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Cord blood inflammatory markers for the diagnosis of early-onset neonatal sepsis

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***Abstract*---**Objective: The present study was design to screen early-onset neonatal sepsis using cord blood inflammatory markers. Additionally, the study compared suitable inflammatory markers for septic screen using cord blood. Study Design: This single-centre observational study included neonates suspected of early onset neonatal sepsis having gestational age >28 weeks and weight >1000gm, delivered at a Tertiary Care Centre. Cord blood was collected immediately after birth (0 hour) and clinical parameters (i.e., C-reactive protein (CRP), interleukin 6 (IL-6), procalcitonin, blood culture) as well as haematological parameters (i.e., absolute neutrophil count (ANC), white blood cell count (WBC)) associated with early-onset neonatal sepsis were examined. Results: The study included 44 neonates of mean gestational age of 36.1 ± 3.2 weeks. Receiver operating characteristic curve (ROC) for sepsis markers showed that procalcitonin demonstrated the highest sensitivity and specificity of 90.9% and 100%, respectively; AUC of 0. 966 (95% CI, 0.862-0.998, p <0.001) with cut off ≤0.48. IL-6 significantly predicted early-onset neonatal sepsis with an AUC of 0.965 (95% CI, 0.861-0.997, p <0.001); optimum cut off ≤ 6.4pg/ml with sensitivity and specificity of 90.9% and 95.5%, respectively. The sensitivity and specificity for CRP were 81.8% and 31.8%, respectively. WBC did not significantly predict early onset neonatal sepsis with sensitivity and specificity of 81.8% and 40.9%, respectively. ANC did not significantly predict early-onset neonatal sepsis with sensitivity and specificity of 86.4% and 36.4%, respectively. Conclusion: Though blood culture is the gold standard for diagnosis of early onset neonatal sepsis, the inflammatory markers can assist in diagnosing early onset neonatal sepsis at the earliest. The procalcitonin and IL-6 provide better

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sensitivity and specificity in diagnosis of early onset neonatal sepsis as compared to CRP, WBC and ANC through cord blood sampling.

***Keywords*---**cord blood, inflammatory markers, early-onset neonatal sepsis.

Introduction

The neonatal sepsis is a clinical syndrome characterized by signs and symptoms of infection with or without accompanying bacteremia in the first month of life. It is either early-onset (EONS) (<72 hours of birth) or late-onset (LONS) (>72 hours) and it is considered one of the crucial leading factors for neonatal mortality and morbidity, particularly in developing countries(1). However, the symptoms and signs of neonatal sepsis in both preterm and term infants are mostly non-specific which makes the diagnosis challenging and the optimal strategy for treatment of such infants become unclear. Consequently, diagnosing early is crucial as it helps in starting antibiotic therapy early, while reducing neonatal mortality(2).

The application of conventional blood culture for the isolation of microorganisms has been the gold standard method for diagnosis(3). False negative results in this method are especially due to antibiotic use. Moreover, false-positive results are due to contamination. Generally, culture results have a time delay for 3-5 days, and it is difficult to obtain serial blood samples from neonates. The delay in getting the blood culture results usually leads to delayed initiation of treatment that may result into serious complications and even mortality as the treatment does not get initiated on time and by then the contamination in the infants flares up that becomes difficult to control(2). To avoid such catastrophic situations, suspected neonates are treated with broad-spectrum antibiotics prophylactically until sepsis is excluded(4). On the other hand, the overuse of such empiric antibiotics favours the development of resistance.

Therefore, improvement in the diagnostic accuracy of tests should avoid the indiscriminative use of antibiotics in non-septic neonates. Several recent studies have included various sepsis markers, like procalcitonin (PCT), C-reactive protein (CRP), and interleukin-6 (IL-6) to improve diagnostic accuracy of neonatal sepsis(3). C-reactive protein (CRP), an acute-phase protein, is produced in the liver in response to inflammation or infection. It is useful in excluding infections in suspected septic neonates and enabling earlier discontinuation of antibiotics. Cost-effectiveness is improved, decreasing the hospital stay duration and development of resistance. Procalcitonin (PCT) is synthesized by the C cells of the thyroid gland in very low concentrations in normal physiologic state. Procalcitonin (PCT) is induced specially as an acute-phase reactant in bacterial sepsis. It is produced by monocytes and macrophages of many organs in response to bacterial lipopolysaccharide (LPS), which is a potent inducer of procalcitonin (PCT) in circulation during severe bacterial infection(5). Interleukin-6 (IL-6), is one of the most significant cytokines, having an early rise in concentration during the inflammatory cascades. IL-6 is produced by macrophages and monocytes as a response to bacterial infection. It is essential for the survival of the plasma cells, which are responsible for antibodies secretion and enhancing cytotoxic T-cells

differentiation. IL-6 along with CRP, is also a hepatocyte stimulating factor giving rise to the acute-phase response. These inflammatory markers can be helpful in detection of neonatal sepsis as adjuvant to blood culture(1).

