

Huseynova N.F.

Scientific Research Institute of Pediatrics, Baku, Azerbaijan

DIAGNOSTIC VALUE OF PRESEPSIN AND INTERLEUKIN-33 IN BACTERIAL INFECTIONS IN TERM NEONATES

e-mail: mic_amu@mail.ru

This study evaluated the diagnostic value of presepsin and interleukin-33 (IL-33) in term neonates with bacterial infections. The prospective study included 36 term neonates with neonatal pneumonia or sepsis and 13 healthy controls. Serum C-reactive protein, procalcitonin, presepsin, IL-33, and lactate were assessed. Infected neonates had significantly higher concentrations of all biomarkers than controls ($p < 0.05$), with the greatest increases observed for presepsin and IL-33. ROC analysis demonstrated excellent diagnostic performance for presepsin (AUC=0.971), CRP (AUC=0.971), lactate (AUC=0.965), and IL-33 (AUC=0.961), whereas procalcitonin showed lower accuracy (AUC=0.789). Presepsin and IL-33 demonstrated 94.1 % sensitivity and 100 % specificity. A strong positive correlation was found between presepsin and IL-33. These findings indicate that presepsin and IL-33 are highly informative biomarkers whose combined assessment with conventional laboratory markers may improve early diagnosis and clinical management of bacterial infections in term neonates.

Key words: term neonates, bacterial infection, neonatal sepsis, neonatal pneumonia, presepsin, interleukin-33, biomarkers, C-reactive protein, procalcitonin.

Гусейнова Н.Ф.

ДІАГНОСТИЧНЕ ЗНАЧЕННЯ ПРЕСЕПСИНУ ТА ІНТЕРЛЕЙКІНУ-33 ПРИ БАКТЕРІАЛЬНИХ ІНФЕКЦІЯХ У ДОНОШЕНИХ НОВОНАРОДЖЕНИХ

У дослідженні оцінювали діагностичну значимість пресеписину та інтерлейкіну-33 (IL-33) у доношених новонароджених із бактеріальними інфекціями. У проспективне дослідження було включено 36 доношених новонароджених із неонатальною пневмонією або сепсисом та 13 здорових новонароджених у контрольній групі. Визначали рівні С-реактивного білка, прокальцитоніну, пресеписину, IL-33 і лактату. У новонароджених з інфекцією концентрації всіх досліджуваних біомаркерів були статистично значуще вищими, ніж у контрольній групі ($p < 0,05$), при цьому найбільш виражене підвищення відзначалося для пресеписину та IL-33. ROC-аналіз показав високу діагностичну ефективність пресеписину (AUC=0,971), С-реактивного білка (AUC=0,971), лактату (AUC=0,965) та IL-33 (AUC=0,961), тоді як прокальцитонін характеризувався нижчою точністю (AUC=0,789). Чутливість пресеписину та IL-33 становила 94,1 %, специфічність – 100 %. Між пресеписином та IL-33 виявлено сильну позитивну кореляцію. Отримані результати свідчать про високу діагностичну інформативність пресеписину та IL-33 і перспективність їхнього спільного застосування з традиційними лабораторними маркерами для ранньої діагностики бактеріальних інфекцій у доношених новонароджених.

Ключові слова: доношені новонароджені, бактеріальна інфекція, неонатальний сепсис, неонатальна пневмонія, пресеписин, інтерлейкін-33, біомаркери, С-реактивний білок, прокальцитонін.

Neonatal bacterial infections remain a major challenge in modern neonatology despite significant advances in perinatal medicine and neonatal intensive care. According to recent international studies, sepsis and bacterial pneumonia continue to be among the leading causes of neonatal mortality and severe infectious morbidity. Moreover, bacterial infections in the neonatal period are associated with an increased risk of long-term neurological impairment and adverse systemic outcomes, highlighting their substantial impact on both short- and long-term health [2, 13, 15, 16].

The current concept of neonatal sepsis considers it not only as a consequence of bacterial invasion but also as a result of dysregulation of the innate immune response, leading to systemic inflammation, endothelial dysfunction, and subsequent multiple organ failure. The intensity of the immune-inflammatory response is considered a key determinant of disease severity and clinical outcome [1, 6].

One of the major challenges in clinical practice remains the early diagnosis of bacterial infections in neonates. The initial clinical manifestations of sepsis

and pneumonia are often nonspecific and may mimic a variety of other neonatal conditions, including respiratory distress, transient adaptation disorders, and metabolic disturbances. At the same time, blood culture, which remains the reference method for laboratory confirmation of infection, does not allow timely clinical decision-making because of its prolonged turnaround time and limited sensitivity. Consequently, the identification of reliable biomarkers capable of detecting infection at an early stage and objectively assessing the severity of the systemic inflammatory response has become one of the most rapidly evolving areas of modern neonatology. Particular interest has focused on molecules reflecting different components of the innate immune response and demonstrating high diagnostic sensitivity during the first hours following the onset of infection [4 – 6].

