

PROMISING DIAGNOSTIC MARKER AT NICU AND PICU-NEW APPROACHES FOR DIAGNOSTIC AND TREATMENT

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Abstract

The aim of this study is evaluate the initial PCT levels on the outcome of patients in pediatric intensive care units and find out if these biomarker can be used to predict sepsis.

The study was designed as a prospective, clinical, investigation conducted in the period sixth months, which included 45 (M:F=25:20) newborns with two or three clinical signs of sepsis hospitalized in the Intensive Care Unit at the PHI University Clinic for Children Diseases. The patient have been divided into two groups: I group included 31 septic newborns with negative blood culture and II group - 14 septic newborns with positive blood cultures. Results of blood count (WBC), CRP and PCT, were recorded.

Procalcitonin PCT levels at first 24 hours of the admission were increased in all 45 newborns (≥ 2 ng/mL). The values of C-reactive protein gradually increase after 12-36 hours at admission. The second measurement, after 3 days usage of an adequate antibiotic treatment, the levels of PCT is decreased, regardless of whether blood culture is positive or negative, except 5 patients develop severe sepsis, and three patients develop septic shock. After the third measurement the levels of PCT and CRP is decreased. NIV was used in 39.8% patients and Invasive MV was used in 29.8% patients. Sensitivity of procalcitonin 83.5%, Specificity of procalcitonin 81.3%.

Procalcitonin value is a early prognostic factor for sepsis and it is a reliable parameter whether an appropriate antibiotic for the treatment is used, thus increasing newborns safety, and reducing costs .

Key words: sepsis, C-reactive protein, procalcitonin PCT

Introduction

For treating pediatric patients urgently and effectively in pediatric intensive care units it is very important to determine the severity and the prognostic factors of underlying disease. Procalcitonin (PCT)-biochemical markers is an endogen peptide secreted in the C cells of thyroid tissue, is commonly used in the early diagnosis of sepsis and infectious diseases. Procalcitonin (PCT) is a precursor of the calcitonin hormone (CT). It is a 116 amino acid peptide with an approximate molecular weight of 14.5 kDa. The structure of procalcitonin can be divided into three parts: amino terminus (57 amino acids), immature calcitonin (33 amino acids), since the crystalline structure of procalcitonin is not yet available), and calcitonin carboxyl-terminal 1 (CC) known as catacalcin (21 amino acids). Its production is regulated by the calcitonin 1 gene (CALC-1) on chromosome 11. Production of this gene, prePCT, is subject to proteolytic cleavage that produces PCT, which is further processed in mature calcitonin molecules [1,2]. Transcription and translation of the CALC-1 gene is normally confined to thyroid S-cells and, to a lesser extent, other neuroendocrine cells. However, production is activated in all parenchymal tissues in response to bacterial infection mediated by cytokines interleukin-6 (IL-6), tumor necrosis factor- α (TNF- α) and interleukin-1 β (IL- β). These other tissues lack the ability to adhere PCT to its mature form, calcitonin, which leads to PCT accumulation [3, 4]. In contrast, PCT production is attenuated by interferon- γ primarily secreted in response to viral infection. This feature makes PCT a more specific marker for bacterial infection [5].

In healthy individuals the level of PCT is below 0.05 ng / ml. Concentrations of procalcitonin in the blood increase with systemic inflammation, especially when it is caused by a bacterial infection [6]. The risk of local bacterial infection occurs when the procalcitonin value exceeds 0.25 ng / ml. The risk of systemic bacterial infection occurs when procalcitonin values exceed 0.5 ng / ml, high procalcitonin values are present in severe sepsis and septic shock [7]. Bacterial lipopolysaccharide (LPS) has been shown to be a potent inducer of procalcitonin release in systemic circulation. Procalcitonin is detected in the body only 3 to 4 hours after infection, reaches its peak after 6 hours, and then plateau for up to 24 hours. In contrast, S-reactive protein (CRP) levels increase between 12 and 36 hours after bacterial infection. In the blood, procalcitonin has a half-life of between 25 and 30 hours[8].