Blood sample collection from neonates can be challenging as well as painful to the neonates. The peripheral vein sampling procedure mandates skilled health care workers who must dedicate quality time for sampling. Whereas umbilical cord blood on the other hand does not involve pain botheration, avoids stress and other procedural complications(6). Large quantities of blood can be obtained for sampling using umbilical cord blood sampling. Thus, this method of blood collection through umbilical cord can be applied for overall betterment in neonates.

Materials and Methods

This as an observational, single-centre study conducted at a Tertiary Care Centre in India. The study consisted of randomly selected eligible neonates that were delivered at this Tertiary Care Centre for a period of 14 months. Of total 2395 deliveries in the institution during the study period, 44 neonates were selected for the study. Neonatal inclusion criteria were weight of newborn must be more than 1000 gm and gestational age of newborn must be more than 28 weeks. Maternal inclusion criteria required at least 2 of the following: Maternal fever (>100.4 F) within 2 weeks prior to delivery; Premature rupture of membranes >16 hours; Foul smelling liquor; Prolonged Labor (1st stage + 2nd stage >24 hrs). The main exclusion criteria were neonate with gross congenital malformation, neonate with birth asphyxia, and negative consent. Early onset neonatal sepsis was defined as- TLC<5,000/mm^3 or more than 14000/mm^3, CRP-positive, IL-6 >60 pg/ml, procalcitonin >6 ng/ml, blood culture-positive. Cord blood was collected immediately after birth (0 hour) for the estimation of TLC, IL-6, CRP, procalcitonin and blood culture. The study was approved by institutional ethics committee before initiation. Written informed consent was obtained from the parents of newborns prior to the enrolment of subjects in the study.

Method of umbilical cord blood collection

Umbilical cord was properly clamped at the placenta side as well as the infant side. Cord was then wiped with 70% isopropyl alcohol using sterile method. Using a sterile 22-guage needle and syringe, around 4 ml to 4.5 ml of blood was taken out into syringe from umbilical vein. Needle was then replaced from syringe with new sterile needle and culture bottle top was cleaned with alcohol. 2 ml blood (1 ml in preterms) was placed in aerobic blood culture bottle, 1 ml blood was taken into EDTA bulb and remaining 1 ml blood sample was then sent to laboratory for serum separation and other diagnostic tests.

Laboratory investigations

The following laboratory investigation were carried out.

a. Cord blood sample in EDTA bulb for Complete Blood Count (aperature impedance technology)

b. Cord blood sample in plain bulb for C-reactive protein estimation with titres (agglutination slide test)
c. Cord blood sample in plain bulb for Interleukin-6 estimation (solid phase enzyme amplified immunoassay)
d. Cord blood sample in plain bulb for procalcitonin estimation (fluorescence immunoassay)
e. Cord blood sample in preterm neonates and in term neonates for blood culture (Aerobic growth media)

Statistical analysis

Continuous variables are expressed as means and standard deviations (range) and categorical variables are reported as frequencies and percentages. The optimal cut-off levels of CRP, IL-6, procalcitonin, WBC, and ANC were determined by receiver operating curve (ROC) analysis using Youden's index. Besides, specificity, sensitivity, positive predictive value (PPV), and negative predictive value (NPV) were computed. Descriptive analysis was performed using the Statistical Package for Social Sciences (SPSS) version 16.0 software (SPSS Inc., Chicago, IL, USA). Analysis of ROC and specificity, sensitivity, PPV, and NPV were done using MedCalc Version 20.014. P-value <0.05 was considered statistically significant.

Results

Total 44 neonates with clinically suspected early-onset neonatal sepsis were included in the study. The detailed description about maternal and neonatal factors is mentioned hereby.