Among these biomarkers, presepsin (sCD14-ST) has attracted particular attention. Presepsin is a soluble subtype of the CD14 receptor that is released upon activation of monocytes and macrophages in response to bacterial infection. According to recent prospective studies and meta-analyses, presepsin

demonstrates high diagnostic accuracy for neonatal sepsis and may provide important additional diagnostic information compared with conventional inflammatory markers such as CRP and procalcitonin [3, 10, 11].

Another promising area of research is the investigation of interleukin-33 (IL-33), a member of the IL-1 cytokine family that functions as an alarmin. The release of IL-33 following tissue injury promotes activation of the innate immune response and contributes to the maintenance of inflammatory processes. Available evidence suggests that IL-33 may have diagnostic value in neonatal sepsis; however, the number of neonatal studies remains limited [7, 14].

Despite the growing body of research on inflammatory biomarkers, data regarding the combined diagnostic value of presepsin and IL-33 in bacterial infections among term neonates remain limited. Evaluation of their integrated use may improve the accuracy of early diagnosis, facilitate timely risk stratification, and contribute to the refinement of clinical management strategies for neonates with suspected infection [3, 7, 10, 14].

Therefore, investigating the diagnostic value of presepsin and IL-33 in bacterial infections in term neonates appears to be a promising area of research. Such an approach may enhance the accuracy of early diagnosis and optimize the clinical management of newborns with suspected infectious diseases.

The purpose of the study was to evaluate the diagnostic value of presepsin and interleukin-33 in term neonates with bacterial infections and to compare their diagnostic performance with conventional inflammatory biomarkers.

Materials and methods. A prospective study with consecutive enrollment included 36 term neonates admitted between 2021 and May 2024 to neonatal intensive care units and departments of neonatal pathology (Educational Surgical Clinic of Azerbaijan Medical University, Baku, Azerbaijan; Scientific Research Institute of Pediatrics, Baku, Azerbaijan). The study population comprised infants born in maternity hospitals across various cities and regions of Azerbaijan who developed critical conditions during the neonatal period and required transfer to the Neonatal Intensive Care Unit of the Scientific Research Institute of Pediatrics. All enrolled term neonates were divided into two groups: a study group and a control group. The study group included 23 term neonates aged 0 – 28 days with bacterial infections, including neonatal pneumonia and sepsis. Based on comprehensive clinical and laboratory evaluation and etiological assessment, neonatal pneumonia was diagnosed in 12 (52.2 %) infants, while sepsis was diagnosed in 11 (47.8 %) infants.

The control group consisted of 13 healthy term neonates without bacterial infections or congenital malformations. These infants were observed at the

Department of Neonatology of the Scientific Research Institute of Obstetrics and Gynecology. The control group did not differ significantly from the study group with respect to age and sex distribution.

Neonates with a diagnosis of hemolytic disease of the newborn, chromosomal abnormalities, metabolic diseases, multiple congenital malformations, and TORCH infections specific to the perinatal period were excluded from the study.

The average body weight of neonates in the study group was 3162.0 ± 112.3 g. Among neonates with infection, there were 14 (60.9 %) boys and 9 (39.1 %) girls; the control group included 9 boys (69.2 %) and 4 girls (30.8 %). All patients underwent clinical, laboratory, and instrumental examinations, which included cardiorespiratory monitoring, temperature measurement, complete blood count and biochemical blood tests, analysis of blood gas composition and acid-base status, and evaluation of CRP, procalcitonin, presepsin, and IL-33 concentrations. Neurosonography, ultrasonography, echocardiography, and bacteriological culture of samples from various sites were also performed. The nSOFA and NEOMOD scores were used to diagnose organ dysfunction. Quantitative determination of serum presepsin and IL-33 was performed using a sandwich enzyme-linked immunosorbent assay (ELISA) with a commercial Sunred Bio (Shanghai) kit produced by Algen Diagnostik Medical LTD (Ankara, Turkey) on a BioScreen MS- 500 analyzer (Stat Fax, USA).

The study was conducted in accordance with the ethical principles of the Declaration of Helsinki of the World Medical Association, the UNESCO Universal Declaration on Bioethics and Human Rights, and internationally accepted principles of biomedical research involving human participants. Participation in the study did not interfere with the standard diagnostic and therapeutic management of the newborns. Written informed consent for participation in the study and the use of clinical and laboratory data for scientific purposes was obtained from the parents or legal representatives of all neonates before enrollment. All collected data were anonymized and processed confidentially.