Sepsis is a leading cause of death in the pediatric age, characterized by severe systemic inflammation, tissue damage and impaired systemic response of the organism to infection [9]. Pediatric sepsis is thought to be a life-threatening condition caused by microorganisms, which consists of a range of disorders, with the immune system instead of controlling the infection causing damage to its own tissues and organs [10]. Early recognition and treatment significantly improve outcomes in pediatric patients with sepsis infections. Initial treatment should lead to stabilization and correction of metabolic, circulatory and respiratory disorders. The incidence of sepsis, severe sepsis and septic shock continues to increase, and although Gram-positive bacterial pathogens remain the most common cause of sepsis, fungal microorganisms are on the rise [11]. Significant progress has been made in the diagnosis and treatment of patients with sepsis over the past half-century, while decreasing mortality rates. However, due to the increasing incidence of sepsis, the number of people dying every year continues to increase, 10% -20% of sepsis, 20% -50% of severe sepsis and 40% -80% of septic shock [12].

Diagnosis of infection caused by bacteria or other microbiological organisms is essential for effective treatment and prognostic evaluation. Current clinical and laboratory methods for diagnosing bacterial infections are either nonspecific or require a longer time to develop the agent. Procalcitonin (PCT) is a biomarker that is more specific than other proinflammatory markers in identifying pediatric patients with sepsis and can be used to diagnose bacterial infections.

Procalcitonin (PCT), an early marker for the diagnosis of sepsis in pediatric patients. Specificity and sensitivity of procalcitonin to correlate with other classic inflammatory markers (WBC, Neutr%, CRP) and hemoculture as a gold standard for infection surveillance are required. The dynamics of procalcitonin concentration also determine the duration of antibiotic therapy or its modification [13].

Methods

The study is designed as prospective study, which is done at the PHI University Clinic for Children Diseases - Skopje. Diagnosis of sepsis in pediatric patients diagnosed according to standard protocols for diagnosis of disease.

Sepsis was defined as the identification of two or more Systemic Inflammatory Response Syndrome criteria in addition to known or suspected infection. SIRS criteria are Temperature $>38^{\circ}\text{C}$ or $<36^{\circ}\text{C}$, Heart rate $>90/\text{min}$, Respiratory rate $>20/\text{min}$ or $\text{PaCO}_2 <32 \text{ mm Hg}$ (4.3 kPa) White blood cell count $>12\,000/\text{mm}^3$ or $<4000/\text{mm}^3$ or $>10\%$ immature bands. Severe sepsis was defined as clinical sepsis accompanied by organ dysfunction, hypoperfusion or hypotension. Severe sepsis criteria are Hypotension from sepsis (systolic blood pressure $<90 \text{ mm Hg}$, mean arterial pressure $<70 \text{ mm Hg}$, or a decrease in SBP by $>40 \text{ mm Hg}$), Elevated lactate (above upper limit of normal), Decreased urine output ($<0.5 \text{ ml/kg/hr}$ for more than 2 hours despite fluid resuscitation), Acute lung injury / ARDS (if no pneumonia present: $\text{PaO}_2/\text{FiO}_2 <250$; if pneumonia present: $\text{PaO}_2/\text{FiO}_2 <200$), Creatinine $>2 \text{ mg/dL}$, Total Bilirubin $>2 \text{ mg/dL}$, Thrombocytopenia (platelet count $<100,000$), Coagulopathy (INR >1.5). Septic shock was defined as sepsis-induced hypotension (average artery blood pressure $\leq 70 \text{ mmHg}$) persisting despite adequate fluid resuscitation. [14,15].

Procalcitonin (PCT and other inflammatory markers (WBC, Neutr%, CRP) are employed in the Clinical Laboratory at the PHI University Clinic of Pediatric Diseases-Skopje

Hemoculture The gold standard in the diagnosis of sepsis and the Filmarray method for identifying positive hemoculture was also worked in the Clinical Laboratory at the PHI University Pediatric Disease Clinic - Skopje.

