Diagnostic utility of procalcitonin versus C-reactive protein as markers for early-onset neonatal sepsis at Korle-Bu Teaching Hospital

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Research

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Abstract

Introduction: Symptoms of sepsis are non-specific among neonates and diagnosis requires a high index of suspicion. The study sought to evaluate the utility of procalcitonin (PCT) versus C-reactive protein (CRP) in diagnosing early-onset neonatal sepsis. **Methods:** This was a cross-sectional study in which neonates admitted to the neonatal intensive care unit, with signs suggesting sepsis were categorized according to an adapted criteria from Tollner's sepsis score and case definition of bloodstream infection as: "highly probable", "probable" and "less probable". Laboratory investigations including blood culture, complete blood count, PCT and CRP levels were done before first antimicrobial drug administration. **Results:** A total of 62 neonates less than 12 hours postnatal age (0.16-9.82 hours) were recruited. Proportion of neonates with PCT>2 ng/mL was 91% (20/22) in the "highly probable" group compared to 31.6% (6/19) in the "probable group" (p<0.001). Neonates with CRP>5 mg/L was 54.4% (12/22) in the "highly probable" group compared to 26.3% (5/19) in the "probable group" (p = 0.07). The receiver operator characteristics for PCT and CRP were; sensitivity (87.5% vrs 50%), specificity (63.0% vrs 72.2%), positive predictive value (44.1% vrs 37.5%) and negative predictive value (93.8% vrs 81.3%), respectively. **Conclusion:** PCT was a better predictive marker for neonatal sepsis within the first 12 hours of life than CRP in this setting, however, its low specificity relative to CRP suggests that neonates without patent infection are more likely to be incorrectly diagnosed with sepsis using this test.

Introduction

Neonatal sepsis represents an important cause of morbidity and mortality especially in low-resource settings. Early diagnosis and prompt treatment of neonatal sepsis improves outcome [1]. However, the diagnosis of neonatal sepsis is complicated by many factors, including the non-specific clinical symptomatology and absence of specific biomarkers for diagnosis. Although blood culture remains the gold standard for diagnosis, the high false negative rate [2] and delay in obtaining results, places limitations on its clinical utility, thus the need for additional methods and continuous validation for existing markers in neonatal populations. C-reactive protein (CRP), an acute-phase reactant of hepatic origin that increases following secretion of interlukin-6 from macrophages and T-cells [3, 4], is elevated in acute inflammatory conditions, including infection. CRP is a valuable screening marker for the diagnosis of neonatal sepsis [5, 6], however, the time needed for its release, the non-specific response to inflammation, as well as variations in levels depending on physiological state (e.g birth weight or gestational age) [7-10], exerts a limitation on the use of CRP as a marker for neonatal sepsis diagnosis. Furthermore, CRP level has been shown to be low or normal in infants infected with coagulase-negative staphylococcus [11] and the magnitude of the CRP response is known overall to be higher in Gram-negative compared to Gram-positive infections [7].

Procalcitonin (PCT), a pro-hormone of calcitonin, synthesized by thyroid C cells [12] and which level increases markedly in septic conditions [13], is considered by some as a more sensitive marker than CRP in the early identification of bacterial infection in neonates [14, 15]. After exposure to bacterial agents, PCT levels increase within 3-4 hours, while CRP levels increase 12-18 hours later [16]. However, the role of these two markers in diagnosis of neonatal sepsis remain contentious [17, 18]: procalcitonin levels have been shown to vary between preterm and late-preterm neonates [19-21]. In term neonates, PCT demonstrates age-specific variation between the time of birth up to 48 hours [22], suggesting that cut-off levels of this acute-phase reactant should be age-dependent. Nonetheless, the features of physiological changes and reference in various infant populations have not been extensively studied. The incidence of early-onset neonatal sepsis vary between countries and study centers [23]. Also, depending on etiological agents, Gram-positive or Gram-negative bacteria, inflammatory response to infection could vary [24]. The heterogeneity of responses as well as difficulties in adopting a uniform definition for neonatal sepsis underlines the need to generate data from different settings in determining cut-off values of acute-phase reactants in the diagnosis of neonatal sepsis. Early-onset neonatal sepsis is commonly associated with co-morbid conditions but studies conducted to evaluate diagnostic utility of CRP and/or PCT rarely exclude confounders like perinatal asphyxia or meconium aspiration, which could affect predictive accuracy [25-27]. This study investigated the utility of CRP and PCT in neonates with suspected sepsis, excluding possible confounders and compared levels of these acute-phase reactants with clinical data and blood culture results.

