Original Article

Assessment of the Biomarkers in Neonatal Sepsis: An Observational Study

Dr. Brajesh Kumar[1], Dr. Sujit Nath Choudhury[2]

1. Post Graduate Trainee, Dept of Pediatrics, Silchar Medical College and Hospital, Assam, India.
2. Associate Professor, Dept of Pediatrics, Silchar Medical College and Hospital, Assam, India.

***Corresponding author's E-mail:** brajeshjlnmc07@gmail.com

Received: 18-12-2024; **Revised:** 23-04-2025; **Accepted:** 06-05-2025; **Published online:** 15-05-2025.

ABSTRACT

Introduction: Infants who exhibit clinical signs of illness are given sepsis workups and put on broad-spectrum antibiotics unless a definitive diagnosis is obtained. Furthermore, inadequate clinical evaluation and delays in identifying sepsis episodes have an effect on the management of the illness. Furthermore, little is known about the precise clinical predictors and biomarkers of poor outcomes from newborn sepsis in low- and middle-income countries.

Aims/ objective: To determine and compare the biomarkers in neonatal sepsis versus healthy neonates (control).

Materials and Method: In this observational study, 50 patients with neonatal sepsis and 50 neonates without sepsis were evaluated for serum biomarkers. Blood specimens were taken from all neonates before the start of antimicrobial agents for the sepsis work up. The samples included haematological parameters such as platelet count, degenerative manifestation in the neutrophils, culture of the blood, antibiotic sensitivities, total leukocyte count, total neutrophil count, immature neutrophils divided by total neutrophil count (I/T ratio), procalcitonin, and CRP estimation.

Results: Incidence of prematurity, and low birth weight were greater in neonates with sepsis as compared to control, but the difference was not significant ($p>0.05$). CRP, PCT and serum lactate levels were significantly raised in patients with neonatal sepsis as compared to healthy controls. ($p<0.0001$) CRP, PCT, and I:T ratio was more associated with neonatal sepsis, as compared to leucocytosis and micro-ESR.

Conclusion: The present study concludes that serum PCT and CRP levels are a more reliable indication for the early identification of newborn sepsis and for assessing the response of the illness to antibiotic therapy compared to ESR or leukocyte counts.

Keywords: Neonatal Sepsis, Biomarker, C-Reactive Protein, Procalcitonin, Leukocyte Count.

INTRODUCTION

In the initial 28 days of life, neonatal sepsis is a clinical state of systemic sickness associated with the bacteremia. There are two forms of neonatal sepsis: late-onset sepsis, which appears as septicemia beyond 72 hours after delivery, and early-onset sepsis, which presents as breathing problems within 72 hours of birth. [1-3]

Neonatal sepsis, comprising either early-onset sepsis (EOS) or late-onset sepsis (LOS), has been reported to occur in one to five cases out of each thousand live births, whereas one to two instances occurring in term babies per 1000 live births. The total mortality rate is up to 24.4%, but for infants delivered around 22 to 24 weeks of pregnancy as well as between 25 and 28 weeks, it can reach as high as 54% and 30%, respectively. [4]

Maternal risk factors such as a condition called PROM, premature pregnancy, extended rupture of membranes, as well as intrapartum maternal fever, as well as neonatal risk factors such as premature newborn, low birth weight at delivery, foetal distress, lower APGAR score, as well as multiple gestations, can all contribute to a neonate's development of EOS. [1] Several signs and symptoms of neonatal sepsis include decreased feed acceptance, difficulty breathing, respiratory infections, apnoea, delayed capillary refill time, cold peripheral regions, mottling, off colour, necrotizing enterocolitis, temperature instabilities (hypothermia as well as hyperthermia), reduced muscular tone, epilepsy, protruding fontanels, DIC, haemorrhaging manifestation, as well as prolonged jaundice. [4,5]

Furthermore, an infant suffering from neonatal sepsis may experience serious problems such as post-infectious encephalopathy, convulsions, hydrocephalus, encephalomalacia, cerebral infarction, delayed neurodevelopment, and sensory abnormalities. [1]

Infants who exhibit clinical signs of illness are given sepsis workups and put on broad- spectrum antibiotics unless a definitive diagnosis is obtained. The identification of microorganism through blood cultures is the gold standard for making a conclusive diagnosis of neonatal sepsis. [6, 7]

The majority of neonatal sepsis care guidelines were created in high-income nations. [8–9]

Although these recommendations are very helpful in identifying the best course of treatment, it is challenging to apply them to healthcare institutions in India with lower resources. [10–12] Furthermore, inadequate clinical evaluation and delays in identifying sepsis episodes have an effect on the management of the illness. Furthermore, little is known about the precise clinical predictors and biomarkers of poor outcomes from newborn sepsis in low- and middle-income countries. [11, 12]

So, this study was planned to determine and compare the biomarkers in neonatal sepsis versus healthy neonates (control).