Statistical analysis was performed using SPSS 27. Data were presented as median (Me) and quartile values (Q1; Q3), corresponding to the 25th and 75th percentiles. The significance of differences between the compared groups was assessed using the non-parametric Mann – Whitney U test. Differences were considered statistically significant at $p < 0.05$ [12].

Results of the study. The study revealed that most newborns were born to mothers with extragenital pathology and a complicated obstetric and gynecological history. The examined newborns were predominantly born from repeated pregnancies and deliveries: 14 (60.9 %) in the study group and 9 (69.2 %) in the control group. In most cases, delivery in the study group was performed by cesarean section

in 17 (73.9 %) cases, whereas vaginal delivery predominated in the control group, accounting for 8 (61.5 %) cases ($p < 0.05$). Among newborns with infection, the 1-minute Apgar score was 6.0 ± 0.3 (range 4 – 8) points. Mild asphyxia was diagnosed in 12 (52.2 %) newborns, while moderate asphyxia was observed in 11 (47.8 %). The 5-minute Apgar score was 7.1 ± 0.2 (range 6 – 9) points. At 5 minutes, 19 (82.6 %) newborns were classified as having mild asphyxia, whereas 4 (17.4 %) had moderate asphyxia.

Upon admission, 11 (47.8 %) newborns were in severe condition and 12 (52.2 %) were in very severe condition. Resuscitation measures were performed in all newborns included in the study group. The mortality rate was 13.0 % (3 patients).

The observed newborns presented with respiratory failure in 19 (82.6 %) cases, congenital pneumonia in 11 (47.8 %), perinatal infections in 3 (13.0 %), necrotizing enterocolitis in 3 (13.0 %), hepatitis in 1 (4.3 %), peritonitis in 1 (4.3 %), cardiovascular insufficiency in 2 (8.7 %), disseminated intravascular coagulation (DIC) syndrome in 1 (4.3 %), pneumothorax in 3 (13.0 %), intracranial hemorrhage in 1 (4.3 %), neonatal seizures in 2 (8.7 %), and osteomyelitis in 1 (4.3 %) case.

Bacteriological examination was performed in 20 of the 23 preterm newborns included in the study group. Positive culture results were obtained in 15 (75.0 %) newborns, whereas no microbial growth was detected in 5 (25.0 %) newborns. The most

frequently identified pathogens were coagulase-negative Gram-positive staphylococci, which were isolated in 7 (46.7 %) cases. The isolation rates of other microorganisms were as follows: *Staphylococcus aureus* – 1 (6.7 %), *Klebsiella pneumoniae* – 2 (13.3 %), *Candida albicans* – 1 (6.7 %), *Enterobacter aerogenes* – 1 (6.7 %), *Pseudomonas* spp. – 1 (6.7 %), and *Moraxella* spp. – 1 (6.7 %). In several cases, mixed microbial growth was observed. In the majority of cases, microorganisms were isolated from blood cultures. Thus, among newborns who underwent bacteriological examination, an infectious pathogen was identified in 75.0 % of cases, most commonly from blood and typically from a single anatomical site upon admission to the NICU.

Comparative analysis of hematological parameters demonstrated that bacterial infection in term newborns was associated with a statistically significant decrease in hemoglobin concentration, red blood cell count, and hematocrit, accompanied by an increase in red blood cell distribution width (RDW-SD) and mean corpuscular hemoglobin (MCH). Changes in the leukocyte profile were characterized by increased absolute counts of neutrophils, monocytes, eosinophils, and basophils, whereas total leukocyte and lymphocyte counts did not differ significantly from those observed in the control group. No significant differences were identified in platelet indices between the groups (Table 1).

Table 1

Hematological Parameters of Term Newborns with Bacterial Infection and Healthy Control Newborns

	Group 1								Pu
	Control				Study				
	M	Me	Q1	Q3	M	Me	Q1	Q3	
HGB	17.1	17.0	16.0	18.5	13.8	14.4	12.2	15.8	<0.001*
RBC	4.81	4.90	4.10	5.10	3.88	4.20	3.80	4.41	<0.002*
WBC	14.4	14.6	12.0	18.4	12.2	11.4	7.1	15.6	0.311
HCT	51.3	51.5	47.3	55.2	42.0	43.3	37.5	47.3	<0.001*
MCV	96.1	98.0	87.0	99.0	105.3	106.0	99.2	110.0	0.089
MCH	33.4	34.0	32.0	35.0	34.7	34.6	33.3	35.9	0.001*
MCHC	32.5	32.0	31.2	33.0	33.1	33.3	32.0	34.6	0.352
RDWSD	40.2	39.2	38.3	43.0	53.5	54.0	49.5	58.0	<0.001*
RDWCV	17.2	17.0	16.0	18.8	16.8	16.0	15.0	18.8	0.548
PLT	239.9	223.0	213.0	265.0	227.5	229.0	109.0	309.0	0.709
PCT	0.40	0.40	0.30	0.50	0.29	0.20	0.15	0.3	0.233
BAS	0.81	0.70	0.60	0.90	1.39	1.10	0.90	2.0	0.011
EOS	0.75	0.70	0.54	1.10	1.46	1.40	0.90	2.0	<0.001*
NEUT	3.14	1.80	1.60	4.70	6.13	4.90	3.06	9.7	<0.016*
MON	0.38	0.10	0.08	0.50	1.43	1.23	0.45	1.9	<0.044*
LYM	3.29	3.00	1.70	4.80	6.40	4.10	2.75	7.1	0.106