Methods

Study design and site: This was a cross-sectional study, a sub-component of a larger study that evaluated the disposition of amikacin in neonates with suspected sepsis at the Neonatal Intensive Care Unit (NICU), Korle-Bu Teaching Hospital, Accra, Ghana, from November 2013 to June 2014. The full details of the larger study has been reported elsewhere [**28**]. The NICU is a 55-bed tertiary unit that admits annually an average of 2000 sick and preterm neonates mainly from southern Ghana. The NICU restricts admission to neonates with postnatal age less than 48 hours, unless in exceptional situations.

Criteria for inclusion and exclusion: Admitted neonates that fulfilled criteria (described below) were purposely selected. Eligible neonates were categorized as; "highly probable", "probable" and "less probable" neonatal sepsis, based on an adapted criteria from Tollner's sepsis score [29] and case definition of bloodstream infection by Vergnano et al [30] (Table 1).

Inclusion: neonates with maternal risk factors (prolonged rupture of amniotic membrane >18 hrs, chorioamnionitis), neonatal risk factors (low birth weight and premature birth) and clinical symptoms (feeding intolerance, lethargy, temperature instability, tachypnea, bradycardia, tachycardia, abdominal distension or vomiting) and deemed by the admitting physician to have a presumptive diagnosis of sepsis, were enrolled.

Exclusion: neonates presenting with meconium aspiration, perinatal asphyxia or neonates who required resuscitation for any reason, were excluded.

Blood sampling and laboratory analysis: After initial examination by study pediatrician and before first antimicrobial drug dose, blood samples were taken from recruited neonates, for the following investigations: blood culture (1-2 mL), full blood count (0.5 mL), CRP and PCT (0.5 mL). The sample for culture was collected into pediatric culture vials (BACTEC Peds plus/F, Becton-Dickinson, Gauteng, South Africa) and those for full blood count, CRP and PCT analysis into EDTA and gel microtainer tubes, respectively. The samples for measurement of CRP and PCT levels were immediately centrifuged to obtain serum. Blood culture was done using a fully automated BACTEC 9240 blood culture system (Becton Dickinson Diagnostic Instrument Systems, Sparks, Maryland). Isolates from positive bottles were sub-cultivated and identified using biochemical methods. Briefly, Gram-positive bacteria were identified by catalase, slide and tube coagulase test and Gram-negative by API 20E and 20NE (BioMerieux, France). Antibiotic susceptibility tests were done using the disc diffusion method on Mueller-Hinton agar (Oxoid, UK), in accordance with Clinical Laboratory Standards Institute (CLSI) criteria. Complete blood count was done by means of a Sysmex Autoanalyzer (Sysmex KX-21N, Sysmex Corporation, Kobe, Japan). CRP assay was performed with a BNII automated system (Dade-Behring Inc, Newark, Delaware), according to manufacturer's instructions. The assay uses particle-enhanced immunonephelometry to quantify CRP in serum samples [31]. Limit of detection of assay was 0.17 mg/L. Electrochemiluminescence immunoassay, which is based on a sandwich principle [32], was used to analyze PCT in serum of neonates. An automated Elecsys (Roche Diagnostics, Rotkreuz, Switzerland) was used for this purpose according to manufacturer's instructions. Limit of detection of assay was 0.1 ng/mL.

Ethical issues: Approval of this research was from the Ethical and Protocol Review Committee of the School of Medicine and Dentistry, University of Ghana (Protocol ID: MS-Et/M.8-P.5.3/2011-2012). Written informed consent was obtained from parents of all recruited neonates.