Maternal factors

The youngest mother was 19 years old, and the oldest was 41 years old. The mean age of the mothers was 26.6 ± 5.1 years. The maximum number of mothers was aged <25 years (n =18, 40.9%), followed by aged between 25-30 years (n = 16, 36.4%), 31-35 years (n = 5, 11.4%), 36-40 years (n = 3, 6.8%). Fever within two weeks prior to delivery was found in 29 (65.9%) mothers, use of antibiotics in 44 (100%) mothers, premature rupture of membranes in 20 (45.5%) mothers, foul-smelling liquor in 27 (61.4%) mothers, and prolong labour (1st and 2nd stage >24 hrs) in 19 (43.2%) mothers. Out of 44 mothers, 20 (45.5%) mothers were delivered by lower (uterine) segment caesarean section (LSCS) and 24 (54.5%) mothers were delivered by normal vaginal delivery (NVD).

Neonatal factors

The mean gestational age of 44 included neonates was found to be 36.1 ± 3.2 weeks. Total 24 (54.5%) neonates were term and 20 (45.5%) neonates were preterm. The range of birth weight was 1000 to 3842 grams. The mean birth weight of 44 neonates was 2214.1 ±777.3 grams. Out of 44, none of the neonates required resuscitation manoeuvre. Of the total 44 neonates, 30 (68.2%) neonates were admitted to NICU in <15 days, 11 (25%) between 15-30 days, and 3 (6.8%) in ≥31 days.

Neonates with early-onset neonatal sepsis commonly presented with respiratory distress (59.1%) followed by lethargy (34.1%), feed intolerance (29.5%), temperature instability (27.3%), hypotonia (15.9%), and shock (13.6%). Other clinical findings included hypoglycaemia (9.1%) and tachycardia (2.3%).

Results of blood culture, CRP, IL-6, procalcitonin, WBC and ANC

Blood culture was positive in 22 (50%) and negative in 22 (50%) neonates. Moreover, the proportion of neonates with CRP positive and negative were 11 (25%) and 33 (75%) neonates, respectively. Further, IL-6 and procalcitonin were raised in 22 (50%) and 24 (54.5%) neonates, respectively. WBC and ANC were found to be abnormal in 25 (56.8%) and 7 (15.9%) neonates. A total of 25 variety of organisms were isolated in the blood culture of neonates with early-onset neonatal sepsis. Of which, Escherichia Coli (20%), Staphylococcus Haemolyticus (16%), and Klebsiella Pneumonia (16%) were the most found organisms. pon analysis, maternal risk factors (e.g. Fever within 2 weeks prior to delivery, premature rupture of membranes, foul smelling liquor, and prolonged labour) were not significantly associated with CRP, IL-6, procalcitonin, WBC and ANC positivity. There was no significant association was found between mode of delivery and CRP, IL-6, procalcitonin, WBC and ANC positivity.

Predictors of early-onset neonatal sepsis

The CRP did not significantly predict early-neonatal sepsis with an AUROC of 0.568 (95% CI, 0.410-0.717, P = 0.306). The optimum cut-off value was ≤0 mg/dl with sensitivity and specificity of 81.8% and 31.8%, respectively (Blood culture positive n = 22; CRP positive n = 11). IL-6 significantly predicted early-onset neonatal sepsis with an AUROC of 0.965 (95% CI, 0.861-0.997, P <0.001). The optimum cut-off value was ≤6.4 pg/ml with sensitivity and specificity of 90.9% and 95.5%, respectively (Blood culture positive n = 22; raised IL-6 n = 22). Procalcitonin significantly predicted early-onset neonatal sepsis with an AUROC of 0.966 (95% CI, 0.862-0.998, P <0.001). The optimum cut-off value was ≤0.48 ng/ml with sensitivity and specificity of 90.9% and 100%, respectively (Blood culture positive n = 22; raised procalcitonin n = 24). WBC did not significantly predict early-onset neonatal sepsis with an AUROC of 0.511 (95% CI, 0.356-0.665, P = 0.902). The optimum cut-off value was ≤13,900/mm3 with sensitivity and specificity of 81.8% and 40.9%, respectively (Blood culture positive n = 22; abnormal WBC n = 25). ANC did not significantly predict early-onset neonatal sepsis with an AUROC of 0.540 (95% CI, 0.384-0.691, P = 0.660). The optimum cut-off value was >2457/mm3 with sensitivity and specificity of 86.4% and 36.4%, respectively (Blood culture positive n = 22; abnormal ANC n = 7). The AUC of IL-6 and procalcitonin were higher than that of other studied parameters (Table 13). Thus, we can say that IL-6 and procalcitonin were effective predictors of early-onset neonatal sepsis as an alternative to blood culture testing.

