Chinese Journal of Evidence-Based Medicine

DIAGNOSTIC UTILITY OF INTERLEUKIN-6, INTERLEUKIN- 8 AND C-REACTIVE PROTEIN FOR EARLY DETECTION OF NEONATAL SEPSIS AT A TERTIARY CARE HOSPITAL

Dr. Harish. N [1], Dr. R.Vidhya [2*], Dr. S. Sundari [3]

[1] JUNIOR RESIDENT, SREE BALAJI MEDICAL COLLEGE AND HOSPITAL, CHROMPET, CHENNAI.

[2] ASSISTANT PROFESSOR, DEPARTMENT OF MICROBIOLOGY, KARPAGA VINAYAGA INSTITUTE OF MEDICAL SCIENCES AND RESEARCH CENTRE, CHENNAI.

[3] PROFESSOR AND HEAD OF THE DEPARTMENT, PAEDIATRICS, SREE BALAJI MEDICAL COLLEGE AND HOSPITAL, CHROMPET, CHENNAI.

DOI: https://doie.org/10.0729/Cjebm.2024188633

Abstract:

Introduction and aim: Neonatal sepsis is a clinical illness that occurs in the first 28 days of life. Non-specific clinical appearance makes the early diagnosis difficult. C-reactive protein (CRP), Interleukin-6 and 8 (IL-6 & 8) have been of great interest in detecting the disease in recent days. So this study aims to determine the cut-off values for IL-6 & 8 and to investigate the diagnostic accuracy of the same.

Materials and Methods: The case-control study included 280 neonates, 140 were clinically suspected cases of sepsis and 140 were healthy neonates. Immunoassay Kits were used to determine the levels of IL 6 & 8 and CRP in the blood.

Results: All three given biomarkers were found to be extremely significant between the cases and control population. CRP levels were found to be statistically significant between the early-onset sepsis group and the late-onset sepsis group. CRP findings in EOS can be crucial. So in EOS samples, it would be beneficial if CRP results were interpreted with other biomarkers like IL 6 and IL 8.

Conclusion: We found a considerable increase in IL 6 and IL 8 levels, which could be used as a particular biomarker for detecting neonatal sepsis. The combination of these immunological markers could thus be critical for the diagnosis, as well as better indicators of the disease pathophysiology.

Keywords: Neonatal sepsis, C-reactive protein, Interleukin-6 & 8, Immunoassay and biomarker.

Introduction:

Neonatal sepsis is a severe illness that occurs in the initial 28 days after birth and it is one of the leading causes of death around the world[1]. Invasive procedures along with immunological immaturity are two risk factors linked to its higher prevalence[2]. Depending on the time of infection it is divided into two types: early onset (< 72 hours of childbirth) and late onset sepsis (> 72 hours of childbirth)[3]. EOS is linked to vertical infection as a result of

maternal colonization. LOS is linked to infection acquired from the hospital or community[3]. Non-specific clinical appearance makes the early diagnosis difficult. The gold standard for microbiological diagnosis is blood culture, although it has low sensitivity and is a time-consuming procedure[4]. A missing diagnosis causes medication to be delayed and raises the chance of complications. The prognosis of newborn sepsis is determined by early detection and effective antibiotic treatment[5]. As a result, early and precise detection of sepsis is critical for improving clinical outcomes and lowering medical expenses[6].

Acute phase proteins, pro-inflammatory components like cytokines and chemokines, such as C-reactive protein (CRP), pro-calcitonin (PCT), Interleukin-6 (IL-6) and interleukin 8 (IL-8) have been of great interest to detect the progression of neonatal sepsis in recent days[7].

The liver produces CRP which participates in the acute phase reaction. It has a half-life of 24 to 48 hours. Increased CRP production can be caused by infections and non-infectious disorders[8]. The CRP level may be modest in the early stages of infection, but serial assessments can provide more valuable information. It is a useful marker to diagnose neonatal sepsis, however, it is unreliable since it takes a long time to respond to an infection[9].

IL-6 is an immune-regulatory cytokine that is found in the bloodstream during the initial stages of infection. The generation of CRP by the liver is aided by IL-6. The amount of IL-6 rises during the early stages of bacterial infections, which could help with the early detection of newborn sepsis[10]. Furthermore, Interleukin 6 (IL 6), generated by T and B cells, is more sensitive than CRP, its short half-life limits its utility as a solitary diagnostic of sepsis[11].

IL-8 is a pro-inflammatory cytokine that aids in the diagnosis and detection of neonatal sepsis severity. IL-8 is the only interleukin from the chemokine family. Within 2-4 hours following the initiation of an infection, the amount of IL-8 rises significantly[11].

In neonates, these immunological markers have been helpful in the timely identification of sepsis[12]. However investigations have indicated that both the markers have diagnostic significance, their cut-off values fluctuate and the effectiveness varies from study to study. The relationship between these biomarkers and the prognosis of neonatal sepsis is critical for early diagnosis[13].

A good diagnostic biomarker will have high sensitivity and specificity[14]. The aim of this study is to determine the best cut-off values for IL-6 & 8, as well as to calculate the diagnostic accuracy for the early detection of neonatal sepsis.

Materials & methods:

The prospective case-control study was done over two years and it included 280 neonates, 140 were clinically suspected cases of sepsis admitted to the NICU and 140 were healthy neonates. The patients and controls were both under the age of 28 days.