Statistical analysis: One-way ANOVA was used to compare the means of selected clinical and laboratory parameters (PCT and CRP levels) between the "highly probable", "probable" and "less

probable" neonatal sepsis groups. Chi-square was used to compare proportions of neonates with elevated PCT and CRP among the groups. Blood culture was used as the gold standard for receiver operator characteristics (ROC) of PCT and CRP. Area under the ROC curve (AUC) for PCT and CRP were compared using Chi-square. A p-value <0.05 was considered statistically significant.

Results

Patient characteristics and outcome: A total of 62 neonates fulfilled criteria for inclusion and were categorized as: highly probable (22 neonates), probable (19 neonates) and less probable (21 neonates) sepsis. All neonates were recruited within 12 hours after birth (0.16-9.82 hours) and prior to first antimicrobial drug dose. Selected baseline demographic and clinical parameters are shown in Table 2. The overall mortality was 9.7% (n=6) and case fatality per group was 13.6% (n=3), 10.5% (n=2) and 4.8% (n=1) in the highly probable, probable and less probable groups, respectively. All neonates who died were less than 34 weeks gestational age and weighed less than 1500 grams.

PCT and CRP levels between groups: Mean PCT levels were significantly different between the three groups. The PCT levels followed the order: "highly probable" > "probable" > "less probable" groups (Table 2). Mean CRP levels showed a similar trend, however, this difference did not attain statistical significance (p = 0.08). The PCT levels were elevated (>2 ng/mL) in 91% (20/22) of neonates in the highly probable group compared to 31.6% (6/19) in the probable group (p<0.001). The proportion of neonates with elevated (>5 mg/L) CRP levels was higher in the highly probable group (54.5%; 12/22) compared to probable (26.3%; 5/19), but the difference (p=0.07) showed only a trend towards but did not attain statistical significance.

Positive blood culture and sepsis risk score: In all, 36.4% (8/22) of neonates in the "highly probable" group had positive blood cultures. Among the "highly probable" neonatal sepsis risk group, there was no statistically significant difference in mean (SD) PCT (4.3 (2.4) ng/mL versus 4.1 (0.6) ng/mL; p = 0.9) and CRP (11.3 (21.7) mg/L versus 5.2 (1.2) mg/L; p = 0.3) between those with positive blood cultures and those with negative blood cultures respectively. Overall, CRP levels were more reflective of blood culture positivity even though the difference was not statistically significant. Clinical characteristics, PCT and CRP levels of neonates with positive blood culture are shown in Table 3.

Diagnostic accuracy of PCT and CRP: The receiver operator characteristic curves (ROCs) of PCT and CRP are shown in Figure 1. The area under the receiver operating characteristic curve (AUC) for PCT was 0.646 compared to that of 0.569 for CRP (p = 0.09) and these were at optimal thresholds of 2 ng/mL and 5 mg/L for PCT and CRP, respectively. Using the above thresholds, the sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) of PCT and CRP were: 87.5% versus 50.0%, 63.0% versus 72.2%, 44.1% versus 37.5% and 93.8% versus 81.3%, respectively.

Discussion

Diagnosis of neonatal sepsis remains a difficult task due to the non-specific nature of clinical signs and symptoms in this age group. As sepsis remains a major cause of neonatal deaths, the standard of care includes the use of empiric antibiotic treatment on wide indications. As a result, the fear of missing a case of neonatal

infection invariably leads to an overuse of antibiotics at NICUs. To ensure antimicrobial accountability while reducing neonatal deaths, there is the need for newer, faster and reliable markers of infection, and continuous validation of existing ones. In the current study, the proportion of neonates with elevated PCT in the highly probable sepsis group was significantly different compared with the probable sepsis group. Generally, serum PCT levels begin to rise 4 hours after exposure to bacterial endotoxin, peaks at 6-8 hours, and remain raised for at least 24 hours [33]. On the other hand, CRP levels start to increase 4 to 6 hours later than PCT and reaches peak 36 hours later [34]. Thus, the high proportion of neonates in the highly probable sepsis group with elevated PCT, relative to CRP, is consistent with the kinetics of these biomarkers after exposure to bacterial agents [33, 34]. PCT levels greater than 10 ng/mL have been associated with high mortality [35], however, the PCT levels of all neonates who died in this study were less than 10 ng/mL. A possible reason for this observation could be the fact that all neonates who died were delivered preterm [36].

