

Evaluation of Neutrophil-To-Lymphocyte Ratio, Platelet-To-Lymphocyte Ratio and Procalcitonin in Early Onset Neonatal Sepsis At A Tertiary Care Center In India

Nandini Singh¹, Bindu T Nair^{2*}, Poonam Chhillar Vij³, Meenakshi Bothra⁴, Praneta Swarup⁵, Rajeev Kumar Thapar⁴, Gurmeet Singh¹, Alisha Jain¹, Adeeba Ahmed¹

¹ Resident, Department of Pediatrics, Sharda School of Medical Sciences & Research, Greater Noida

^{2*} Professor, Department of Pediatrics, (Corresponding Author), Sharda School of Medical Sciences & Research, Greater Noida

³ Assistant Professor, Department of Pediatrics, Sharda School of Medical Sciences & Research, Greater Noida

⁴ Professor, Department of Pediatrics, Sharda School of Medical Sciences & Research, Greater Noida

⁵ Associate Professor, Department of Pediatrics, Sharda School of Medical Sciences & Research, Greater Noida

*Corresponding Author: Dr. Bindu T Nair,

Professor (Department of Paediatrics), Sharda School of Medical Sciences & Research, Sharda University, Greater Noida, Uttar Pradesh, India, PIN – 201306. Email ID: bindu.nair@sharda.ac.in, ORCID ID: 0000 0003 4957 0437

ABSTRACT

Background:

Early-onset neonatal sepsis (EONS) is a major cause of neonatal morbidity and mortality, particularly in low- and middle-income countries where early diagnostic facilities are limited. Simple and rapid biomarkers are needed for early identification of neonatal sepsis. The present study aimed to evaluate the predictive role of neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), and procalcitonin (PCT) in diagnosing EONS.

Materials and Methods:

This prospective observational cross-sectional study was conducted in the Department of Pediatrics, of a tertiary care hospital of North India. A total of 120 neonates with suspected early-onset sepsis within the first 72 hours of life were enrolled. Clinical evaluation, complete blood count, blood culture, and procalcitonin levels were assessed. NLR and PLR were calculated from routine hematological parameters.

Results:

Among the 120 neonates studied, 14 (11.7%) were diagnosed with EONS. Septic neonates showed significantly higher mean NLR (4.62 ± 1.48) and PLR (84.0 ± 53.0) compared to non-septic neonates (1.92 ± 0.87 and 61.9 ± 26.1 respectively). Mean procalcitonin levels were also significantly elevated in the sepsis group (7.2 ± 3.4 ng/mL) compared with the non-sepsis group (4.64 ± 7.66 ng/mL). ROC analysis demonstrated that PCT had the highest diagnostic accuracy with an area under the curve (AUC) of 0.788, followed by NLR (AUC 0.678) and PLR (AUC 0.644). The optimal cut-off values identified were 6.49 ng/mL for PCT, 3.23 for NLR, and 67.2 for PLR. At these thresholds, PCT showed sensitivity of 71.4% and specificity of 79.2%, while NLR and PLR demonstrated moderate diagnostic performance.

Conclusion:

Neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, and procalcitonin are useful biomarkers for early detection of neonatal sepsis. Among these, procalcitonin showed the highest diagnostic accuracy, while NLR and PLR offer inexpensive and easily available screening tools derived from routine blood tests.

Keywords: Early-onset neonatal sepsis, neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, procalcitonin, neonatal biomarkers, diagnostic accuracy.

INTRODUCTION

Neonatal sepsis remains a major cause of neonatal morbidity and mortality worldwide, particularly in low- and middle-income countries where limitations in infection control and diagnostic facilities contribute to poor outcomes.^{1,2} It is defined as a systemic infection occurring within the first 28 days of life and may manifest as bacteremia, pneumonia, meningitis, urinary tract infection, or other systemic infections.^{3,4} Based on the time of onset, neonatal sepsis is classified into early-onset neonatal sepsis (EONS), occurring within the first 72 hours of life, and late-onset neonatal sepsis thereafter.^{5,6} EONS is commonly acquired through vertical transmission of microorganisms from the maternal genital tract before or during delivery.⁶ Common causative organisms include Group B Streptococcus, Escherichia coli, Klebsiella species, and Listeria monocytogenes.⁷