The study comprised preterm and term babies (<28 days old) of both sexes who showed symptoms of neonatal sepsis. Temperature instability, abdominal distension, food intolerance, tachycardia (HR>190bpmin), bradycardia (HR>90bpmin), dyspnea, tachypnea (>70/min), hepatosplenomegaly and irritability are the signs and symptoms of sepsis. The study excluded newborns born with congenital abnormalities, type 1 diabetes, or those who had experienced any surgical procedure. For both cases and controls, routine biochemical

parameters were measured. In both groups, either parent gave their informed consent. The Institution's Ethics Committee approved the study.

A standard proforma was constructed to collect patient's demographic data and information such as delivery history, symptoms, risk factors for infection, laboratory results, and clinical outcomes.

Clinical signs and symptoms are used to diagnose neonatal sepsis in the study setting, as well as laboratory findings such as complete blood count (CBC), CRP and or blood cultures.

***Measurements of Biomarkers*:**

Blood samples were received and serum was separated and stored at -20^{o}C till analysis.

***Human Interleukin 6 level*:**

A human Interleukin 6 ELISA kit was used for quantitative detection (Bioassay technology laboratory). The assay procedure was done according to kit instructions. Readings were measured at 450nm of wavelength. The coefficient of variation among intraassay and interassay had been approximately 8% and 10% respectively.

***Human Interleukin 8 level*:**

Human Interleukin 8 ELISA kit was used for quantitative detection (Bioassay technology laboratory). Kit instructions were followed throughout the assay. Readings were measured at 450nm of wavelength. The coefficient of variation among intra-assay and interassay had been approximately <8% and <10% respectively.

***Statistical analysis*:**

All variables were given as median (inter-quartile range). A statistically significant value was taken as $p < 0.05$. Graph Pad Prism software was applied for all statistical analysis (version 5).

Results:

***Classification of case participants*:**

Table 1: The table shows the classification of case participants

Characteristics	Total (%)
EOS	91 (65%)
LOS	49 (35%)
Number of Participants	140 (100%)

The prevalence of early-onset sepsis (65%) was found to be higher in the case population.

***Blood culture*:**

Blood culture showed no growth in 105 (75%) of neonates and growth in 35 (25%) of neonates. Thirty-five positive cultures were included, with 32 (91.4%) showing bacterial growth within 24 hours. The remaining 3 cultures became positive on day 2.

***Distribution of pathogens by Blood culture*:**

Table 2: The table shows the different aetiological agents in blood culture-positive sepsis.

S.no	Pathogen isolated	EOS	LOS	Total number	Percentage (%)
1	*Klebsiella pneumoniae*	10	7	17	48.6
2	*Acineto bacterbaumannii*	1	4	5	14.3
3	*Escherichia coli*	2	2	4	11.4
4	*CoNS*	1	2	3	8.6
5	*Pseudomonas aeruginosa*	1	1	2	5.7
6	*Staphylococcus aureus*	2	-	2	5.7
7	*Enterobacter aerogenes*	1	-	1	2.9
8	*Enterococcus fecalis*	1	-	1	2.9

The majority of the pathogens were Gram-negative organisms (82.9%). *Klebsiella pneumoniae* (48.6%) is the most common isolate followed by *Acinetobacter baumannii* complex as shown in **Table 2**.

Analysis of Biomarkers – CRP, IL6 and IL8

Table 3: Analysis of biomarkers between cases and control.

Biomarkers	Cases Median (IQR)	Control Median (IQR)	p-value
CRP	11 (4.1, 41.8)	3 (1.5, 4.4)	<0.001
IL 6	32.99 (24.98, 39.29)	9.58 (7, 12.47)	<0.001
IL 8	44.08 (37.46, 50.58)	13.58 (10.8, 18.9)	<0.001

All three given biomarkers were found to be extremely significant between the cases and the control population.

Table 4: Analysis of biomarkers between EOS and LOS

Biomarkers	EOS - Median (IQR)	LOS- Median (IQR)	p-value
CRP	6.6 (3.3, 17.2)	41.8 (16.3, 66.1)	<0.001
IL 6	32.66 (23.28, 38.78)	33.24 (25.89, 41.02)	0.122
IL 8	44.00459 (35.41, 48.71)	46.94 (37.60, 52.99)	0.106

When the CRP values were compared between the EOS and LOS cases, extremely significant statistical difference ($p<0.001$) was seen. The cut-off value for CRP was < 6mg/dl. It must be noted that the median CRP level in EOS was 6.6mg/dl. CRP findings in EOS can be crucial. Box Whisker plot showing the level of CRP, IL6 and IL8 in control, EOS and LOS groups are shown in figure 1-3.