The higher AUC of the ROC curve for PCT compared to that of CRP is consistent with studies [37, 38], on early-onset neonatal sepsis. In contrast Park et al [39], studied newborns with postnatal age ranging between 4 and 30 days and reported a lower AUC (0.803) of PCT compared to CRP (0.951). These dissimilarities could be due to differences in postnatal age and the kinetics of these biomarkers. PCT showed higher sensitivity, PPV and NPV compared to CRP in this study. These results, together with others [37, 38, 40], suggest that PCT is a more sensitive marker for sepsis diagnosis within the first 24 hours of life. The utility of the high NPV of PCT in this study may be its potential in excluding neonates without sepsis. This may be helpful in decision-making about early discontinuation of antibiotics in neonates without strong clinical indicators of sepsis. PCT is a relatively more reliable diagnostic marker for diagnosis of early-onset neonatal sepsis, the cost of the test precludes its routine use in clinical management of sepsis [41]. Therefore, a cost benefit analysis is recommended if PCT is to be used as diagnostic marker, alone or in combination with others, for sepsis at NICUs, especially in low-resource settings. A limitation of this study is the weakness inherent in the adapted criteria used for sepsis classification. One example is the criteria based on tachypnea (rate >60 cpm), as most neonates born preterm (especially <34 weeks gestation) have some degree of respiratory distress syndrome, which can manifest with tachypnea in the absence of sepsis. The use of blood culture in the sepsis definition in this age group is another limitation because of its low sensitivity given the limited volume of blood used for the test.

Conclusion

In summary, this study has shown that within the first 12 hours of life, PCT is a more reliable acute phase reactant in diagnosing neonates with sepsis than CRP. The use of PCT during the first 12 hours of life may limit number of neonates started on antibiotics due to suspected risk of sepsis when there are no strong clinical indicators of illness. Limiting unnecessary use of antibiotics in NICUs in low-resource settings will improve efficiency in clinical care and decrease the rising trend of antimicrobial resistance.

What is known about this topic

The role of PCT and CRP, in the diagnosis of neonatal sepsis remain controversial;
CRP is a valuable screening marker for the diagnosis of neonatal sepsis, however, there are variations in levels depending on physiological state of neonate (e.g birth weight or gestational age).

What this study adds

Within the first 12 hours of life, PCT is a more reliable acute phase reactant in diagnosing neonates with sepsis than CRP in this cohort of neonates;
Although PCT has low specificity (relative to CRP), it may improve antimicrobial use accountability in low-income countries as clinicians would treat fewer newborns without sepsis than current practice where most newborns in NICUs are given antibiotics.

Tables and Figure

Table 1: Criteria for classification based on Tollner's sepsis score and case definition of bloodstream infection
Table 2: Characteristics [mean (SD)] of recruited neonates with clinical suspicion of sepsis
Table 3: Selected characteristics of neonates with positive blood culture in the highly probable sepsis group
Figure 1: Receiver operator characteristic curves of PCT and CRP

References

1. Shah BA, Padbury JF. Neonatal sepsis: an old problem with new insights. Virulence. 2014; 5(1): 170-178.**PubMed** | **Google Scholar**

2. Brown DR, Kutler D, Rai B, Chan T, Cohen M. Bacterial concentration and blood volume required for a positive blood culture. J Perinatol. 1995; 15(2): 157-159. **PubMed** | **Google Scholar**

3. Worthman H, Tryc A, Dirks M, Schuppner R, Brand K, Klawonn F, Lichtinghagen R,Weissenborn K. Lipopolysaccharide binding protein, interleukin-10, interleukin 6 and C-reactive protein blood levels in acute ischemic stroke patients with post-stroke infection. J Neuroinflammation. 2015; 12: 13. **PubMed** |**Google Scholar**