Discussion

The main findings of the present study were: (1) fever in mothers prior to delivery was the major risk factor; (2) gestational age of neonates did not affect the occurrence of EONS; (3) Procalcitonin and IL-6 had better specificity and

sensitivity than CRP, WBC, and ANC in accordance with blood culture results. In view of the aim of this study to detect EONS using a varied array of inflammatory markers and to screen EONS at the earliest, the blood sample collection through umbilical cord upheld to be the first step towards early diagnosis. Avoidance of pain along with early sample collection was the major boon of this procedure. Some previous studies have demonstrated the application and advantages of umbilical cord blood sampling over peripheral blood sampling. Jain P, et al.(6) Have recently reported sensitivity of 81.0% and accuracy of 87% in prediction of EONS by umbilical cord blood collection method over peripheral vein blood collection method stating its reliability and applicability as an alternative for prediction of outcome in patients with EONS. Another study also determined that umbilical cord blood collection showed appropriate diagnostic correctness in identification of neonatal sepsis even in high-risk patients when compared with peripheral cord blood collection(7).

In the present study, fever was found in 65.9% mothers, premature rupture of membranes in 45.5% mothers, foul smelling liquor in 61.4% mothers, and prolong labour in 43.2% mothers. In the study by Heutz N, et al.(8), only 5.9% mothers had fever prior to delivery and premature rupture of membrane was observed in 3.5% of mothers. Similarly, in a study by Mukhopadhyay S, et al.(9) The maternal related risks like fever was present in 33.8% mothers whereas premature rupture as observed in 5.4% patients. In another study, Cancelier AC, et al.(10) Reported use of antenatal antibiotics in 20% of the mothers and premature membrane rupture in 5% of patients. In present study, clinical features of neonates were majorly respiratory distress (59.1%), lethargy (34.1%), hypotonia (15.9%), temperature instability (27.3%), and feed intolerance (29.5%). In the study by Jain P, et al.(6), the observed clinical features were feed Intolerance, vomiting, jaundice, abdominal distension, respiratory distress, hypoglycaemia and hypothermia.

The ROC curves for sepsis markers showed that procalcitonin demonstrated the highest sensitivity, specificity, PPV, and NPV of 90.9%, 100%, 100% and 99% than other studied markers. This was followed by IL-6, CRP, WBC and ANC. The IL-6 significantly predicted early-onset neonatal sepsis with an AUC of 0.965 (95% CI, 0.861-0.997, $p < 0.001$); with sensitivity and specificity of 90.9% and 95.5%, respectively. CRP did not significantly predict early onset neonatal sepsis with an AUC of 0.568 (95% CI, 0.410-0.717, $p = 0.306$). The sensitivity and specificity for CRP were 81.8% and 31.8%, respectively. WBC did not significantly predict early onset neonatal sepsis with sensitivity and specificity of 81.8% and 40.9%, respectively. ANC did not significantly predict early-onset neonatal sepsis with sensitivity and specificity of 86.4% and 36.4%, respectively.

In present study, the sensitivity, specificity, PPV, and NPV of WBC were 81.8%, 40.9%, 13.3%, and 95.3%. However, Su H, et al. had demonstrated in their meta-analysis that WBC had variable range of sensitivity and specificity observed in different studies when compared to other markers(11). In present study, the sensitivity and specificity of ANC were 86.4% and 36.4%. Tam P-Y I, et al.(12) Had reported in their review that sensitivity and specificity of ANC in various studies ranged from 0.8%–68% and 95% – 99%, respectively in neonates with early-onset sepsis.

In agreement to the results of this study, Fan Y, et al. had stated superiority of IL-6 and procalcitonin over CRP(13). Similarly, Ruan et al. found the PCT more sensitive than CRP. Ruan et al. also observed that the procalcitonin was more sensitive than CRP in diagnosis of EONS(14). Parallely, Al- Zahrani, et al.(5), proposed that procalcitonin was more accurate than IL-6 and CRP in diagnosis of EONS.