MATERIALS AND METHODS

This was an observational study conducted in patients of neonatal sepsis. in Department of General Medicine in a tertiary care hospital of India from January 2018 to June 2018. Informed consent was taken from children with and without FC as per recommendation of Declaration of Helsinki and Good Clinical Practice.

Inclusion Criteria:

Cases:

- Neonates of age less than or equal to 28 days
- Patients admitted in neonatal ICU
- Neonates with diagnosis of culture confirmed neonatal sepsis Control
- Healthy neonates with matched gestational age and gender

Exclusion Criteria:

- Length of hospital stay less than 24 hours
- Patients with recurrent sepsis
- Failure to obtain informed consent

Sample Size: Consecutive sampling was done and 50 patients with neonatal sepsis (case) and 50 healthy neonates (control) were enrolled in the study.

Individualized case report forms were utilized to gather patient data. These included demographic information, information about the mother, clinical features, biomarkers like procalcitonin and CRP, evaluations, therapies, and outcomes.

Specimens and Tests

Blood specimens were taken from all neonates before the start of antimicrobial agents for the sepsis work up. The samples included haematological parameters such as platelet count, degenerative manifestation in the neutrophils, culture of the blood, antibiotic sensitivities, total leukocyte count, total neutrophil count, immature neutrophils divided by total neutrophil count (I/T ratio), procalcitonin, and CRP estimation.

Serum CRP

Following the manufacturer's instructions, an immune-turbidimetric approach was used in the laboratory to quantify the serum's CRP quantitatively. Reagent linearity was seen as much as 150 mg/L. Up to 6 mg/L was the reference standard. [13]

Serum PCT

Using a quantitative immuno-luminometry technique, the serum PCT level was determined. A PCT value ≥0.5 ng/ml was deemed abnormal in this assay. PCT values were classified as mildly positive, positive, and significantly positive, respectively, at 0.5‥2 ng/ml, 2‥10 ng/ml, and >10 ng/ml.[13]

Statistical Analysis: Data from patients with or without neonatal sepsis were presented in tabular form using Microsoft Excel 365 and transferred to SPSS version 24 for further statistical analysis. Continuous data such as age, birth weight, levels of CRP, PCT, and serum lactate were expressed as mean ± SD (standard deviation). Statistical significance of difference in continuous data between patients with and without neonatal sepsis was evaluated by unpaired t-test. Categorical data, including incidence of deranged biomarker, gender, and type of delivery were reported as percentages and frequencies and then compared by chi-square test or fisher's exact test. A p-value of less than 0.05 was taken as cut-off for statistical significance.

RESULTS

- Most of the patients were between 36-40 weeks of gestation with vaginal delivery in both groups and most of them were males. Incidence of prematurity, and low birth weight were greater in neonates with sepsis as compared to control but the difference was not significant (p>0.05).

Table 1: Baseline Characteristics of Patient with Neonatal Sepsis and Healthy Control

Variable	Cases (n=50)	Control (n=50)	P-Value
Gestational age in week	38.14 ± 2.39	38.58 ± 1.92	0.31
Male Gender	31	32	>0.99
Female Gender	19	18	
Prematurity	18	13	0.39
Birth weight in kg	2.68 ± 0.51	2.78 ± 0.39	0.27
Low birth weight (<2.5 kg)	21	16	0.41
Vaginal Delivery	31	34	0.68
Caesarean Section Delivery	19	16	

Table 2: Clinical Features of Patient with Neonatal Sepsis

Variable	Number of Patients	%
Fever >38.5°C	16	32.00
Requirement for mechanical ventilation	18	36.00
Impaired peripheral perfusion	9	18.00
Feeding intolerance	23	46.00
Lethargy	8	16.00

36% of patients required mechanical ventilation and 18% had impaired peripheral perfusion.

Table 3: Comparison of Biomarkers between Patients with Neonatal Sepsis and Healthy Control

Variable	Cases (n=50)	Control (n=50)	P-Value
CRP in mg/L	23.26 ± 3.26	7.69 ± 0.72	<0.0001
PCT in ng/ml	7.90 ± 0.53	0.78 ± 0.05	<0.0001
Serum Lactate in mmol/L	4.78 ± 0.41	2.71 ± 0.23	<0.0001

CRP, PCT and serum lactate levels were significantly raised in patients with neonatal sepsis as compared to healthy controls. (p<0.0001)

Table 4: Comparison of Incidence of deranged Biomarkers between Patients with Neonatal Sepsis and Healthy Control

Variable	Cases (n=50)	Control (n=50)	P-Value
Elevated PCT	46	7	<0.001
Elevated CRP	26	4	<0.001
Leucocytosis	7	3	0.32
Micro ESR	3	0	0.24
I:T Ratio	8	1	0.03

CRP, PCT, and I:T ratio was more associated with neonatal sepsis, as compared to leucocytosis and micro-ESR.