Note: M – mean; Me – median; Q1, Q3 – quartiles; p – statistical significance between the values of newborns in the study and control groups. P_U – statistical accuracy of the Mann – Whitney U test; “*” the null hypothesis is rejected.

ROC curve analysis demonstrated that RDW-SD had the highest discriminatory ability for identifying bacterial infection in term newborns (area under the ROC curve (AUC)=0.983, $p < 0.001$). High diagnostic performance was also observed for EOS (AUC=0.839), MCV (AUC=0.801), NEYT

(AUC=0.799), MON (AUC=0.786), and BAS (AUC=0.781), all of which showed statistically significant areas under the ROC curve ($p < 0.01$).

In contrast, MCH, MCHC, LYM, RDW-CV, PLT, and total leukocyte count did not demonstrate statistically significant discriminatory ability

($p > 0.05$). HGB, RBC, HCT, and PCT showed statistically significant inverse associations with bacterial infection, as reflected by AUC values below 0.5 ($p < 0.05$), indicating lower levels of these hematological parameters in infected newborns. Although WBC also yielded an AUC below 0.5 (AUC=0.348), its discriminatory ability was not statistically significant ($p = 0.134$) (Fig. 1).

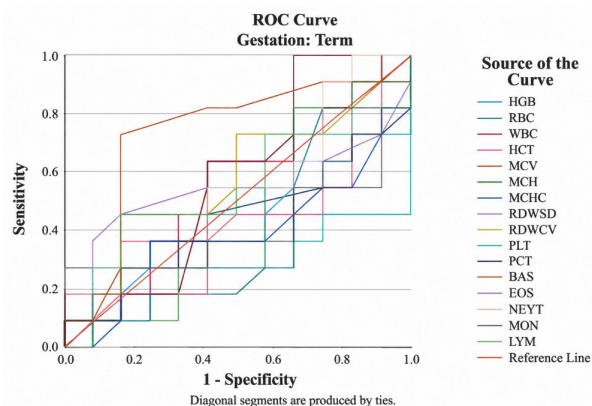


Fig. 1. ROC curves reflecting the hematological parameters of term infants with bacterial infection.

ROC analysis demonstrated limited diagnostic performance of the investigated hematological parameters. The AUC values ranged from 0.265 to 0.735. The highest AUC was observed for basophils (AUC=0.735; 95 % CI: 0.513 – 0.956); however, statistical significance was not achieved ($p = 0.056$). Similarly, the platelet count showed an AUC of 0.265 (95 % CI: 0.044 – 0.486; $p = 0.056$), indicating an inverse association with the studied outcome. For the remaining parameters, AUC values ranged from 0.371 to 0.576, with no statistically significant differences detected ($p > 0.05$), suggesting limited diagnostic value. The obtained results are consistent with current concepts regarding the diagnosis of neonatal bacterial infections. Despite statistically significant changes in certain complete blood count

parameters, most of them demonstrated relatively low diagnostic performance. This may be explained by the pronounced physiological variability of hematological parameters during the early neonatal period and the nonspecific nature of inflammatory changes.

Particular interest in our study was drawn to RDW-SD, which demonstrated the highest area under the ROC curve among all hematological parameters (AUC=0.983). This finding may reflect the impact of systemic inflammation on erythropoiesis, oxidative stress, and microcirculatory disturbances accompanying bacterial infection, resulting in increased anisocytosis. Nevertheless, despite its excellent discriminatory ability, RDW-SD cannot replace specific inflammatory biomarkers, as elevated values may also be observed in other critical conditions affecting newborns.

Comparative analysis of inflammatory biomarkers revealed significantly higher levels of all investigated markers in term newborns with bacterial infection compared with the control group. The most pronounced differences were observed for presepsin and IL-33, whose concentrations were increased by more than fivefold and approximately sevenfold, respectively ($p < 0.001$). Significant elevations were also found for CRP ($p = 0.003$), procalcitonin ($p = 0.015$), and lactate ($p < 0.001$). These findings indicate activation of the systemic inflammatory response and the development of tissue hypoperfusion in newborns with bacterial infection.