Figure 1: Box Whisker plot showing the level of CRP in control, EOS and LOS groups

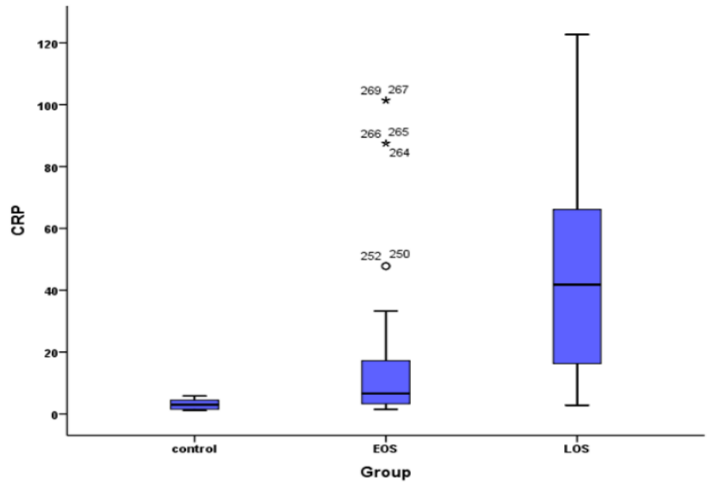


Figure 2: Box Whisker plot showing the level of IL-6 in control, EOS and LOS groups

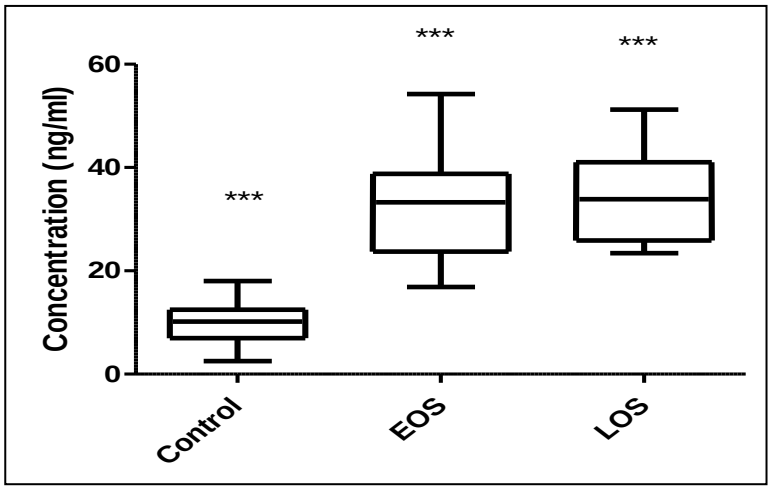
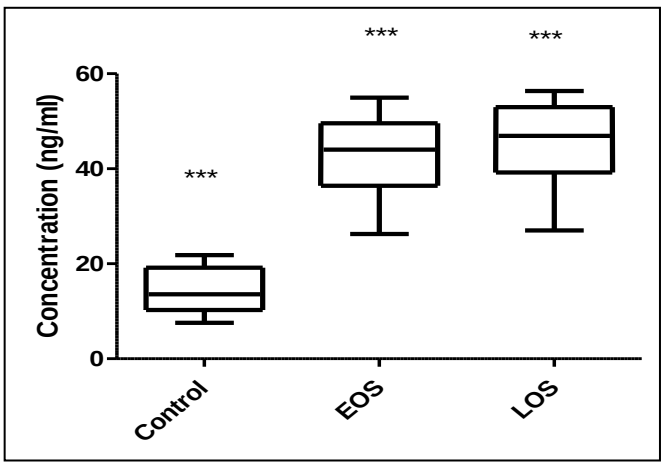


Figure 3: Box Whisker plot showing the level of IL-8 in control, EOS and LOS groups

If the p-value is less than 0.001 it is assigned three stars (***)

Receiver operating characteristic curve analysis for biomarkers

Figure 4: ROC for IL6



Vol. 20, No. 1. (2024)
E ISSN: 1672-2531

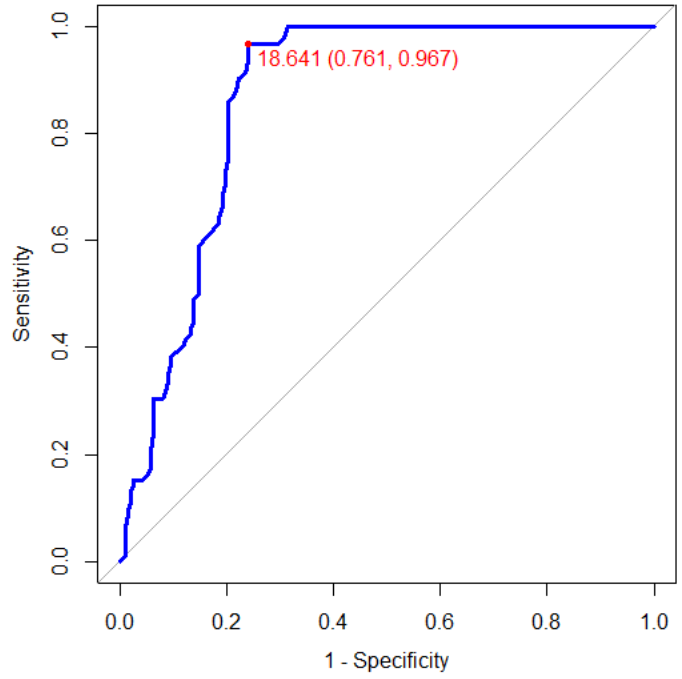
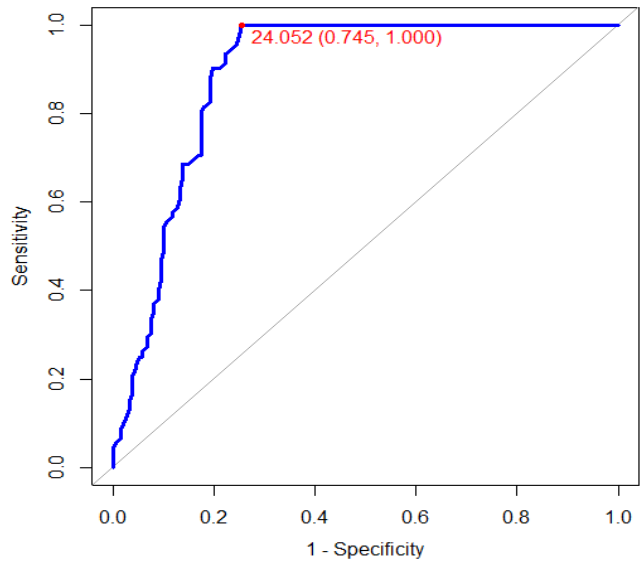