Hemoculture was obtained prior to initiation of antibiotic therapy, and the Film Array method for the identification of positive hemoculture was used to obtain the cause of sepsis. the third measurement is growing, and at febrile.

Procalcitonin was determined by immunoassay: patented ELFA (Enzyme-linked fluorescent assay) technology, automated Vidas Biomerieux immunoassay (ng / ml). In healthy individuals the level of PCT is below 0.05 ng / ml . Sepsis occurs when the procalcitonin value exceeds 0.5 ng / ml , high procalcitonin values above 2 ng / ml are found in severe sepsis and septic shock.

The first sample of procalcitonin was taken during the first 24 hours of sign of infection.

A second sample of procalcitonin for 3-5 days and antibiotic treatment initiated after anamnesis, clinical finding and increased values of PCT, and other inflammatory markers

Third sample of procalcitonin of 6-14 days.

Statistic method

The material was statistically analyzed using the methods of descriptive statistics. To determine the significance of differences in the parameters, the tests for independent samples were analyzed. Statistical significance was determined for the values of $p < 0.05$.

SPSS program was used for statistical analysis. Continuous variables are expressed as mean \pm standard deviation, and categorical variables are expressed as frequency. The relationship between PCT, CRP, and leucocyte values was determined with receiver operating characteristic (ROC) curves. For all tests, $p < 0.05$. was considered to be statistically significant.

Results

In The study was designed as a retrospective and prospective, we included 45 (M:F=25:20) newborns with two or three clinical signs of sepsis hospitalized in the Intensive Care Unit at the PHI University Clinic for Children Diseases - Skopje. The frequent diagnoses of pediatric patients with sepsis were respiratory disorders (41.2%) and the others were asphyxia and neurological disease. The mean duration of stay in PICU was 15.56 ± 10.51 days. NIV was used in 39.8% patients and Invasive MV was used in 29.8% patients. Analysis of ROC curves of correlation between sepsis and biomarkers are showed in Table 1. PCT at a cut-off value of 6.41 ng/dl has a sensitivity of 83.5% and a specificity of 81.3% (area under curve 0.829).

Parameter	Area under curve	P value	Cutoff value	Sensitivity (%)	Specificity (%)
Procalcitonin (ng/ml)	0.829	≤0.001	6.41	83.5	81.3
C-reactive protein (ml/L)	0.535	0.137	163.00	61.2	59.0
Leucocyte (mm3)	0.412	0.410	32.15	50.5	51.1

Table 1. Analysis of ROC curves of correlation between sepsis and biomarkers

The mean gestational age of newborns with negative blood culture was 37.40 ± 3.2 weeks and with positive blood cultures 36.46 ± 3.9 weeks . The mean birth weight of newborns with negative blood culture was 2886.5 ± 768.3 grams, while with positive blood cultures was 2685.6 ± 889.7 grams.

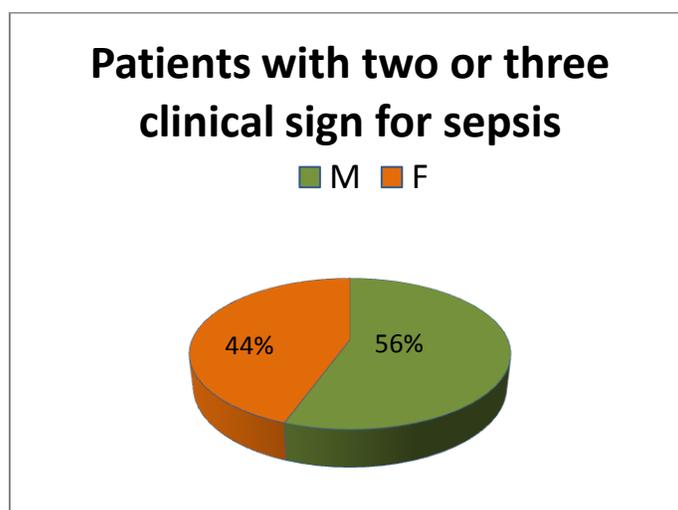


Figure 1. Distribution of newborns with two or three clinical signs of sepsis depending of gender