Blood culture remains the gold standard for diagnosis, but its clinical utility is limited by low sensitivity, delayed reporting, and reduced yield following maternal antibiotic exposure.^{8,9} In addition, clinical manifestations such as respiratory distress, lethargy, feeding intolerance, and temperature instability are nonspecific and often overlap with non-infectious neonatal conditions.⁴ Conventional hematological indices derived from complete blood count have limited diagnostic accuracy because of physiological variations in neonates.¹⁰

Recently, neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR), obtained from routine blood counts, have gained attention as inexpensive and readily available inflammatory markers.^{11,12} Earlier studies demonstrated that elevated NLR and PLR values are associated with increased inflammatory activity and may aid in the early identification of neonatal sepsis.^{13,14,15} More recent studies have further highlighted their diagnostic accuracy and potential utility as adjunctive biomarkers in early-onset neonatal sepsis.^{16,17,18} Procalcitonin (PCT) has also emerged as a promising biomarker because of its rapid rise during bacterial infection and systemic inflammatory response.^{8,9,19,20} Therefore, this study aimed to evaluate the predictive value of NLR, PLR, and PCT in early-onset neonatal sepsis.

Materials and Methods

This study was a observational cross-sectional study conducted in the Department of Pediatrics, of a tertiary care hospital of North India. The study was carried out over a period of 18 months from April 2024 to November 2025 after obtaining approval from the Institutional Ethics Committee of the institute. The research was carried out following the guidelines of the ethics committee as listed in the ethics statement and was in accordance with Helsinki Declaration. Written informed consent was taken from the parents or legal guardians of the enrolled neonates.

A total of 120 neonates admitted to the neonatal intensive care unit (NICU) with clinical suspicion of early-onset neonatal sepsis within the first 72 hours of life were enrolled. Neonates presenting with features suggestive of sepsis such as respiratory distress, temperature instability, lethargy, poor feeding, apnea or circulatory compromise were evaluated for inclusion. Neonates with major congenital anomalies, chromosomal abnormalities, or those who had received antibiotic therapy prior to admission were excluded from the study.

For each enrolled neonate, detailed maternal and perinatal histories were recorded, including gestational age, birth weight, sex, parity of the mother, and mode of delivery. A thorough clinical examination was performed at the time of admission. Laboratory investigations were performed as part of the standard sepsis evaluation protocol. Blood samples were obtained under strict aseptic precautions before the initiation of antibiotic therapy. Investigations included complete blood count (CBC) with differential leukocyte count, platelet count, blood culture, and serum procalcitonin levels.

From the hematological parameters obtained from the CBC, the neutrophil-to-lymphocyte ratio (NLR) was calculated by dividing the absolute neutrophil count by the absolute lymphocyte count, while the platelet-to-lymphocyte ratio (PLR) was calculated by dividing the platelet count by the absolute lymphocyte count. Serum procalcitonin levels were measured using the institutional laboratory immunoassay method. Blood cultures were processed using standard microbiological techniques to identify bacterial pathogens.

Statistical Analysis

All collected data were entered into Microsoft Excel and subsequently analyzed using Statistical Package for the Social Sciences (SPSS) software, version 25. Continuous variables were summarized as mean $\pm$ standard deviation, while categorical variables were expressed as frequencies and percentages. Comparisons between septic and non-septic neonates were performed using appropriate statistical tests depending on the nature and distribution of the data.

Associations between categorical variables were analyzed using the Chi-square test. Differences in continuous variables between groups were evaluated using the independent samples t-test. The correlation between procalcitonin, neutrophil-to-lymphocyte ratio, and platelet-to-lymphocyte ratio was assessed using Pearson's correlation coefficient.

Receiver operating characteristic (ROC) curve analysis was performed to determine the diagnostic performance of procalcitonin, NLR, and PLR in predicting early-onset neonatal sepsis. The area under the curve (AUC) was calculated to evaluate the discriminative ability of each biomarker. Optimal cut-off values were determined using Youden's index, and corresponding sensitivity, specificity, and accuracy were calculated. A p-value of less than 0.05 was considered statistically significant for all statistical tests.

RESULTS

This prospective observational study included a total of 120 neonates. Sepsis screen was sent between 6 – 12 hours after birth.