Our results further support the important role of innate immunity biomarkers in the early diagnosis of neonatal infection. The most substantial increases were observed for presepsin and IL-33, suggesting marked activation of the monocyte – macrophage immune pathway during the early stages of the infectious process (Table 2).

Table 2

Levels of inflammatory biomarkers in term infants with bacterial infection

	Group								P _u
	Control				Study				
	M	Me	Q1	Q3	M	Me	Q1	Q3	
CRP mg/L	2.8	2.8	2.2	3.2	44.3	29.0	7.6	70.0	0.003*
Procalcitonin ng/mL	0.4	0.4	0.3	0.5	11.8	5.6	0.6	18.5	0.015*
Presepsin pg/mL	111.2	104.1	96.3	121.0	537.2	535.0	315.0	738.0	<0.001*
IL-33 pg/mL	4.3	4.3	3.6	4.9	32.0	28.5	21.3	32.8	<0.001*
Lactate mmol/L	1.28	1.20	1.10	1.50	3.68	3.50	2.50	4.40	<0.001*

Note: M – mean; Me – median; Q1, Q3 – quartiles; p – statistical significance between the newborns in the study and control groups. P_u – statistical accuracy of the Mann – Whitney U test; “*” the null hypothesis is rejected.

ROC curves reflecting the levels of proinflammatory markers in preterm newborns with bacterial infection showed that the investigated inflammatory biomarkers demonstrated high diagnostic value for bacterial infection in term newborns. The highest diagnostic performance was observed for CRP and presepsin (AUC=0.971 for both markers), indicating an almost perfect ability to

discriminate between infected and healthy newborns. Similarly high diagnostic accuracy was demonstrated by lactate (AUC=0.965) and IL-33 (AUC=0.961), confirming their substantial value in the diagnosis of neonatal bacterial infection. Procalcitonin showed a lower, although still statistically significant, diagnostic performance (AUC=0.789, $p = 0.010$). Thus, the ROC analysis identified CRP, presepsin, IL-

33, and lactate as the most informative laboratory markers of bacterial infection in term newborns.

The diagnostic performance of the investigated biomarkers varied considerably. Presepsin and IL-33 demonstrated the highest diagnostic accuracy, with a sensitivity of $94.1 \pm 5.7\%$ and specificity of 100.0% for both biomarkers. Their overall diagnostic value (GDV) was $96.6 \pm 3.4\%$, while the positive predictive value (PPV) reached 100.0% and the negative predictive value (NPV) was $92.4 \pm 7.4\%$. The optimal cut-off values were >181.8 for presepsin and >9.26 for IL-33. Both positive and negative likelihood ratios were classified as excellent. CRP also showed high diagnostic performance at a cut-off value of >4.7 , with a sensitivity of $91.3 \pm 5.9\%$ and specificity of 100.0% . Its GDV was $94.3 \pm 3.9\%$, with a PPV of 100.0% and an NPV of $85.7 \pm 9.4\%$. The positive and negative likelihood ratios were both rated as excellent. Procalcitonin, at a cut-off value of >1.42 , demonstrated lower sensitivity ($68.8 \pm 11.6\%$) compared with the other biomarkers, although its specificity and PPV remained 100.0% . The GDV was $82.1 \pm 7.2\%$, and the NPV was $70.6 \pm 11.1\%$. The positive likelihood ratio was classified as excellent, whereas the negative likelihood ratio showed only moderate diagnostic performance. Lactate, with a cut-off value of >2.05 , showed a sensitivity of $87.0 \pm 7.4\%$ and a specificity of $92.3 \pm 7.4\%$. Its GDV was $88.9 \pm 5.2\%$, with a PPV of $98.7 \pm 1.3\%$ and an NPV of $80.0 \pm 10.3\%$. The positive likelihood ratio was classified as excellent and the negative likelihood ratio as good. Overall, presepsin and IL-33 demonstrated the most favorable combination of sensitivity, specificity, predictive values, and likelihood ratios for identifying bacterial infection in term infants.

Evaluation of the diagnostic performance of the investigated biomarkers demonstrated that presepsin and IL-33 were the most informative markers for the detection of bacterial infection in term newborns. Both biomarkers showed the highest sensitivity, diagnostic accuracy, and negative predictive value, while maintaining absolute specificity and a positive predictive value of 100% . CRP exhibited similarly high specificity and positive predictive value; however, its sensitivity was slightly lower than that of presepsin and IL-33. Lactate also demonstrated high diagnostic performance, although its sensitivity, specificity, and negative predictive value were somewhat lower. Among the investigated biomarkers, procalcitonin showed the lowest diagnostic informativeness despite maintaining high specificity and positive predictive value. Analysis of likelihood ratios indicated that a positive test result had excellent diagnostic value for most of the studied biomarkers, whereas a negative test result was most informative for CRP, presepsin, and IL-33. Odds ratio analysis revealed a statistically significant association between elevated levels of all investigated

biomarkers and the presence of bacterial infection in term newborns.