The newborns have been divided into two groups: I group included 31 septic newborns with negative blood culture and II group - 14 septic newborns with positive blood cultures. Results of blood count (WBC), CRP and PCT, were recorded.

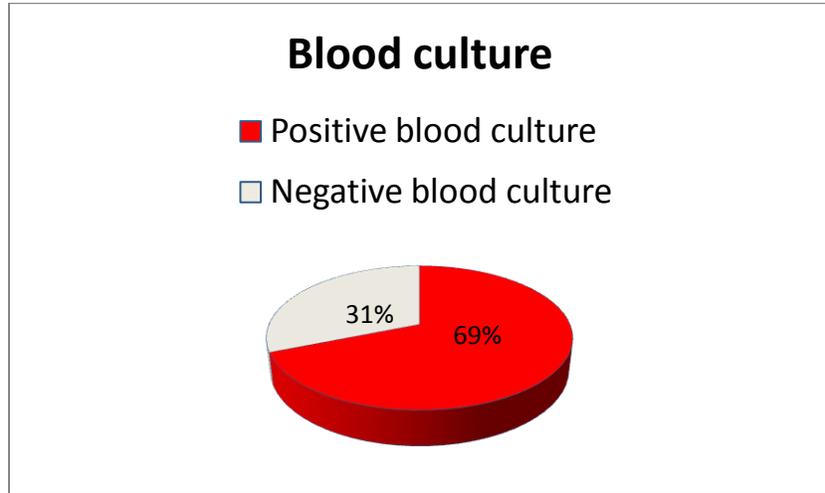


Figure 2. Distribution of newborns with two or three clinical signs of sepsis depending of blood culture

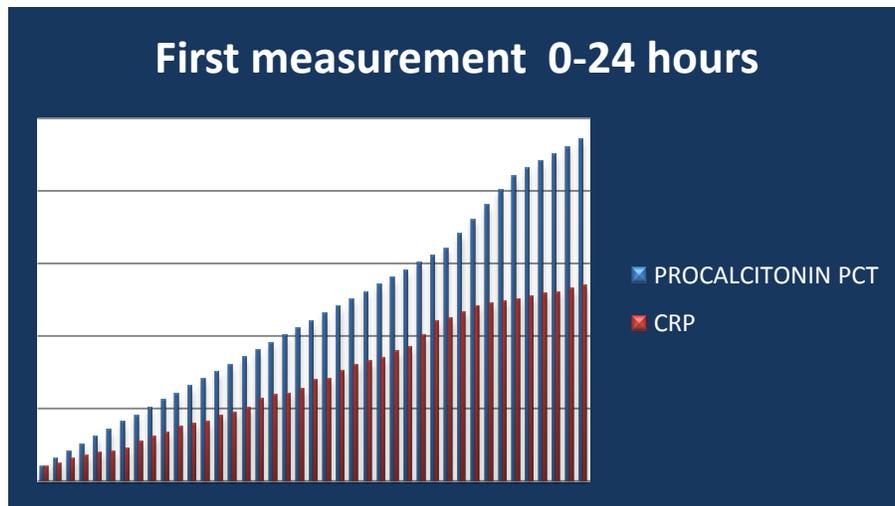


Table 2. First measurement 0-24 hours

Procalcitonin PCT levels at first 24 hours of the admission were increased in all 45 patients (≥ 2 ng/mL). The values of C-reactive protein gradually increase after 12-36 hours at admission. PCT level was significantly higher in all newborns in comparison with CRP levels