Figure 5: ROC for IL8



The area under the curve (AUC), sensitivity and specificity, and cut-off value for biomarkers like Il 6 and IL 8 were calculated (**Figure 4-5**) and given in **Table 5**. Both the biomarkers showed higher sensitivity and specificity.

Table 5: ROC analysis of IL 6 and IL 8

Biomarkers	Cut off value	Sensitivity	Specificity	Accuracy	PPV	NPV
IL 6	18.64 ng/l	96.73%	76.06%	0.828	0.664	0.979
IL 8	24.05 ng/l	100%	74.46%	0.828	0.657	1

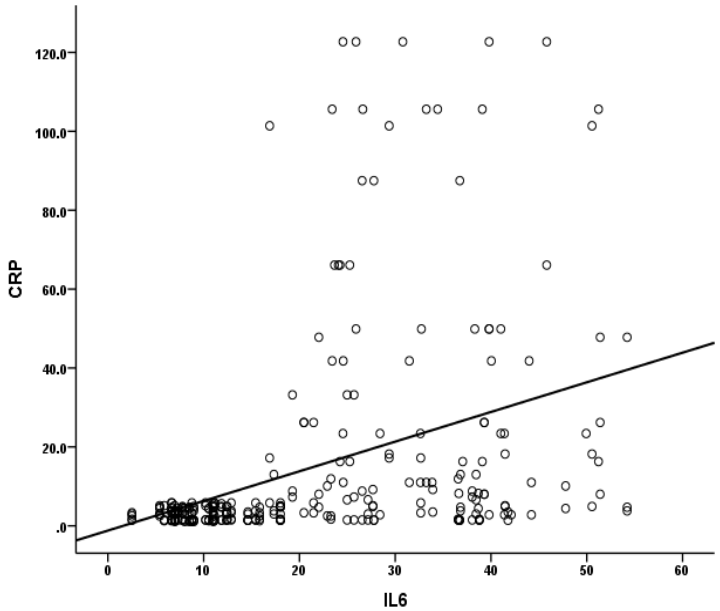
***Correlation between biomarkers*:**

- The correlation between serum concentration of IL 6 and IL 8 with CRP was carried out. The correlation values along with their significance level are given in **Table 6**.
- The scatter diagram shows the correlation for all the biomarkers like IL 6, IL8 and CRP. (Figure 6-8)
- IL 6 and IL 8 showed a strong positive correlation while CRP with IL 6 and IL 8 showed moderate positive correlation.

Table 6: Correlation of Biomarkers

	Spearman's rho (P value)	
	IL8	CRP
IL6	.790 (<0.001)	.506 (<0.001)
IL8		.511 (<0.001)