Results of the Fisher – Snedecor criterion (F-test) in term infants with bacterial infection. Analysis of the association between biomarker levels and bacterial infection demonstrated statistically significant results for all investigated parameters. CRP at a cut-off value of >4.7 was associated with markedly increased odds of bacterial infection ($OR=215.0$; $95\% \text{ CI: } 9.5 - 4846.4$; $p=0.002$). Procalcitonin >1.42 was also significantly associated with bacterial infection, although the magnitude of the association was lower than that observed for the other biomarkers ($OR=52.3$; $95\% \text{ CI: } 2.6 - 1053.9$; $p=0.016$). The strongest associations were observed for presepsin and IL-33. Presepsin >182 was associated with an OR of 275.0 ($95\% \text{ CI: } 10.3 - 7336.9$; $p=0.002$), while IL-33 >9.26 demonstrated an identical OR of 275.0 ($95\% \text{ CI: } 10.3 - 7336.9$; $p=0.002$). Lactate levels >2.05 were also significantly associated with bacterial infection ($OR=80.0$; $95\% \text{ CI: } 7.5 - 858.9$; $p=0.001$).

Thus, all five biomarkers showed statistically significant associations with bacterial infection. Among them, presepsin and IL-33 demonstrated the highest odds ratios, followed by CRP and lactate, whereas procalcitonin showed the lowest OR. However, the wide 95% confidence intervals observed for all biomarkers indicate considerable uncertainty in the magnitude of these effect estimates and should be taken into account when interpreting the results. These findings are consistent with current clinical recommendations, according to which the combined use of several biomarkers can substantially improve the accuracy of early sepsis diagnosis and reduce unnecessary antibiotic administration.

Spearman correlation analysis demonstrated predominantly positive and statistically significant relationships between the investigated proinflammatory markers in term infants with bacterial infection. The strongest correlation was observed between presepsin and IL-33 ($\rho=0.843$; $p<0.001$), indicating a close relationship between these two biomarkers. Strong positive correlations were also found between IL-33 and lactate ($\rho=0.732$; $p<0.001$), as well as between CRP and lactate ($\rho=0.692$; $p<0.001$). Presepsin was significantly correlated with lactate ($\rho=0.647$; $p<0.001$) and CRP ($\rho=0.541$; $p=0.002$). CRP showed significant positive correlations with IL-33 ($\rho=0.580$; $p=0.001$) and procalcitonin ($\rho=0.498$; $p=0.007$). Procalcitonin was also significantly correlated with lactate ($\rho=0.552$; $p=0.002$). In contrast, correlations of procalcitonin with presepsin ($\rho=0.217$; $p=0.321$) and IL-33 ($\rho=0.171$; $p=0.435$) were weak and did not reach statistical significance.

Thus, the obtained results indicate a coordinated change in the majority of the investigated biomarkers during bacterial infection, with presepsin and IL-33 showing the strongest association with each other.

The strongest correlation was observed between presepsin and IL-33, suggesting synchronous activation of different components of the innate immune response. The strong correlations of these biomarkers with CRP and lactate reflect a close relationship between the severity of the inflammatory response and the development of tissue hypoperfusion. These findings support the usefulness of a comprehensive assessment of multiple biomarkers rather than relying on a single laboratory parameter for the diagnosis of bacterial infection in term newborns.

Discussion. Early diagnosis of bacterial infection in newborns remains challenging because of nonspecific clinical manifestations and the limited diagnostic performance of conventional laboratory tests. Although CRP and procalcitonin are widely used, increasing attention is being directed toward biomarkers reflecting activation of innate immunity, particularly presepsin and pro-inflammatory cytokines [4–6].

In the present study, bacterial infection in term newborns was associated with decreased hemoglobin, erythrocyte count, and hematocrit, together with increased RDW-SD and absolute counts of neutrophils, monocytes, eosinophils, and basophils. However, ROC analysis showed that most hematological parameters had limited independent diagnostic value. RDW-SD was a notable exception, demonstrating the highest discriminatory performance among the hematological indices (AUC=0.983). This finding may reflect inflammation-associated disturbances in erythropoiesis, oxidative stress, and changes in erythrocyte morphology. Nevertheless, RDW-SD remains a nonspecific marker and should be interpreted in combination with other laboratory parameters. This interpretation is consistent with Eichberger et al. (2022), who emphasized the limitations of conventional inflammatory markers in neonatal sepsis, and with Gopal et al. (2023), who highlighted the advantages of combining conventional laboratory indices with emerging biomarkers [4, 5].