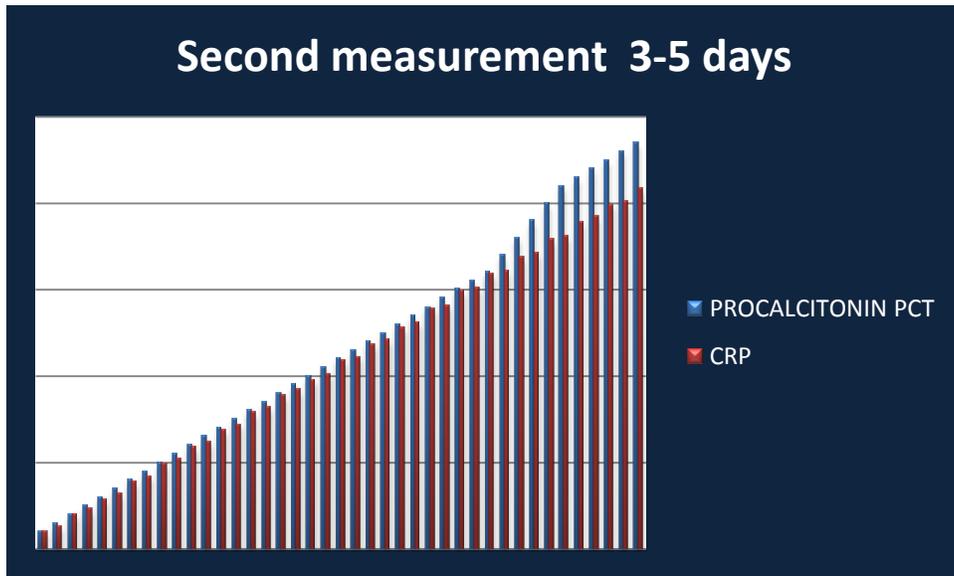


Table 3. Second measurement 3-5 days

The second measurement, after 3 days usage of an adequate antibiotic treatment, the levels of PCT is decreased, regardless of whether blood culture is positive or negative, except 6 patients develop severe sepsis, and three patients develop septic shock.

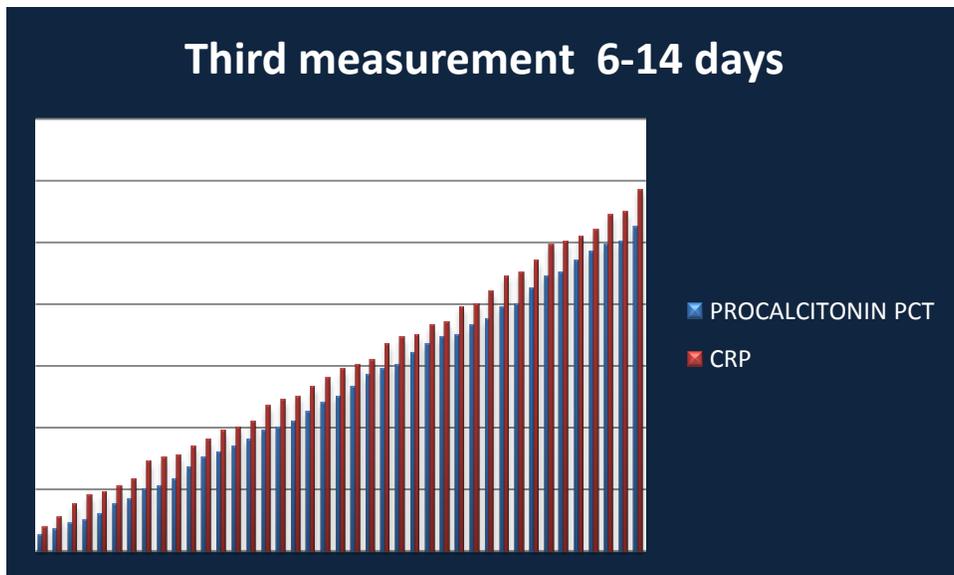


Table 4. Third measurement 6-14 days

After the third measurement (6-14 days) the levels of PCT and CRP is decreased.