4. Cekic C, Arabul M, Alper E, Pakoz ZB, Saritas E, Yuksel, Unsal B. Evaluation of the relationship between serum ghrelin, C-reactive protein and interleukin-6 levels, and disease activity in inflammatory bowel diseases. Hepatogastroenterology. 2014; 61 (133): 1196-1200. **PubMed** | **Google Scholar**

5. Chan DK, Ho LY. Usefulness of C-reactive protein in the diagnosis of neonatal sepsis. Singapore Med J. 1997; 38(6): 252-255. **PubMed** | **Google Scholar**

6. Philip AG, Mills PC. Use of C-reactive protein in minimizing antibiotic exposure: experience with infants initially admitted to a well-baby nursery. Pediatrics. 2000; 106(1): E4. **PubMed** | **Google Scholar**

7. Hofer N, Zacharias E, Muller W, Resch B. An update on the use of C-reactive protein in early-onset neonatal sepsis: current insights and new tasks. Neonatology. 2012; 102 (1): 25-36. **PubMed** |**Google Scholar**

8. Mussap M. Laboratory medicine in neonatal sepsis and inflammation. J Matern Fetal Neonatal Med. 2012; 25(Suppl 4): 32-34. **PubMed** | **Google Scholar**

9. Chiesa C, Signore F, Assumma M, Buffone E, Tramontozzi P, Osborn JF, Pacifico L. Serial measurements of C-reactive protein and interleukin-6 in the immediate postnatal period: reference intervals and analysis of maternal and perinatal confounders. Clin Chem. 2001; 47 (6): 1016-1122. **PubMed** | **Google Scholar**

10. Dammann O, Phillips TM, Allred EN, O'Shea TM, Paneth N, Van Marter LJ, Bose C, Ehrenkranz RA, Bednarek FJ, Naples M, Leviton A. Elgan study investigators, Mediators of fetal inflammation in extremely low gestational age newborns. Cytokine. 2001; 13(4): 234-239. **PubMed** | **Google Scholar**

11. Lai MY, Tsai MH, Lee CW, Chiang MC, Lien R, Fu RH, Huang HR, Chu SM, Hsu JF. Characteristics of neonates with culture-proven bloodstream infection who have low levels of C-reactive protein (≤10 mg/L). BMC Infect Dis. 2015; 15: 320. **PubMed** | **Google Scholar**

12. Maruna P, Nedelnikova K, Gurlich R. Physiology and genetics of procalcitonin. Physiol Res. 2000; 49(Suppl1): S57-S61. **PubMed** | **Google Scholar**

13. Gendrel D, Bohuon C. Procalcitonin as a marker of bacterial infection. Pediatr Infect Dis J. 2000; 19(8): 679-687. **PubMed** | **Google Scholar**

14. Yu Z, Liu J, Sun Q, Qiu Y, Han S, Guo X. The accuracy of the procalcitonin test for the diagnosis of neonatal sepsis: a meta-analysis. Scand J Infect Dis. 2010; 42(10): 723-733. **PubMed** | **Google Scholar**

15. Aikawa N, Fujishima S, Endo S, Sekine I, Kogawa K, Yamamoto Y, Kushimoto S, Yukioka H, Kato N, Totsuka K, Kikuchi K, Ikeda T, Ikeda K, Harada K, Satomura S. Multicenter prospective study of procalcitonin as an indicator of sepsis. J Infect Chemother. 2005; 11(3): 152-159. **PubMed** | **Google Scholar**