Figure 6: Scatter plot of IL 6 and CRP



Vol. 20, No. 1. (2024)
E ISSN: 1672-2531

Figure 7: Scatter plot of IL 8 and CRP

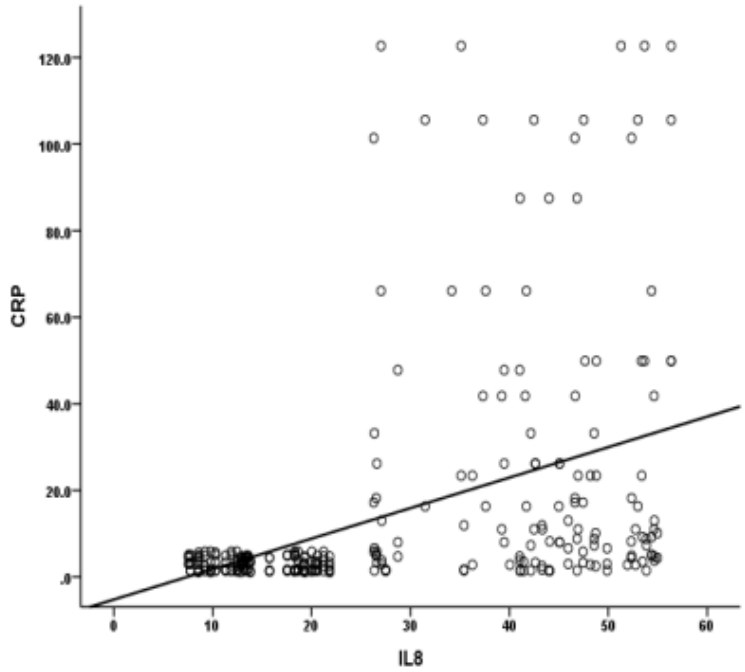
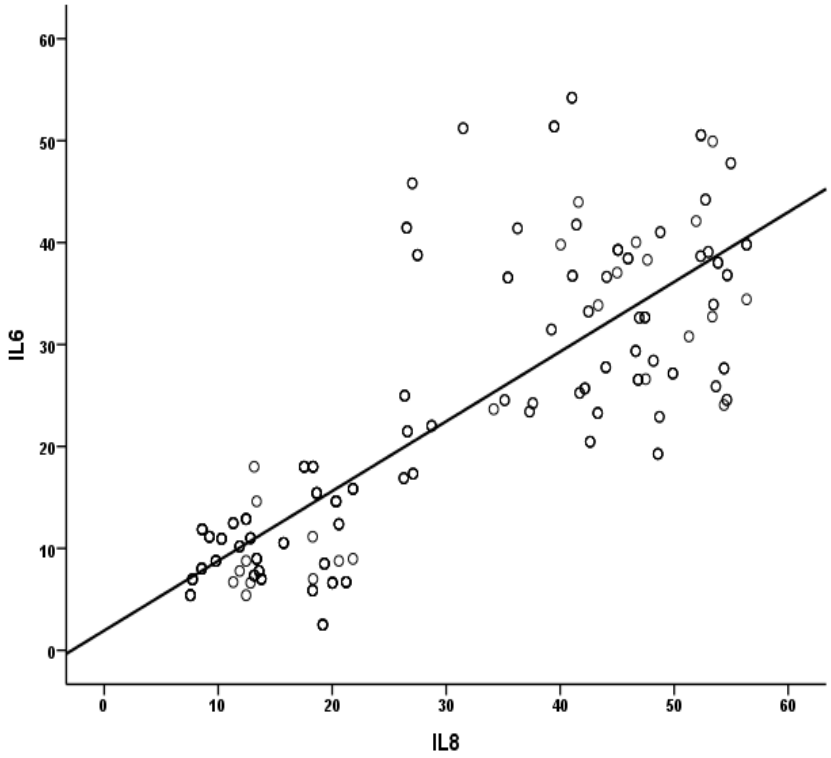


Figure 8: Scatter plot of IL 6 and IL 8



Discussion:

Vol. 20, No. 1. (2024)
E ISSN: 1672-2531

***Analysis of Biomarkers*:**

The early detection of neonatal sepsis continues to remain challenging for clinicians. Because of the delayed onset of symptoms and vague clinical presentation, neonatal sepsis is a major health issue[15]. The gold standard blood culture takes time and can be negative due to insufficient sample collection or intrapartum antibiotic use[16]. Unfortunately, if antibiotic treatment is delayed after the onset of clinical signs, it may be difficult to prevent a fulminant clinical progression that leads to septic shock and death[17].

However, both blood culture positive and negative neonates were employed in this investigation to examine serum IL-6, IL-8 and the commonly used CRP. These markers were evaluated to determine whether the above markers, alone or in combination, would be a more effective predictor of newborn sepsis.

It is possible to raise the specificity under the 'both positive' criterion or the sensitivity under the 'either positive' rule by combining two component tests. In practical practice, boosting sensitivity in a screening setting may be beneficial, whereas improving specificity in prominent infants may be beneficial in distinguishing an infection from other illnesses[18].

***C Reactive Protein*:**

An extremely significant statistical difference ($p<0.001$) was seen in CRP levels in our study. CRP level was 11mg/dl and 3mg/dl in cases and control participants respectively.

Morad *et al.*, (2020)studied the sepsis markers in neonates and found the median CRP level was 48 mg/dl (range: 4–140 mg/dl). The cut-off value in their study was ≥10 mg/dl. In our study, the median CRP serum level in cases was 11 mg/dl and ranged from 4.1 to 41.8 mg/dl. The median value was considerably low in our findings[19].

Tosson *et al.*,(2021)evaluated the diagnostic utility of CRP levels in full-term neonates LOS. They claimed that CRP demonstrated higher specificity and positive predictive value in LOS diagnosis at a cut-off value of 7.2 mg/L. In our study, the cut-off value was 6mg/dl[20].

When the CRP values were compared between the EOS and LOS cases, extremely significant statistical difference ($p<0.001$) was seen. The cut-off value for CRP was < 6mg/dl. It must be noted that the median CRP level in EOS was 6.6mg/dl. CRP findings in EOS can be crucial. So in EOS samples, it would be beneficial if CRP results are interpreted with other clinical findings or other biomarkers like IL 6 and IL 8.

According to a study by YochpazS *et al.*, (2020), the clinical EOS (cEOS) is only weakly predicted by the first plasma CRP values measured 6 to 8 hours after delivery. This finding was in concordance with our study findings[21].

Khan F *et al.*,(2019)conducted a study to measure the validity of CRP as a screening test for EONS and LONS. He concluded that CRP as a screening test has low screening validity in EOS which was similar to our results[22].

In our investigation, 65.7 % of the case participants had positive CRP levels. Bunduki GK *et al.*,(2020)studied the efficacy of CRP as a biomarker in forecasting neonatal sepsis and found 41.2% of infected neonates had elevated CRP.65.7% of cases had raised CRP levels in our study[23].