The literature states that there is a difference in specificity and sensitivity of these diagnostic markers when assessed through cord blood and maternal serum sampling. In the meta-analysis, Su H, et al.(11) had amalgamated diagnostic accuracy of procalcitonin, CRP and IL-6 in cord blood and maternal serum samples that were observed in various studies. Su H, et al.(11) had reported equivalent accuracy of IL-6 in maternal serum sample, but higher accuracy of IL-6 was observed in cord blood sample. The 95% confidence interval of sensitivity and specificity of CRP in cord blood sample was 0.71 (0.62–0.79) and 0.71 (0.68–0.74), respectively. Moreover, cord blood procalcitonin had better sensitivity and specificity (sensitivity 0.82, 95% CI: 0.72 to 0.89; specificity 0.86, 95% CI: 0.58 to 0.96). Thus, cord blood sampling can be useful in providing important diagnostic information for making clinical decisions.

On a broader note, many studies suggest use of combination of two or more diagnostic markers for better results rather than relying on single marker. Rashwan, et al.(15) described that combined application of procalcitonin and IL6 could be better in appropriate diagnosis than either of the single markers. Another study also reported that amalgamation of markers like CRP, procalcitonin, and IL-6 provided better accuracy and results than single markers in diagnosing sepsis(5). Moreover, Fan Y, et al.(13) had stated that upon combining with other haematological markers and proper clinical observation, the dependability of procalcitonin, IL-6 and IL-8 could be improved. Hence, further research concentrating on the combination of various umbilical cord biomarkers in diverse clinical settings can be applied to achieve better conclusions.

The study had some limitations, firstly, it was an observational study with small sample size, and secondly due to small sample size certainty that the sensitivity and specificity calculated from the ROC curves becomes difficult to extrapolate to real life population. Thirdly, there was a lack of follow-up to allow a prognostic estimation and face the clinical challenges.

Conclusion

Neonatal sepsis has been one of the main causes of morbidity and mortality in neonates during the initial days of life. Therefore, proper diagnosis of neonatal sepsis, especially the early onset neonatal sepsis is very important. Though blood culturing is the gold standard for diagnosis, the inflammatory markers can assist in diagnosing early onset neonatal sepsis at the earliest. The procalcitonin and IL-6 provide better sensitivity and specificity in diagnosis of early onset neonatal sepsis when compared to CRP, WBC and ANC through cord blood sampling. Combined use of inflammatory markers than using any singe marker for diagnosis of neonatal sepsis is recommended. The timely detection of early onset

neonatal sepsis in neonates will help in the judicious use of antibiotics, thus reducing the morbidity and mortality of neonates.

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Tables

Table 1: Baseline characteristics

Variables	Patients (N = 44)
Mean age (Range)	26.6 ± 5.1 (19-41)
Maternal risk factors	
Fever within 2 weeks prior to delivery	29 (65.9%)
Use of antibiotics	44 (100%)
Premature rupture of membranes	20 (45.5%)
Foul smelling liquor	27 (61.4%)
Prolonged labour (1st+2nd stage >24hours)	19 (43.2%)
Maternal history	
Diabetes mellitus	7 (15.9%)
Hypertension	13 (29.5%)
Hyperthyroidism	3 (6.8%)
Hypothyroidism	4 (9.1%)
TORCH infections	0 (0%)
Clinical features of sepsis	
Lethargy	15 (34.1%)
Hypotonia	7 (15.9%)
Temperature Instability	12 (27.3%)
Respiratory Distress	26 (59.1%)
Feed Intolerance	13 (29.5%)
Shock	6 (13.6%)
Others	
Hypoglycemia	4 (9.1%)
Tachycardia	1 (2.3%)
Mode of delivery	
LSCS	20 (45.5%)
NVD	24 (54.5%)
Gestational age	
Preterm (<37 weeks)	20 (45.5%)
Term (≥37 weeks)	24 (54.5%)
Mean gestational age	36.1 ± 3.2 weeks
Birth weight (Grams)	
< 1500	11
1500-1999	7
2000-2499	9
>2499	17
Mean weight (range)	2214.1 ± 777.3 (1000-3842)
Duration of NICU admission (Days)	
< 15 days	30 (68.2%)
15-30	11 (25%)

≥ 31	3 (6.8%)

Table 2: Results of blood culture, CRP, IL-6, PCT with respect to the isolated organisms from blood

