of 3.16%.[17] In a study carried out in Korea, the sensitivity of NLR was found to be 83.3% and specificity of 93.3% using a cut-off value of 1.27%.[18] They found a strong relationship between C reactive protein and neutrophil to lymphocyte ratio in sepsis in the first week of premature new-born babies.

LIMITATIONS OF STUDY

The study demonstrated the potential use of NLR as a diagnostic marker for neonatal sepsis. However, the maternal characteristics, the neonates' clinical presentation, and the sepsis's timing were not recorded. Similarly, the early and late onset of neonatal sepsis was also not considered.

CONCLUSION

The results of the study demonstrated a sufficient correlation between the use of NLR in estimating neonatal sepsis in patients. Furthermore, it was evident that NLR can be used as a surrogate marker for neonatal sepsis, especially in developing countries, by dividing the absolute number of lymphocyte neutrophils from the complete blood count. This is a simple and inexpensive check.

Conflict of Interest: None.

Author's Contribution

Following authors have made substantial contributions to the manuscript as under:

NN: Supervision, Conception, Study design, analysis and Interperitation of data, Critically reviewed manuscript & approval for the final version to be published.

TKS: Co-supervision, Data entry, analysis and interpretation, manuscript writing & approval for the final version to be published.

SZ & HB: Critically reviewed, Drafted manuscript & approval for the final version to be published.

SH & JJ: Data colllection, Entry and analysis of data, preparation of rough draft & approval for the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investi-gated and resolved.

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