Table 1: Demography of study population (N = 120)

Variable	Category	EONS Present n (%)	EONS Absent n (%)	p-value	Odds Ratio (95% CI)
Overall EONS Status	Present	14 (11.7%)	—	—	—
	Absent	106 (88.3%)	—	—	—
Gender	Female (n=51)	5 (9.8%)	46 (90.2%)	0.310	0.68 (0.21–2.20)
	Male (n=69)	9 (13.0%)	60 (87.0%)		
Mode of Delivery	LSCS (n=65)	7 (10.8%)	58 (89.2%)	0.730	1.07 (0.36–3.18)
	Vaginal (n=55)	7 (12.7%)	48 (87.3%)		
Gestational Age	Preterm (n=39)	8 (20.5%)	31 (79.5%)	0.036*	5.48 (1.71–17.55)
	Term (n=81)	6 (7.4%)	75 (92.6%)		
Maternal Parity	Primigravida (n=56)	8 (14.3%)	48 (85.7%)	0.403	0.53 (0.16–1.76)
	Multigravida (n=64)	6 (9.4%)	58 (90.6%)		

Preterm neonates had a significantly higher incidence of EONS compared to term neonates (20.5% vs. 7.4%, $p = 0.036$), with an approximately 5.5-fold increased risk. No statistically significant association was observed between EONS and gender, mode of delivery, or maternal parity. Prematurity emerged as the major risk factor for EONS in this cohort (Table 1).

Table 2: Microbiological Profile and Hematological Parameters in EONS and Non-EONS Neonates (N = 120)

Variable	Category/Parameter	EONS Present (n=14)	EONS Absent (n=106)	p-value
Blood Culture Isolates	Klebsiella pneumoniae	3 (21.4%)	—	—
	Citrobacter spp.	2 (14.3%)	—	—
	Acinetobacter spp.	2 (14.3%)	—	—
	Candida spp.	2 (14.3%)	—	—
	Escherichia coli	1 (7.1%)	—	—
	MRCONS	1 (7.1%)	—	—
	MSCONS	1 (7.1%)	—	—
	Enterococcus spp.	1 (7.1%)	—	—
	Staphylococcus spp.	1 (7.1%)	—	—
Hematological Parameters	TLC (/mm ³), Mean $\pm$ SD	17,534 $\pm$ 5,804	19,632 $\pm$ 6,465	0.251
	Neutrophil %, Mean $\pm$ SD	70.8 $\pm$ 8.83	65.1 $\pm$ 9.76	0.042*
	Lymphocyte %, Mean $\pm$ SD	19.4 $\pm$ 8.09	25.7 $\pm$ 9.44	0.018*
	Platelet count (/mm ³), Mean $\pm$ SD	226,857 $\pm$ 104,596	262,236 $\pm$ 72,749	0.108
	Procalcitonin (ng/mL), Mean $\pm$ SD	7.2 $\pm$ 3.4	4.64 $\pm$ 7.66	<0.001*

Microbiological and hematological parameters of the EONS and non- EONS neonates are shown in Table 2. Procalcitonin levels were significantly elevated in the EONS group ($p < 0.001$), supporting its role as an important biomarker for early neonatal sepsis (Table 2).

Table 3: Comparison, Correlation, and Diagnostic Accuracy of PCT, NLR, and PLR in Early-Onset Neonatal Sepsis (EONS)

Variable	Findings	Statistical Significance
----------	----------	--------------------------

Comparison Between EONS and Non-EONS Neonates	NLR significantly higher in EONS (5.23 ± 5.22 vs. 3.01 ± 1.62)	$p < 0.001^*$
	PLR significantly higher in EONS (84.0 ± 53.0 vs. 61.9 ± 26.1)	$p = 0.012^*$
Comparison by Gestational Age (Non-EONS)	NLR higher in term neonates (3.26 ± 1.73 vs. 2.42 ± 1.13)	$p = 0.014^*$
	PLR higher in preterm neonates (70.15 ± 25.23 vs. 58.50 ± 25.85)	$p = 0.036^*$
	PCT showed no significant difference	$p = 0.106$
Comparison by Gestational Age (EONS)	PCT, NLR, and PLR were higher in preterm neonates compared to term neonates	Not significant
Correlation Analysis	PCT positively correlated with NLR ($r = 0.248$)	$p = 0.006^*$
	NLR strongly correlated with PLR ($r = 0.641$)	$p < 0.001^*$
	No significant correlation between PCT and PLR ($r = 0.086$)	$p = 0.348$
Diagnostic Accuracy (ROC Analysis)	PCT: AUC = 0.788 (0.642–0.934), Cut-off = 6.49 ng/mL, Sensitivity = 71.4%, Specificity = 79.2%	Highest diagnostic accuracy
	NLR: AUC = 0.678 (0.524–0.831), Cut-off = 3.23, Sensitivity = 71.4%, Specificity = 62.3%	Moderate accuracy
	PLR: AUC = 0.644 (0.487–0.800), Cut-off = 67.2, Sensitivity = 64.3%, Specificity = 63.2%	Moderate accuracy