DISCUSSION

With its substantial mortality rate, neonatal sepsis continues to provide challenges for neonatal healthcare practitioners in terms of diagnosis and treatment. Neonatal septicaemia death rates can be decreased by prompting the clinician to start antibiotic therapy. This is made possible by a quick identification of the condition. Early detection of an infected baby also assists in avoiding antibiotic therapy to a non-infected neonate needlessly. The blood culture has a low yield, is difficult, and takes a long time. The sensitivity of the leukocyte differential analysis and the easily attainable complete blood count for sepsis diagnosis is somewhat low.

For many years, the most examined criterion for bacterial infection prediction has been the CRP.[14,15]

This protein functions as a "scavenger" by opsonizing bacteria, triggering the complement system, and promoting phagocytosis during the process of inflammatory reactions. A biomarker of bacterial sepsis was recently proposed as procalcitonin (PCT). PCT has an advantage over CRP in that it increases bacterial infection more quickly and returns the body to normal more quickly. [16] The accurate marker of sepsis didn't have an elevated micro-ESR level. In only eight occurrences was an anomalous I/T ratio noted. The findings of Rodwell, Zipursky, and Basu et al. were in conflict with this.[17–19]

In addition, low PCT levels were useful in ruling out septicemia as a diagnosis. In newborns, a high PCT level may aid in the prediction of neonatal sepsis. As a result, the PCT evaluation may assist doctors in reducing the quantity of prescriptions for antibiotics they write. In the current investigation, both the suspected and confirmed instances of sepsis in newborns had exceptionally high PCT levels. This result was similar to the findings of studies by Monneret et al. as well as Yadolla Zahedpasha et al. [20, 21]

The blood PCT level and (p<0.001) showed a substantial association, which is consistent with the findings of the study by Koksal et al. [22] According to Chiesa et al., 100% of neonates could be diagnosed with late-onset sepsis at a time beyond 48 hours after birth. [23]

In 67 newborns, Vazzalwar et al. evaluated PCT as the detection of late-onset sepsis. Sensitivity and specificity were determined to be 97% and 80%, respectively, at a PCT threshold value of 1.0 ng/ml, and 72% and 93%, respectively, with CRP. [24]

In While the remaining sepsis screening tests came back negative in the majority of the culture-positive patients, the PCT level was increased. This was consistent with the findings of Boo et al. [25] These results bolster the value of PCT for determining the earlier detection of sepsis in neonates. The current study supported the findings of other researchers, who found that PCT had been more sensitive compared to CRP in identifying neonatal sepsis and that PCT levels increased earlier in sepsis than CRP levels did. In a recent study, Koksal et al. came to the conclusion that the serum PCT level was more effective than the serum CRP

level in identifying the severity of the disease, assessing the response to antibiotic treatment, and providing an early diagnosis of newborn sepsis. [22]

Nevertheless, when PCT and CRP are combined, an adverse PCT test outcome may aid in "ruling out," whereas a high CRP result aids in "ruling in," the potential of sepsis, especially late-onset sepsis. We suggest that the PCT findings on the day of the newborn's admittance to the NICU be used to determine when to start antibiotics, based on the findings of this study. Of the fifty instances, forty-six had elevated PCT levels, whereas only twenty-six had elevated CRP levels.

PCT is a useful diagnostic and therapeutic tool for newborn sepsis, as evidenced by the high serum levels found in nearly all cases of sepsis with culture-proven sepsis. PCT helps distinguish bacterial infections from viral infections and is very specific for bacterial infections. It has a strong correlation with the infection's severity and rate of progression. Prior to the blood culture report being available, which is typically three to five days following admission, PCT aids in the early detection of sepsis on the day of admission. By preventing antibiotic medication when it is not necessary, PCT lowers costs and the likelihood of bacterial resistance. PCT may additionally be employed to predict the outcome of sepsis.

CONCLUSION

The present study concludes that serum PCT and CRP levels are a more reliable indication for the early identification of newborn sepsis and for assessing the response of the illness to antibiotic therapy compared to ESR or leukocyte counts. Hospital expenses are decreased when serum PCT is frequently measured for the detection and follow-up of newborn sepsis. Such an advantage could encourage the test to be accepted more widely in everyday practice. In conjunction with supporting the training as well as education of healthcare personnel in adopting a more hazard-based approach to patient care, emphasizing the significance of measures to prevent and control infections, these data will enable early risk classification and appropriate actions.

Acknowledgement: We are thankful to the healthcare workers of Silchar Medical College and Hospital, Assam, India

Source of Support: The author(s) received no financial support for the research, authorship, and/or publication of this article

Conflict of Interest: The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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