Parameters	Blood culture	CRP (mg/dl) (mean ± SD)	IL-6 (pg/ml) (mean ± SD)	PCT (ng/ml) (mean ± SD)
Staphylococcus haemolyticus	4 neonates	0.6 ± 0.0	181.9 ± 101.2	10.6 ± 4.4
Escherichia coli	5 neonates	0.3 ± 0.4	68.3 ± 7.3	7.3 ± 9.5
Coagulase-positive staphylococcus aureus	3 neonates	0.6 ± 0.6	28.4 ± 19.8	3.3 ± 4.5
Acinetobacter baumannii complex	3 neonates	0.2 ± 0.3	82.7 ± 34.3	5.5 ± 7.4
Pseudomonas aeruginosa	2 neonates	0.4 ± 0.5	72.4 ± 101.1	1.2 ± 1.5
Klebsiella pneumonia	4 neonates	0.6 ± 1.2	91.3 ± 108.5	0.6 ± 0.1
Staphylococcus hominis	2 neonates	0.0 ± 0.0	27.4 ± 26.9	0.6 ± 0.0
Burkholderia caryophylli	1 neonate	0.0 ± 0.0	8.3 ± 0.0	0.6 ± 0.0
Staphylococcus warneri	1 neonate	0.0 ± 0.0	187± 0.0	0.73 ± 0.0

Table 3: Association of maternal risk factors with CRP, IL-6, PCT, WBC and ANC positivity

Parameters	Fever within 2 weeks prior to delivery, n(%)	Premature rupture of membranes, n(%)	Foul smelling liquor, n(%)	Prolonged labour n(%)
Blood culture positive	15/22 (68.2%)	9/22 (40.9%)	11/22 (50%)	10/22 (45.5%)
	P-value = 0.750	P-value = 0.545	P-value = 0.120	P-value = 0.761
CRP positive	9/11 (81.0%)	3/11 (27.3%)	6/11 (54.5%)	5/11 (45.5%)
	P-value = 0.182	P-value = 0.155	P-value = 0.594	P-value = 0.861
Raised IL-6	14/22 (63.6%)	9/22 (40.9%)	11/22 (50%)	11/22 (50%)
	P-value = 0.750	P-value = 0.545	P-value = 0.120	P-value = 0.360
Raised PCT	16/24 (66.7%)	10/24 (41.7%)	12/24 (50%)	11/24 (45.8%)
	P-value = 0.908	P-value = 0.580	P-value = 0.086	P-value = 0.697
Abnormal WBC	17/25 (68%)	10/25 (40%)	14/25 (56%)	11/25 (44%)
	P-value = 0.737	P-value = 0.404	P-value = 0.400	P-value = 0.900
Abnormal ANC	4/7 (57.1%)	3/7 (42.9%)	6/7 (85.7%)	1/7 (14.3%)
	P-value = 0.599	P-value = 0.880	P-value = 0.125	P-value = 0.075

Table 4: Overall summary of sensitivity, specificity, and predictive values of CRP, IL-6, PCT, WBC, and ANC using blood culture as gold standard

Parameters	AUC (95% CI)	P-value	Criterion	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
CRP (mg/dl)	0.568 (0.410-0.717)	0.306	≤0 mg/dl	81.80%	31.80%	11.80%	94.00%
IL-6 (pg/ml)	0.965 (0.861-0.997)	<0.001	≤6.4 pg/ml	90.90%	95.50%	69%	99%
PCT (ng/ml)	0.966 (0.862-0.998)	<0.001	≤0.48 ng/ml	90.90%	100%	100%	99%
WBC/mm^3	0.511 (0.356-0.665)	0.902	≤13,900/ mm^3	81.80%	40.90%	13.30%	95.30%
ANC/mm^3	0.540 (0.384-0.691)	0.66	>2457/mm^3	86.40%	36.40%	13.10%	96%