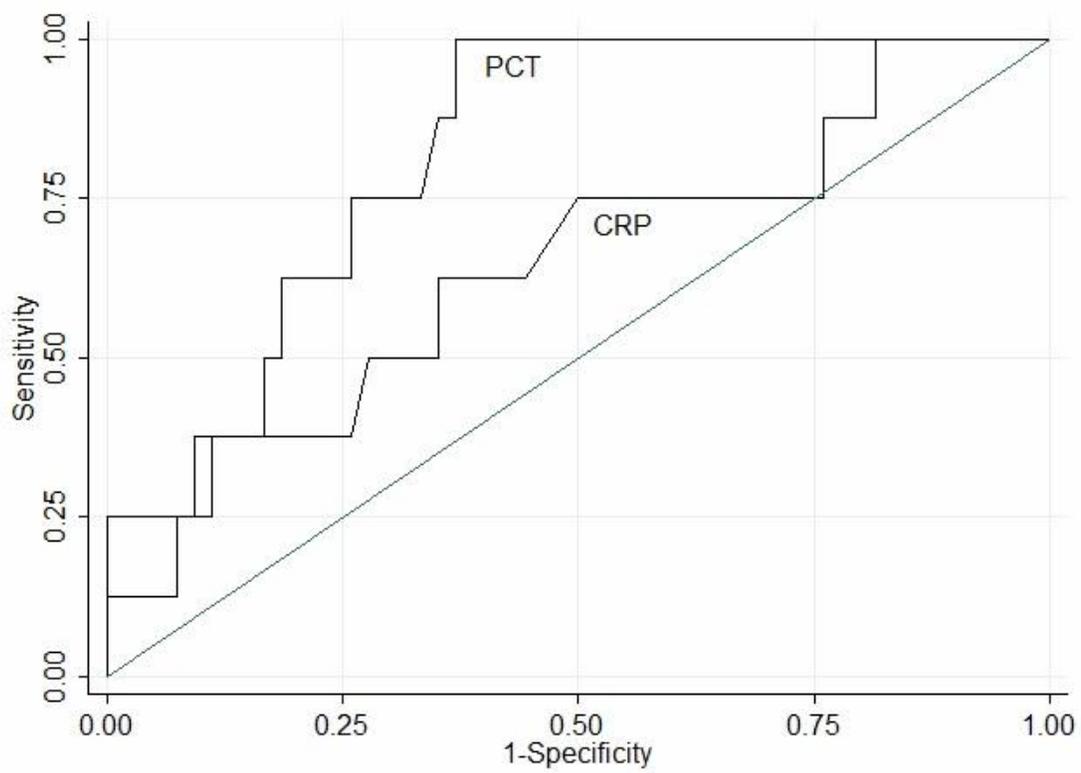16. Monneret G, Labaune JM, Isaac C, Bienvenu F, Putet G, Binevenu J. Procalcitonin and C-reactive protein levels in

neonatal infections. Acta Paediatr. 1997; 86(2): 209-212. **PubMed** | **Google Scholar**

17. Nahar BS, Mannan MA, Noor K, Shahiddullah M. Role of serum procalcitonin and C-reactive protein in the diagnosis of neonatal sepsis. Bang Med Res Counc Bull. 2011; 37(2): 40-46. **PubMed** | **Google Scholar**

18. Janota J, Stranak Z, Belohlavkova S, Mudra K, Simak J. Postnatal increase of procalcitonin in premature newborns is enhanced by chorioamnionitis and neonatal sepsis. Eur J Clin Invest. 2001; 31(11): 978-983.**PubMed** | **Google Scholar**

19. Turner D, Hammerman C, Rudensky B, Schlesinger Y, Goia C, Schimmel MS. Procalcitonin in preterm infants during the first few days of life: introducing an age related nomogram. Arch Dis Child Fetal Neonatal Ed. 2006; 91 (4): F283-286. **PubMed** | **Google Scholar**

20. Lopez Sastre JB, Solís DP, Serradilla VR, Colomer BF, Cotallo GD. Grupo de Hospitales Castrillo, Evaluation of procalcitonin for diagnosis of neonatal sepsis of vertical transmission. BMC Pediatr. 2007; 7: 9.**PubMed** | **Google Scholar**

21. Chiesa C, Natale F, Pascone R, Osborn JF, Pacifico L, Bonci E, De Curtis M. C reactive protein and procalcitonin: reference intervals for preterm and term newborns during the early neonatal period. Clin Chim Acta. 2011; 412 (11-12): 1053-1059. **PubMed** | **Google Scholar**