The most clinically relevant findings concerned presepsin and IL-33. Both biomarkers were markedly elevated in term newborns with bacterial infection and demonstrated excellent diagnostic performance. Presepsin yielded an AUC of 0.971, with 94.1 % sensitivity and 100 % specificity. These results are consistent with Hincu et al. (2025), who demonstrated the usefulness of presepsin in the early diagnosis of neonatal sepsis [8]. Similarly, Dierikx et al. (2025) reported high diagnostic accuracy of presepsin for late-onset neonatal sepsis in a multicenter prospective cohort [3]. Pospisilova et al. (2023) also demonstrated the diagnostic potential of presepsin in newborns at risk of early-onset sepsis [11]. Thus, despite differences in study populations and timing of sepsis onset, these studies and our findings consistently

support presepsin as a valuable neonatal infection biomarker.

Our findings are also in agreement with the meta-analysis by Peng et al. (2025), which demonstrated substantial diagnostic value of presepsin across different age groups [10]. Importantly, in our cohort presepsin outperformed procalcitonin: the sensitivity of presepsin was 94.1 %, compared with 68.8 % for procalcitonin, while presepsin maintained 100 % specificity. This difference supports the potential value of presepsin as an adjunct to conventional biomarkers rather than relying on procalcitonin alone.

IL-33 demonstrated similarly strong diagnostic characteristics, with an AUC of 0.961, sensitivity of 94.1 %, and specificity of 100 %. Although the evidence base for IL-33 in neonatal infection is considerably smaller than that for presepsin, our findings are consistent with Halil et al. (2018), who investigated serum IL-33 as a biomarker of neonatal sepsis in premature infants [7], and Salem et al. (2024), who also reported its diagnostic value in neonatal sepsis [14]. Our study extends these observations specifically to term neonates, suggesting that the diagnostic potential of IL-33 may not be restricted to premature populations.

CRP also showed excellent diagnostic performance (AUC=0.971) and, notably, an AUC identical to that of presepsin in our cohort. Lactate and IL-33 showed similarly high values (AUC=0.965 and 0.961, respectively), whereas procalcitonin demonstrated substantially lower discrimination (AUC=0.789). Thus, our findings do not support the superiority of a single marker in all respects; rather, they suggest that presepsin and IL-33 may complement CRP and lactate while providing additional information on the innate immune response. This interpretation is consistent with contemporary reviews emphasizing multimarker rather than single-marker strategies for neonatal sepsis diagnosis [4–6].

Another important finding was the strong correlation between presepsin and IL-33 ($\rho=0.843$; $p<0.001$). IL-33 was also strongly correlated with lactate ($\rho=0.732$; $p<0.001$), while presepsin correlated with lactate ($\rho=0.647$; $p<0.001$) and CRP ($\rho=0.541$; $p=0.002$). In contrast, procalcitonin showed weak and nonsignificant correlations with presepsin and IL-33. These differences may indicate that the investigated biomarkers reflect partially distinct but interconnected components of the systemic response to bacterial infection. The simultaneous assessment of markers representing different inflammatory pathways may therefore provide a more comprehensive characterization of neonatal infection than any single biomarker alone.

The present study has several limitations. It was conducted at a single center and included a relatively small number of term newborns, resulting in wide

confidence intervals for some diagnostic estimates and limiting generalizability. Biomarkers were measured only at admission, without serial assessment during treatment, and their diagnostic performance was not stratified according to infection etiology or disease severity. Therefore, larger

multicenter prospective studies are required to validate the identified cut-off values, assess temporal changes in presepsin and IL-33, and determine whether their combined measurement provides clinically meaningful advantages over established biomarkers.

Conclusion

Bacterial infections in term newborns are associated with pronounced activation of the innate immune response, as evidenced by significant increases in presepsin, IL-33, C-reactive protein, procalcitonin, and lactate concentrations. Presepsin and IL-33 demonstrated the highest diagnostic performance, exhibiting high sensitivity, absolute specificity, and the strongest association with the infectious process. The combined assessment of presepsin and IL-33 together with conventional laboratory markers may improve the accuracy of early diagnosis of bacterial infections in term newborns, enhance risk stratification, and facilitate the timely initiation of appropriate antimicrobial therapy.

Prospects for further research. The present findings expand current knowledge regarding the role of innate immune biomarkers in the diagnosis of neonatal sepsis and pneumonia and support their potential incorporation into routine laboratory diagnostic algorithms for newborns.