Table 3 shows that neonates with EONS demonstrated significantly higher NLR and PLR values compared to non-septic neonates. Correlation analysis showed a significant positive association between PCT and NLR, and a strong correlation between NLR and PLR. ROC analysis demonstrated that procalcitonin had the best diagnostic performance for predicting EONS, with higher sensitivity and specificity than NLR and PLR, suggesting that PCT is a superior biomarker, while NLR and PLR may serve as useful adjunctive inflammatory markers.

DISCUSSION

Among hematological parameters, septic neonates demonstrated significantly higher neutrophil percentages and lower lymphocyte percentages, resulting in elevated NLR values. NLR reflects the balance between neutrophil-mediated innate immune activation and lymphocyte suppression during systemic inflammation. In the present study, NLR was significantly higher in neonates with EONS compared to non-septic neonates, indicating its potential utility as an inflammatory marker. Previous studies by Can et al.¹², Arcagok et al.¹³, and Nasser et al.¹⁴ also demonstrated elevated NLR and PLR values in neonatal sepsis. Similar findings were further reported by Soliman et al.¹⁵, Zhang et al.¹⁶, Karabulut et al.¹⁷, and Wilar et al.¹⁸, supporting the diagnostic utility of hematological ratios in neonatal sepsis. However, the diagnostic accuracy observed in the present study was moderate compared with some earlier reports, possibly due to differences in patient characteristics, severity of illness, and timing of sample collection.

PLR was also significantly elevated in neonates with EONS. PLR reflects platelet activation along with lymphocyte suppression during systemic inflammatory responses. Although PLR demonstrated lower diagnostic performance compared to NLR, it still showed moderate utility as an adjunctive biomarker. Because both NLR and PLR can be easily derived from routine complete blood count parameters, they represent inexpensive and readily available tools for early sepsis evaluation, particularly in resource-limited settings. The observations of the present study are consistent with previous reports demonstrating the role of PLR as a supportive inflammatory marker in neonatal sepsis^{13,15,17}

Procalcitonin showed the best diagnostic performance among all evaluated biomarkers. Septic neonates had significantly higher PCT levels than non-septic neonates, and ROC analysis demonstrated fair diagnostic accuracy with an AUC of 0.788. Blood culture remains the gold standard for diagnosis of neonatal sepsis, although delayed reporting and low sensitivity limit its early clinical utility.^{8,9} PCT rises rapidly following bacterial infection due to stimulation by inflammatory cytokines and bacterial endotoxins, making it a useful early biomarker for systemic bacterial infection.^{10,11} Previous studies by Chiesa et al.⁸ and Pontrelli et al.⁹ reported good diagnostic performance of PCT in neonatal sepsis. Similar observations were also reported by Shah et al.⁴, Ghosh et al.¹⁹, and Kucuk et al.²⁰, who demonstrated high sensitivity and specificity of PCT for early diagnosis of neonatal sepsis.

In the present study, a cut-off value of 6.49 ng/mL provided optimal sensitivity and specificity for predicting EONS.

Correlation analysis demonstrated a significant positive association between PCT and NLR, while NLR also showed a strong positive correlation with PLR. These findings suggest that all three biomarkers reflect interconnected inflammatory pathways activated during neonatal sepsis. Bacterial invasion stimulates release of pro-inflammatory cytokines such as interleukin-6, interleukin-1 β , and tumor necrosis factor- α , leading to neutrophilia, lymphocyte suppression, platelet activation, and increased PCT synthesis^{10,11,19}. An important observation of the present study was that combining PCT with hematological ratios improved overall diagnostic performance compared with use of individual biomarkers alone. Similar findings regarding combined biomarker utility were reported in earlier neonatal sepsis studies^{12,18,19}.

Despite these findings, certain limitations must be acknowledged. The study was conducted at a single tertiary care center with a relatively small sample size, limiting generalizability of the results. Maternal risk factors such as prolonged rupture of membranes, maternal fever, and chorioamnionitis were not evaluated in detail. In addition, serial measurements of biomarkers were not performed, preventing assessment of dynamic changes during the course of infection. Larger multicenter studies with longitudinal biomarker monitoring are therefore needed to validate these findings and establish standardized diagnostic cut-off values.