Panda SK *et al.*,(2021) found that the mean ± SD of CRP level in cases and control samples were 43.50±26.76 and 3.39±1.50 whereas in our study the median CRP level was 11mg/dl and 3mg/dl in cases and control participants respectively. The median CRP level in cases was considerably low in our investigation[24].

Our findings suggest CRP has a higher specificity and is a stronger indication of severe bacteremia in newborns which is similar to other studies. It is the widely available laboratory test for predicting and diagnosing newborn sepsis. Serial CRP monitoring, coupled with other biomarkers like interleukins, improves the diagnosis of newborn sepsis, according to a study[25].

***Interleukin 6*:**

The median (IQR) IL 6 level in our study was 32.99ng/l (24.98, 39.29) and 9.58ng/l (7, 12.47) in cases and control participants respectively. Extremely significant statistical difference ($p<0.001$) was seen in IL 6 levels in our study. In our study, there was no statistical difference between the levels of IL 6 among EOS and LOS samples.

Morad EA *et al.*, (2020) studied the sepsis markers in neonates. With a median level of 44 pg/ml (range: 6 to 120.8 pg/ml) in cases against a median of 14 pg/ml (range: 5-60 pg/ml) in controls, IL-6 levels in their study demonstrated a significant difference ($p=0.004$)[19].

In our study investigation, the cut-off value of IL 6 was found to be 18.64 ng/l. Sensitivity and specificity were calculated as 96.73% and 76.06% respectively. Accuracy, PPV and NPV were found to be 0.828, 0.664 and 0.979 respectively.

Qiu X *et al.*,(2018) investigated the early diagnostic level of IL-6 for NS with PROM. The overall pooled sensitivity was 0.85 and showed IL-6 high accuracy in diagnosing NS with PROM. Our study results had a higher sensitivity of 96.73%[26].

Sun B *et al.*,(2019)conducted a meta-analysis exploring the diagnostic value of IL-6 for neonatal sepsis. The pooled sensitivity and specificity of IL-6 were 82% and 88% respectively. In our study sensitivity was higher (96.73%) whereas specificity was lower (76.06%) comparatively[27].

Tessema B *et al.*, (2020)studied the diagnostic ability of interleukin-6 for early identification of NS. In their findings, IL-6 showed 73.1%, 80.2%, 37.6%, and 94.8% of sensitivity, specificity, PPV and NPV respectively. In our study, IL 6 showed 96.73% sensitivity, 76.06% specificity, 66.4% PPV and 97.9% NPV. When compared sensitivity and PPV value was found to be higher in our investigation[28].

According to Shoukry LR, *et al.*,(2021), the diagnostic accuracy for IL-6 with a cut-off value of 50 pg/ml was 95.16. The sensitivity and specificity were 100% and 90.32% respectively. In our study investigation, the cut-off value of IL 6 was found to be 18.64 ng/l. Our study results were lower in sensitivity, specificity, PPV and NPV when compared with their findings[29].

Ahmed AM et al., (2019) showed that IL-6 had high diagnostic efficiency with AUC of 0.751. The cutoff value was 24pg/ml, with a sensitivity of 94.4% and specificity of 52.4%, respectively. In our investigation sensitivity was similar whereas the specificity was comparatively higher[30].

According to Cortés JS *et al.*, (2021), there were variations in the levels of CRP and IL-6 between the control and NS groups. Additionally, in the LONS group, CRP demonstrated higher clinical accuracy, but in the EONS group, IL-6 demonstrated higher clinical accuracy. It matched the results of our study[31].

***Interleukin 8*:**

The median (IQR) IL 8 level in our study was 44.08 (37.46, 50.58) and 13.58 (10.8, 18.9) in cases and control participants respectively. Extremely significant statistical difference ($p<0.001$) was seen in IL 8 levels in our study. In our study, there was no significant statistical difference between the levels of IL 8 among EOS and LOS samples.

In our study investigation, the cut-off value of IL 8 was found to be 24.05 ng/l. Sensitivity and specificity were calculated as 100% and 74.46% respectively. Accuracy, PPV and NPV were found to be 0.828, 0.657 and 1 respectively.

Neonatal plasma IL-8 demonstrated a good predictive value (83%) for EONS, according to Nakstad B *et al.*,(2018). IL-8 was 3.6-fold increased in cases when compared to control neonates. An extremely significant statistical difference ($p<0.001$) was seen in IL 8 levels between the cases and control participants in our study[32].

Ahmed AM et al., (2019) showed that IL-8 had a high diagnostic performance for sepsis detection with AUC of 0.775. The cut-off values of IL-8 were 54pg/mL with a sensitivity of 83.3% and specificity of 71.4%. In our study, both sensitivity and specificity were higher (100%, 74.46%) comparatively[30].

Boskabadi H *et al.,* (2018)conducted a systematic review and showed IL8 with a cutoff value of 269.51 pg/ml had 80% sensitivity and 50% specificity. In our study, the cut-off value was 24.05 ng/l. Both sensitivity and specificity were higher (100%, 74.46%) comparatively[33].

Odabasi IO *et al.*,(2020) stated that IL-8 guides in the diagnosis and the assessment of severity of NS. Its sensitivity and specificity range were 80-91% and 76-100% respectively. In our study, specificity was a little lower (74.46%) [5].

Dillenseger L *et al.*,(2018) stated that semi-quantitative IL 8 had greater specificity (92.19%) but low sensitivity (48.15%). It was in contradiction with our study findings. In our study, specificity was lower (74.46%) but sensitivity was highest (100%) [34].