Mean levels of procalcitonin in newborns were significantly higher than CRP in first measurement ($P < 0.005$). Sensitivity, specificity, positive predictive value and negative predictive value were determined for all markers and compared with each other.

Discussion

New awareness of current trials on the level of procalcitonin (PCT) and its sensitivity and specificity for early diagnosis of sepsis in pediatric patients have been reported in large numbers in studies [14]. The major causes of morbidity and mortality in pediatric intensive care units are infection and sepsis. Rapid detection of the infections in PICU is vitally important. C-reactive protein and PCT are the two biochemical markers, which are already being used to evaluate the infections. Procalcitonin rises more rapidly than C-reactive protein in case of infection and decline more quickly in terms of recovery. Procalcitonin is detected in the body only 3 to 4 hours after infection, the peak reaches 6 hours and the plateau reaches 24 hours. Contrary to this, the level of C-reactive protein (CRP) is increased between 18 and 36 hours by bacterial infection [15]. In the blood, procalcitonin has a half-life of between 25 and 30 hours. Blood procalcitonin concentrations increase in systemic inflammation, especially when bacterial infection is caused. [16] The risk of a local bacterial infection is observed when the harmfulness of procalcitonin exceeds 0.25 ng / ml. The risk of a systemic bacterial infection is monitored when the harmfulness of procalcitonin exceeds 0.5 ng / ml, high severity of procalcitonin has severe sepsis and septic shock [16,17]. High PCT levels have been considered as a well diagnostic marker for septic shock

Studies show that early diagnosis and response to therapy improve the outcome of pediatric patients with infections leading to sepsis. This opens the prospect of the use of procalcitonin (PCT) as a marker for the choice and duration of antibiotic therapy for the treatment of sepsis in pediatric patients [18,19]. There are numerous studies on the Cochran base that polyclonal intravenous immunoglobulin reduces case mortality, but monoclonal antibodies may not be effective if the evidence is insufficient [20]. In addition, studies showing that low doses of corticosteroids over 5 days are effective in increasing the possibility of surviving septic shock without causing damage.[21]. The addition of aminoglycosides to beta-lactam cones in sepsis manure does not demonstrate clinical efficacy relative to self-administration of beta lactams. Combined treatment carries the risk of nephrotoxicity [22,23]. Other studies have shown that febrile patients with granulocytopenia (neutropenia), with therapy including beta-lactam with a broad spectrum, such as third-generation cephalosporin 3 (ceftazidime or ceftriaxone) + single ammonoglycoglycosidic [24]. Patients with neutropenia may develop sepsis if they remain unresponsive to antibiotic therapy for several hours after fever [25,26]. Antibiotic change on the basis of change in infection flow and response to therapy prevents the onset of severe sepsis and septic shock [27].

From the studies above, we can see the importance of procalcitonin (PCT) as an early biochemical marker for the diagnosis and prognosis of sepsis in pediatric patients, which shows greater specificity and sensitivity than other proinflammatory markers.

The dynamics of procalcitonin (PCT) concentration determine the duration of antibiotic therapy or its modification.

Conclusion

Pediatric sepsis is a life-threatening condition induced by micro-limitations, which consists of a range of disorders, when the defensive immune system, instead of controlling the infection, leads to damage to its own tissues and organs. Early recognition and treatment clearly improves the outcome of pediatric patients with infection leading to sepsis. We examined two parameters at febrile newborns with two or three clinical signs of sepsis, the values of PCT increased at the moment at admission, while the values of C-reactive protein gradually increase. The PCT measurement may provide a more rapid means of ruling in or out sepsis provided before availability of blood culture results. The value of PCT is a

reliable parameter whether an appropriate antibiotic for the treatment is used,thus increasing patient safety, reducing costs and reducing the development of antibiotic resistance.

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