CONCLUSION

The present study demonstrates that PCT, NLR, and PLR are useful adjunctive biomarkers for early detection of neonatal sepsis. Among these, procalcitonin showed the highest diagnostic accuracy, whereas NLR and PLR provide simple, inexpensive, and readily available inflammatory markers derived from routine hematological investigations. Combined use of these biomarkers may improve diagnostic accuracy and support early clinical decision-making in neonates with suspected EONS, particularly in resource-constrained healthcare settings.

Funding: NIL

Conflicts of Interest: NIL

Clinical trial number: not applicable

Data availability: Data is available to be used by the journal for publishing the article

Consent to publish: YES

REFERENCES

1. Shane AL, Sánchez PJ, Stoll BJ. Neonatal sepsis. *Lancet*. 2017;390:1770-80.
2. Fleischmann-Struzek C, Goldfarb DM, Schlattmann P, Schlapbach LJ, Reinhart K, Kissoon N, et al. Global burden of neonatal sepsis: a systematic review. *Lancet Respir Med*. 2018;6(3):223-30.
3. Lawn JE, Cousens S, Zupan J. 4 million neonatal deaths: when? where? why? *Lancet*. 2005;365:891-900.
4. Shah BA, Padbury JF. Neonatal sepsis: new insights. *Virulence*. 2014;5:170-8.
5. Puopolo KM, Benitz WE, Zaoutis TE. Management of neonates at risk for early-onset sepsis. *Pediatrics*. 2018;142:e20182894.
6. Polin RA. Management of neonates with suspected early-onset bacterial sepsis. *Pediatrics*. 2012;129:1006-15.
7. Camacho-Gonzalez A, Spearman PW, Stoll BJ. Neonatal infectious diseases. *Pediatr Clin North Am*. 2013;60:367-89.
8. Chiesa C, Panero A, Rossi N, Stegagno M, De Giusti M, Osborn JF, et al. Reliability of procalcitonin concentrations for the diagnosis of sepsis in critically ill neonates. *Clin Infect Dis*. 1998;26(3):664-72.
9. Pontrelli G, De Crescenzo F, Buzzetti R, Jenkner A, Balduzzi S, Calò Carducci FI, et al. Accuracy of serum procalcitonin for the diagnosis of sepsis in neonates and children with systemic inflammatory syndrome: a meta-analysis. *BMC Infect Dis*. 2017;17(1):302.
10. Hotchkiss RS, Moldawer LL, Opal SM, Reinhart K, Turnbull IR, Vincent JL. Sepsis and septic shock. *Nat Rev Dis Primers*. 2016;2:16045.
11. Farkas JD. Neutrophil-to-lymphocyte ratio as marker of inflammation. *J Intensive Care Med*. 2020;35:593-600.
12. Can E, Hamilcikan S, Can C. The value of neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio for detecting early-onset neonatal sepsis. *J Pediatr Hematol Oncol*. 2018;40:e229-32.
13. Arcagok B, Karabulut B. Platelet-to-lymphocyte ratio in neonatal sepsis. *J Clin Lab Anal*. 2019;33:e22606.

14. Nasser M, Youssef O, Shalaby S. NLR and PLR in early neonatal sepsis. J Neonatal Perinatal Med. 2020;13:205-12.
15. Soliman AT, El-Sayed HM, Abdelrahman A. Hematological indices in neonatal sepsis. Pediatr Neonatol. 2022;63:270-6.
16. Zhang Y, Zhao J, Wang Y. Diagnostic accuracy of NLR in neonatal sepsis. J Matern Fetal Neonatal Med. 2021;34:2680-7.
17. Karabulut B, Arcagok BC. New inflammatory markers in neonatal sepsis. Pediatr Neonatol. 2020;61:397-402.
18. Wilar R, Wahani A, Suryanto DY. Diagnostic performance of neutrophil-to-lymphocyte ratio in neonatal sepsis. BMC Pediatr. 2023;23:212.
19. Ghosh S, Mukhopadhyay K. Biomarkers in neonatal sepsis. Indian J Pediatr. 2022;89:315-22.
20. Kucuk O, Bas AY, Demirel N. Diagnostic value of procalcitonin in neonatal sepsis. J Matern Fetal Neonatal Med. 2018;31:2357-62