22. Chiesa C, Panero A, Rossi N, Stegagno M, De Giusti M, Osborn JF, Pacifico L. Reliability of procalcitonin concentrations for the diagnosis of sepsis in critically ill neonates. Clin Infect Dis. 1998; 26 (3): 664-672.**PubMed** | **Google Scholar**

23. Seale A, Mwaniki M, Newton CR, Berkley JA. Maternal and early onset neonatal bacterial sepsis: burden and strategies for prevention in sub-Saharan Africa. Lancet Infect Dis. 2009; 9(7): 428-438. **PubMed** |**Google Scholar**

24. Opal SM, Cohen J. Clinical gram-positive sepsis: does it fundamentally differ from gram-negative bacterial sepsis. Crit Care Med. 1999; 27(8): 1608-1616. **PubMed** | **Google Scholar**

25. Monneret G, Labaune JM, Isaac C, Bienvenu F, Putet G, Bienvenu J. Increased serum procalcitonin levels are not specific to sepsis in neonates. Clin Infect Dis. 1998; 27(6): 1559-1561. **PubMed** | **Google Scholar**

26. Pourcyrous M, Bada HS, Korones SB, Baselski V, Wong SP. Significance of serial Creactive protein responses in neonatal infection and other disorders. Pediatrics. 1993; 92(3): 431-435. **PubMed** |**Google Scholar**

27. Ainbender E, Cabatu EE, Guzman DM, Sweet AY. Serum C-reactive protein and problems of newborn infants. J Pediatr. 1982; 101(3): 438-440. **PubMed** | **Google Scholar**

28. Amponsah SK, Adjei GO, Enweronu-Laryea C, Bugyei KA, Hadji-Popovski K, Kurtzhals JA, Kristensen K. **Population pharmacokinetic characteristics of amikacin in suspected cases of neonatal sepsis in a low-resource African setting: a prospective non-randomized single-site study. Curr Ther Res. 2017**. (Accessed 14/4/2017)

29. Tollner U. Early diagnosis of septicemia in the newborn: clinical studies and sepsis score. Eur J Pediatr. 1982; 138(4): 331-337. **PubMed** | **Google Scholar**

30. Vergnano S, Buttery J, Cailes B, Chandrasekaran R, Chiappini E, Clark E, et al,. Neonatal infections: Case definition and guidelines for data collection, analysis, and presentation of immunisation safety data. Vaccine. 2016 Dec 1; 34(49): 6038-6046. **PubMed** | **Google Scholar**

31. De BK, Smith LG, Owen WE, and Roberts WL. Performance characteristics of an automated high-sensitivity C-reactive protein assay on the Dimension RxL analyzer. Clin Chim Acta. 2002; 323(1-2): 151-155. **PubMed** | **Google Scholar**

32. Sano M, Tatsumi N. Electro chemiluminescence immunoassay. Rinsho Byori. 1996; 44(11): 1076-1079.**PubMed** | **Google Scholar**

33. Ng PC. Diagnostic markers of infection in neonates. Arch Dis Child Fetal Neonatal Ed. 2004; 89(3): F229-235. **PubMed** | **Google Scholar**

34. Meisner M. Pathobiochemistry and clinical use of procalcitonin. Clin Chim Acta. 2002; 323(1-20): 17-29.**PubMed** | **Google Scholar**

35. Ali AM, Walid FE, Abdelaziz SS. Procalcitonin versus C-reactive protein in neonatal sepsis. J Immunol Infect Dis. 2014; 1(1): 103. **PubMed** | **Google Scholar**

36. Lemons JA, Bauer CR, Oh W, Korones SB, Papile LA, Stoll BJ, Verter J, Temprosa M, Wright LL, Ehrenkranz RA, Fanaroff AA, Stark A, Carlo W, Tyson JE, Donovan EF, Shankaran S, Stevenson DK. Very low birth weight outcomes of the National Institute of Child health and human development neonatal research network, January 1995 through December 1996, NICHD Neonatal Research Network. Pediatrics. 2001; 107(1): E1. **PubMed** | **Google Scholar**