Figures

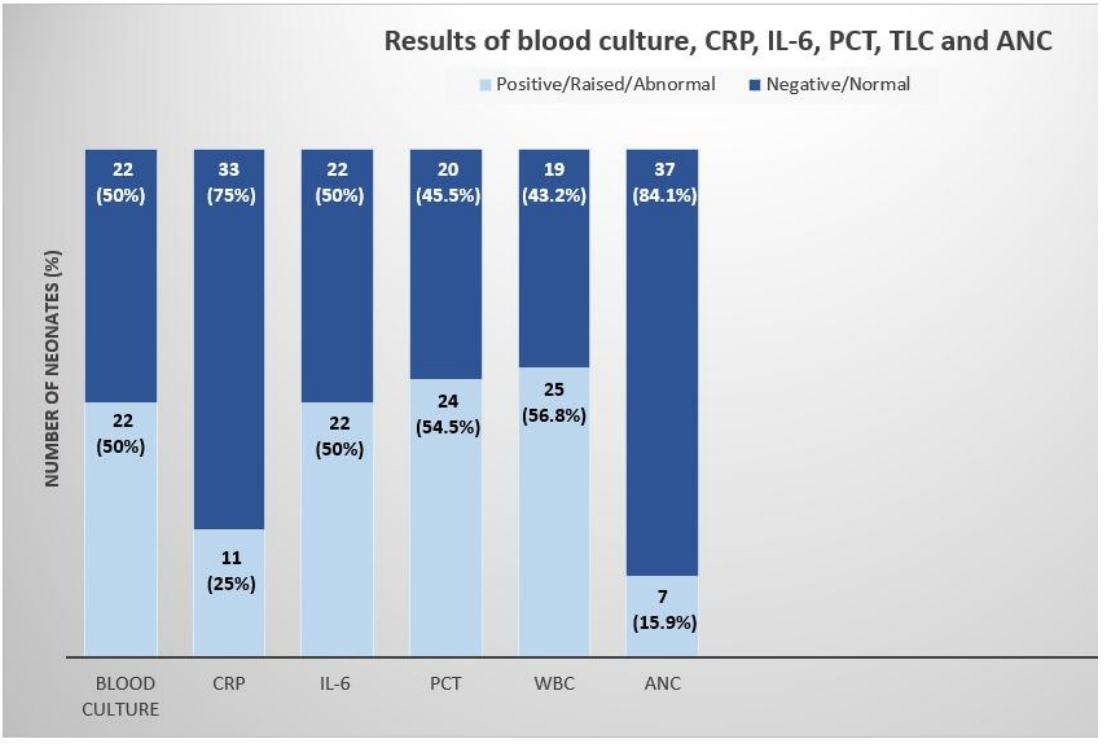

Figure 1: Results of blood culture, CRP, IL-6, PCT, WBC and ANC (N = 44)

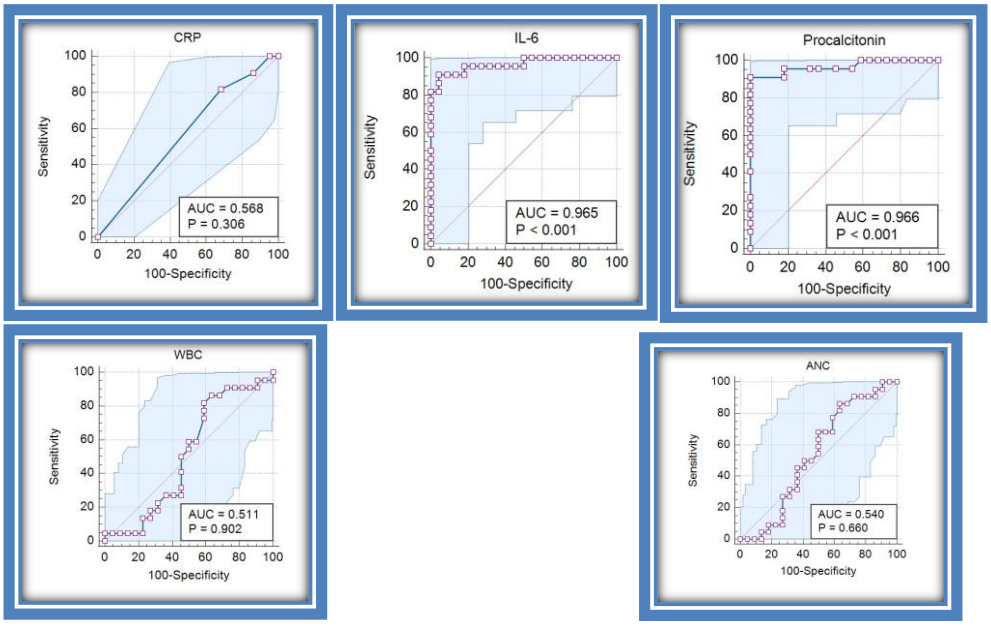

Figure 2: ROC of CRP, IL-6, procalcitonin, WBC, and ANC using blood culture as gold standard