Haroun MA et al.,(2021) stated that IL-8 was significantly raised in cases than control ($p< 0.005$). Extremely significant statistical difference ($p<0.001$) was seen in IL 8 levels in our study. Among cases, there was no statistical difference in IL-8 levels between EOS and LOS. Similarly in our study, there was no statistical significance between EOS and LOS group[35].

***Correlation between biomarkers*:**

Nakstad B *et al.,* (2018) stated that there was no relation between CRP, WBC, and platelet count with PCT, IL-6, IL-8. Whereas in our study, IL 6 and IL 8 showed a strong positive correlation while CRP with IL 6 and IL 8 showed a moderate positive correlation.[32]

Shoukry LR *et al.*,(2021) *evaluated the diagnostic performance of IL-6 and stated that the* use of CRP and IL-6 as a panel for the early identification of NS could improve the sensitivity.[29]

The onset and progression of newborn sepsis are correlated with the inflammatory mediators CRP, IL-6, and IL-8. The diagnostic rate may be successfully increased with combined detection, which is advantageous for early diagnosis and therapeutic intervention.

Conclusion:

Rapid and precise diagnostic methodologies are needed to detect infected neonates to reduce the number of neonates treated empirically. Due to the lack of established cut-off values for sepsis indicators and non-specific clinical presentation, diagnosing newborn sepsis remains a problem for both laboratories and physicians. No currently available biomarker can deliver perfect diagnostic accuracy on its own. Simultaneous screening of several indicators can improve diagnosis. We noticed a large increase in IL 6& 8 levels, which might be used as a marker for detecting newborn sepsis. The combined effect of early and sensitive IL-6& 8 and late and specific CRP has been found to improve sepsis diagnosis sensitivity and specificity. As a result, a combination of biomarkers could be critical for diagnosis and better predictors of neonatal sepsis, as well as being important in the disease's etiology.

REFERENCES:

1. Fleischmann-Struzek C, Goldfarb DM, Schlattmann P, Schlapbach LJ, Reinhart K, Kissoon N. The global burden of pediatric and neonatal sepsis: a systematic review. The Lancet Respiratory Medicine. 2018 Mar 1;6(3):223-30.
2. You T, Zhang H, Guo L, Ling KR, Hu XY, Li LQ. Differences in clinical characteristics of early-and late-onset neonatal sepsis caused by Klebsiella pneumonia. International Journal of Immunopathology and Pharmacology. 2020 Aug;34:2058738420950586.
3. Yadav P, Yadav SK. Progress in Diagnosis and Treatment of Neonatal Sepsis: A Review Article. Journal of the Nepal Medical Association. 2022 Mar 1;60(247).
4. Jethani S, Bhutani N, Yadav A. Diagnostic utility of combined immature and total neutrophil counts along with C-reactive protein in early detection of neonatal sepsis: A cross-sectional study. Annals of Medicine and Surgery. 2022 May 1;77:103589.
5. Odabasi IO, Bulbul A. Neonatal sepsis. The Medical Bulletin of SisliEtfal Hospital. 2020;54(2):142.
6. Al-Matary A, Al Sulaiman M, Al-Otaiby S, Qaraqei M, Al-Matary M. Association between the timing of antibiotics administration and outcome of neonatal sepsis. Journal of Infection and Public Health. 2022 Jun 1;15(6):643-7.
7. Sharma D, Farahbakhsh N, Shastri S, Sharma P. Biomarkers for diagnosis of neonatal sepsis: a literature review. The Journal of Maternal-Fetal& Neonatal Medicine. 2018 Jun 18;31(12):1646-59.
8. Cui X, Shi Y, Zhao X, Xue X, Fu J. The clinical significance of interleukin-6, C-reactive protein, and procalcitonin in the early recognition of nosocomial infections in preterm infants.