References

1. Alansari AN, Zaazouee MS, Elshanbary AA, Mani S, Messaoud M. Diagnostic accuracy of interleukin-6 (IL-6) as a significant biomarker in late-onset neonatal sepsis: an updated systematic review and meta-analysis. *Eur J Pediatr.* 2025;184(9):587. doi:10.1007/s00431-025-06409-w.
2. Cantey JB, Edwards MS. Clinical features, evaluation, and diagnosis of sepsis in term and late preterm neonates. In: Post TW, editor. *UpToDate*. Waltham (MA): UpToDate Inc.; 2022. Accessed January 31, 2023.
3. Dierikx TH, Admiraal J, Nusman CM, van Laerhoven H, van der Schoor SRD, de Meij TGI, et al. The diagnostic accuracy of presepsin for late-onset neonatal sepsis: a multicenter prospective cohort study. *Pediatr Res.* 2025;98(5):1864-1869. doi:10.1038/s41390-025-04008-x.
4. Eichberger J, Resch E, Resch B. Diagnosis of neonatal sepsis: the role of inflammatory markers. *Front Pediatr.* 2022;10:840288. doi:10.3389/fped.2022.840288.
5. Gopal N, Chauhan N, Jain U, Dass SK, Sharma HS, Chandra R. Advancement in biomarker-based effective diagnosis of neonatal sepsis. *Artif Cells Nanomed Biotechnol.* 2023;51(1):476-490. doi:10.1080/21691401.2023.2252016.
6. Gude SS, Peddi NC, Vuppapapati S, Gopal SV, Ramesh HM, Gude SS. Biomarkers of neonatal sepsis: from being mere numbers to becoming guiding diagnostics. *Cureus.* 2022;14(3):e23215. doi:10.7759/cureus.23215.
7. Halil H, Tayman C, Buyuktiryaki M, Okur N, Cakir U, Serkant U. Serum interleukin-33 as a biomarker in predicting neonatal sepsis in premature infants. *Comb Chem High Throughput Screen.* 2018;21(7):510-515. doi:10.2174/1386207321666180911090656.
8. Hincu MA, Gheorghe L, Dimitriu C, Paduraru L, Zonda G, Andronic DC, et al. Procalcitonin, Presepsin, Endocan, and Interleukin-6 in the Early Diagnosis of Neonatal Sepsis-A Prospective Study. *Diagnostics (Basel).* 2025 May 26;15(11):1341. doi: 10.3390/diagnostics15111341.
9. Lee CY, Zven SE, Sathya SA, Abukhalaf D, Sahoo S, Samal P, et al. Using molecular methods to diagnose, classify, and treat neonatal sepsis: a scoping review. *Front Pediatr.* 2025;13:1625449. doi:10.3389/fped.2025.1625449.
10. Peng K, Zhang X, Li Q, Luo Z. Presepsin as a diagnostic biomarker for sepsis across neonates, children, and adults: a meta-analysis. *Biomol Biomed.* 2025;26(2):307-319. doi:10.17305/bb.2025.12909.
11. Pospisilova I, Brodska HL, Bloomfield M, Borecka K, Janota J. Evaluation of presepsin as a diagnostic tool in newborns with risk of early-onset neonatal sepsis. *Front Pediatr.* 2023;10:1019825. doi:10.3389/fped.2022.1019825.
12. Qafarov IA. *Biostatistics*. Baku: Təbib Publishing House; 2025. 250 p. ISBN: 978-9952-39-330-9.
13. Rahimova NJ. Sepsis-associated encephalopathy in newborns: clinical and immunological correlations. *Journal of Azerbaijan Perinatology and Pediatrics.* 2025;11(1):9-17.
14. Salem AMS, Farag DS, Ahmed ME, Botrous OE. Diagnostic value of interleukin-33 in neonatal sepsis. *Egyptian Journal of Medical Research.* 2024;5(1):133-142.
15. Singh M, Agarwal A, Pattnaik P, Alsaleem M. Neonatal sepsis. In: *StatPearls* [Internet]. Treasure Island (FL): StatPearls Publishing; 2026. PMID: 30285373.
16. Stocker M, Rosa-Mangeret F, Agyeman PKA, McDougall J, Berger C, Giannoni E. Management of neonates at risk of early onset sepsis: a probability-based approach and recent literature appraisal: update of the Swiss national guideline of the Swiss Society of Neonatology and the Pediatric Infectious Disease Group Switzerland. *Eur J Pediatr.* 2024;183(12):5517-5529. doi:10.1007/s00431-024-05811-0.

Conflict of interest. The authors have no conflicts of interest to declare.

ORCID: Huseynova N.F. <https://orcid.org/0009-0008-0181-9110>.

Article received: 14.08.2025