37. Bonac B, Derganc M, Wraber B, Hojker S. Interleukin-8 and procalcitonin in early diagnosis of early severe bacterial infection in critically ill neonates. Pflügers Arch-Eur J Physiol. 2000; 440(5 Suppl): R72-4.**PubMed** | **Google Scholar**

38. Koskal N, Harmanci R, Cetinkaya M, HAcimustafaoglu M. Role of procalcitonin and CRP in diagnosis and follow-up of neonatal sepsis. Turk J Pediatr. 2007; 49(1): 21-29. **PubMed** | **Google Scholar**

39. Park IH, Lee SH, Yu ST, Oh YK. Serum procalcitonin as a diagnostic marker of neonatal sepsis. Korean J Pediatr. 2014; 57(10): 4451-456. **PubMed** | **Google Scholar**

40. Altunhan H, Annagur A, Ors R, Mehmetoglu I. Procalcitonin measurement at 24 hours of age may be helpful in the prompt diagnosis of early-onset neonatal sepsis. Int J Infect Dis. 2011; 15(12): e854-e858.**PubMed** | **Google Scholar**

41. Kim EK, Lee BS, Lee JA, Jo HS, Park JD, Kim BI et al. Clinical availability of serum procalcitonin level in the diagnosis of neonatal bacterial infection. J Korean Soc Neonatol. 2001; 8: 211-221. **Google Scholar**

Table 1: Criteria for classification based on Tollner's sepsis score and case definition of bloodstream infection	
I	Positive blood culture (pathogenic microorganism)
II	Negative blood culture
III	Maternal risk (prolonged premature rapture of membranes > 18 hours)
IV	Tachycardia (Heart rate > 160 bpm) or Bradycardia (Heart rate < 100 bpm)
V	Tachypnea (Rate > 60 cpm)
VI	Leukopenia (WBC < 5 x 10^9/L) or Leukocytosis (WBC > 25 x 10^9/L)
VII	Thrombocytopenia (platelet < 150 x 10^9/L)
Highly probable (Group 1): I or II, plus > 3 of remaining criteria	
Probable (Group 2): II, plus 2 or 3 of remaining criteria (I excluded)	
Less probable (Group 3): II, plus < 2 of remaining criteria (I excluded)	

Table 2: Characteristics [mean (SD)] of recruited neonates with clinical suspicion of sepsis

Characteristic	Highly probable (n = 22)	Probable (n = 19)	Less probable (n = 21)	*p-value*
Gestational age (wks)	33.2 (4.3)	32.5 (4.2)	34.8 (4.3)	0.2
Birth weight (kg)	2.1 (0.9)	1.7 (0.7)	2.4 (1.1)	0.08
Sampling time postnatal hours	5.5 (3.3)	4.1 (2.8)	6. (2.2)	0.1
PCT (ng/mL)	4.2 (2.2)	1.5 (1.2)	0.8 (0.6)	<0.001
CRP (mg/L)	7.4 (13.4)	2.9 (3.9)	1.9 (1.8)	0.08

Table 3: Selected characteristics of neonates with positive blood culture in the highly probable sepsis group

N	Isolate	GA (weeks)	BW (kg)	PNA (hours)	CRP (mg/L)	PCT (ng/mL)
1	*Burkholderia cepacia*	26	1.1	1.3	0.6	2.3
2	*Klebsiella pneumonia 2*	26	0.9	1	2.5	3.1
3	*Streptococcus viridians*	30	1.7	0.9	1.6	8.1
4	*Escherichia coli*	31	1.5	3.2	0.4	1.9
5	*Streptococcus agalactiae 2*	33	1.9	0.4	64.5	2.5
6	*Klebsiella pneumonia 1*	33	1.6	0.9	5.2	7.5
7	*Acinetobacter baumannii*	38	3.2	3.3	7.1	3.7
8	*Streptococcus agalactiae 1*	38	3.2	2.3	8.1	4.9
GA = Gestational age, BW = Birth weight, PNA = Postnatal age						

Figure 1: Receiver operator characteristic curves of PCT and CRP