9. Celik IH, Hanna M, Canpolat FE, Pammi M. Diagnosis of neonatal sepsis: the past, present and future. Pediatric research. 2022 Jan;91(2):337-50.
10. Ebenebe CU, Hesse F, Blohm ME, Jung R, Kunzmann S, Singer D. Diagnostic accuracy of interleukin-6 for early-onset sepsis in preterm neonates. The Journal of Maternal-Fetal & Neonatal Medicine. 2021 Jan 17;34(2):253-8.
11. Gude SS, Peddi NC, Vuppalapati S, Gopal SV, Ramesh HM, Gude SS. Biomarkers of Neonatal Sepsis: From Being Mere Numbers to Becoming Guiding Diagnostics. Cureus. 2022 Mar 16;14(3).
12. Rana D, Hazarika H, Agarwal A, Gupta R, Kotru M. Validation of Hematological Markers in Early Onset Neonatal Sepsis. Cureus. 2022 Jun 30;14(6).
13. Eichberger J, Resch E, Resch B. Diagnosis of Neonatal Sepsis: The Role of Inflammatory Markers. Frontiers in Pediatrics. 2022;10.
14. Tosson AM, Koptan DM, Kamal M, AbdElhady M. Assessment of Serum Interleukin-27 and Mean Platelet Volume in Late-Onset Neonatal Sepsis. American Journal of Perinatology. 2022 May 17.
15. Popescu CR, Cavanagh MM, Tembo B, Chiume M, Lufesi N, Goldfarb DM, Kissoon N, Lavoie PM. Neonatal sepsis in low-income countries: epidemiology, diagnosis and prevention. Expert review of anti-infective therapy. 2020 May 3;18(5):443-52.
16. Ershad M, Mostafa A, Dela Cruz M, Vearrier D. Neonatal sepsis. Current emergency and hospital medicine reports. 2019 Sep;7(3):83-90.
17. Kurul S, Fiebig K, Flint RB, Reiss IK, Küster H, Simons SH, Voller S, Taal HR. Knowledge gaps in late-onset neonatal sepsis in preterm neonates: a roadmap for future research. Pediatric Research. 2022 Jan;91(2):368-79.
18. Hincu MA, Zonda GI, Stanciu GD, Nemescu D, Paduraru L. Relevance of biomarkers currently in use or research for practical diagnosis approach of neonatal early-onset sepsis. Children. 2020 Dec 20;7(12):309.
19. Morad EA, Rabie RA, Almalky MA, Gebriel MG. Evaluation of procalcitonin, C-reactive protein, and interleukin-6 as early markers for diagnosis of neonatal sepsis. International journal of microbiology. 2020 Oct 1;2020.
20. Tosson A, Koptan D, Aal RA, Elhady MA. Evaluation of serum and salivary C-reactive protein for diagnosis of late-onset neonatal sepsis: a single center cross-sectional study. Jornal de Pediatria. 2021 Dec 6;97:623-8.
21. Yochpaz S, Friedman N, Zirkin S, Blumovich A, Mandel D, Marom R. C-reactive protein in early-onset neonatal sepsis–a cutoff point for CRP value as a predictor of early-onset neonatal sepsis in term and late preterm infants early after birth?. The Journal of Maternal-Fetal& Neonatal Medicine. 2020 Dec 1:1-6.
22. Khan F. C-reactive protein as a screening biomarker in neonatal sepsis. J Coll Physicians Surg Pak. 2019 Oct 1;29(10):951-3.
23. Bunduki GK, Adu-Sarkodie Y. The usefulness of C-reactive protein as a biomarker in predicting neonatal sepsis in a sub-Saharan African region. BMC research notes. 2020 Dec;13(1):1-5.
24. Panda SK, Nayak MK, Rath S, Das P. The utility of the neutrophil-lymphocyte ratio as an early diagnostic marker in neonatal sepsis. Cureus. 2021 Jan 24;13(1).

25. Gilfillan M, Bhandari V. Neonatal sepsis biomarkers: where are we now?. Research and Reports in Neonatology. 2019 Mar 14;9:9-20.
26. Qiu X, Zhang L, Tong Y, Qu Y, Wang H, Mu D. Interleukin-6 for early diagnosis of neonatal sepsis with premature rupture of the membranes: a meta-analysis. Medicine. 2018 Nov;97(47).
27. Sun B, Liang LF, Li J, Yang D, Zhao XB, Zhang KG. A meta-analysis of interleukin-6 as a valid and accurate index in diagnosing early neonatal sepsis. International wound journal. 2019 Apr;16(2):527-33.
28. Tessema B, Lippmann N, Willenberg A, Knüpfer M, Sack U, König B. The diagnostic performance of interleukin-6 and C-reactive protein for early identification of neonatal sepsis. Diagnostics. 2020 Nov 20;10(11):978.
29. Shoukry LR, Mohamed AN, Sharaf AE, Osman OB. Diagnostic markers for early detection of neonatal sepsis. Journal of Scientific Research in Medical and Biological Sciences. 2021 Aug 12;2(3):13-26.
30. Ahmed AM, Mohammed AT, Bastawy S, Attalla HA, Yousef AA, Abdelrazek MS, Alkomos MF, Ghareeb A. Serum biomarkers for the early detection of the early-onset neonatal sepsis: A single-center prospective study. Advances in Neonatal Care. 2019 Oct 1;19(5):E26-32.
31. Cortés JS, Losada PX, Fernández LX, Beltrán E, DeLaura I, Narváez CF, Fonseca-Becerra CE. Interleukin-6 as a biomarker of early-onset neonatal sepsis. American Journal of Perinatology. 2021 Aug;38(S 01):e338-46.
32. Nakstad B. The diagnostic utility of procalcitonin, interleukin-6 and interleukin-8, and hyaluronic acid in the Norwegian consensus definition for early-onset neonatal sepsis (EONS). Infection and Drug Resistance. 2018;11:359.
33. Boskabadi H, Zakerihamidi M. Evaluate the diagnosis of neonatal sepsis by measuring interleukins: A systematic review. Pediatrics& Neonatology. 2018 Aug 1;59(4):329-38.
34. Dillenseger L, Langlet C, Iacobelli S, Lavaux T, Ratomponirina C, Labenne M, Astruc D, Severac F, Gouyon JB, Kuhn P. Early Inflammatory markers for the diagnosis of late-onset sepsis in neonates: the nosodiag study. Frontiers in pediatrics. 2018 Nov 13;6:346.
35. Haroun MA. Evaluation Plasma Interleukin-8 (IL-8) Levels among Neonatal Sepsis Patients in some Selected Hospitals–Khartoum State-Sudan (Doctoral dissertation, Sudan University of Science & Technology).