The predictive score for early-onset neonatal sepsis

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The aim of the present study was to analyze complete blood count (CBC) and C-reactive protein (CRP) levels to create the predictive score for diagnosis of early-onset neonatal sepsis (EONS).

All neonates treated for suspected EONS between January 2004 and December 2006 were evaluated from their case record. A diagnosis of EONS was made if either clinical findings consistent with sepsis developed within 72 hours of life, or if positive cultures were obtained.

Evaluations for EONS were preformed in 341 neonates, and 199/341 (58.4%) developed EONS. Total white blood count, immature/total ratio, immature/mature ratio, and CRP levels were found to be independent predictors of EONS, and the predictive score for EONS was created. An increase in the predictive score for EONS was directly correlated with possibility of EONS. Receiver operating characteristic (ROC) curve analysis determined a cut-off value of a predictive score for EONS >0.503, with sensitivity of 73% and specificity of 89%. Correct prediction of EONS was found in 78% of all neonates, 80% for positive and 75% for negative outcome (p<0.0001).

In conclusion, for its high sensitivity and prediction rates, the predictive score for EONS is useful in diagnostic evaluation of neonates suspected for EONS.

Key words: neonatal sepsis, early-onset, predictive score, diagnosis.

Neonatal sepsis may be classified according to the time of onset of the disease, as early-onset (EONS) and late-onset (LONS) neonatal sepsis. EONS occurs from birth to 72 hours of life and is associated with transplacental infection or an ascending infection from the cervix caused by microorganisms colonizing the maternal genitourinary tract or birth canal[1,2]. It may have subtle, diverse and non-specific clinical signs. Therefore, early diagnosis and treatment of the neonate with suspected sepsis are essential to prevent severe and life-threatening complications[3].

The incidence of neonatal sepsis in developed countries is 3.5-4.3/1,000 live births, with EONS representing 58% of the cases. Developing countries have both the highest incidence and the highest mortality rates[4,5]. It is a major cause of fatality during the first month of life, contributing to 13-15% of all neonatal deaths[6].

Despite the development of medicine, new antibiotics and other means of treatment, EONS remains an important problem in perinatal medicine. Accurate and timely diagnosis of EONS remains challenging to the clinician and the laboratory. A definitive diagnosis based on the culture of blood, cerebrospinal fluid (CSF) or urine is usually reached only after a delay of a day or two, yet rapid progression of untreated infection may greatly increase morbidity or mortality[7,8]. Moreover, clinical signs of EONS manifest themselves in the absence of a positive culture. This is particularly common in probable EONS with obstetric risk factors where receipt of antenatal maternal antibiotics is common[9].

Thus, over the last decade, a variety of laboratory tests have been developed to enhance the early and accurate identification and treatment of neonates with suspected sepsis[10,11]. As yet, no international consensus regarding screening of EONS has been made[12]

The aim of the present study was to analyze complete blood cell count (CBC) and C-reactive

protein (CRP) levels to create the predictive score for diagnosis of EONS.

Material and Methods

Subject Population

The study was conducted at the Department of Neonatology, Clinic for Gynecology and Obstetrics, University Clinical Center, Tuzla, over a three- year period from January 1, 2004 to December 31, 2006. Subjects were identified prospectively. Relevant data of the neonates during this period were obtained retrospectively from their case records. The clinical characteristics of the analyzed population and characteristics of delivery are shown in Table I.

In very low gestational age neonates, who are routinely admitted to a neonatal intensive care unit (NICU), the neonate can be evaluated and treated with little additional cost. In contrast, older neonates are unlikely to be admitted to the NICU, and there is economic pressure to discharge them sooner rather than later, particularly with vaginal deliveries. The clinician's dilemma is: "Which neonate needs antibiotic treatment?" Thus, neonates were included in the study if they: a) were gestational age >33 weeks; b) were from single pregnancies; and c) were ever evaluated for EONS during the birth hospitalization. A neonate was considered to have been evaluated for EONS if culture (from a normally sterile site) and laboratory studies (CBC and CRP) were obtained in the first 72 hours of life. Some of them were asymptomatic and were evaluated for sepsis because of maternal intrapartum sepsis risk factors (prolonged rupture of membranes, maternal urinary tract infection, maternal intrapartum fever >38°C, chorioamnionitis, and excessive vaginal discharge) according to the Centers for Disease Control and Prevention[13]. Neonates were excluded if they: a) were critically ill and received antibiotic treatment before blood sampling; b) had a major congenital anomaly; c) had inborn errors of metabolism, hemolytic jaundice or respiratory distress syndrome (due to surfactant deficiency); d) underwent the first evaluation after 72 hours of life; and e) were born outside the clinic.

Clinical signs consistent with EONS included: poor feeding, feeding intolerance, lethargy, irritability, temperature instability, poor respiratory effort (apnea, need for supplement oxygen, need for ventilation), tachypnea, tachycardia/bradycardia, hypotension, abnormal glucose homeostasis, metabolic acidosis, nonphysiologic jaundice, abdominal distention, and necrotizing enterocolitis[14].

Gender, gestational age, birth weight, Apgar score, and delivery characteristics were recorded for each neonate.

Subject Classification

Patient classification was based on culture results or clinical factors (results of physical examinations or laboratory studies). A proven EONS was defined as an infection confirmed by a positive culture from a normally sterile site. Cultures were obtained using a standard clinical pathway. Bacteria recovered in cultures were considered to be pathogenic unless they were normal skin or upper respiratory flora, all other laboratory studies were normal, and the neonate either had no clinical signs of infection

Table I. Clinical Characteristics of Neonates and Delivery Characteristics

Neonatal characteristics		
Gender (number)	Male/Female	163/178
Gestational age (weeks)	X±SD	38.6±1.4
Birth weight (g)	X±SD	3167.0±721.3
Apgar score 1st minute	X±SD	8.4±1.5
Apgar score 5th minute	X±SD	8.58±1.2
Delivery characteristics		
Spontaneous vaginal delivery	n (%)	86 (25.2%)
Induced vaginal labor	n (%)	146 (42.8%)
Elective cesarean section	n (%)	71 (20.8%)
Urgent cesarean section	n (%)	38 (11.1%)
Duration of labor (h)	X±SD	4.8±3.4
Duration of rupture of membranes (h)	Median (range)	24 (0-96)

or such signs resolved without antimicrobial therapy. A probable EONS was determined if: clinical or laboratory studies were consistent with this diagnosis but cultures were negative. No EONS (who served as control group) indicated there were no clinical or laboratory study results attributable to sepsis.

Statistical Analysis

Statistical analyses were preformed using SPSS version 10 for Windows. The Kolmogorov-Smirnov test was used to test for normality; statistics determined to be normal are displayed as mean ± standard deviation, while those determined to be nonparametric are shown as median and range. Student *t*-test was used to compare categorical variables. CBC and CRP levels were tested by multiple regression analysis, and according to weight factors, their regression coefficients were calculated. Receiver operating characteristic (ROC) analysis was used to determine the cut-off value of the predictive score for EONS. The area under the ROC curve was calculated using the method of Hanley and McNeil[15]. The predictive score levels were tested by logistic regression analysis to calculate likelihood of EONS.

A two-sided p value <0.05 was considered significant.

Results

During the study period, there were 12,298 live births in the Clinic for Gynecology and Obstetrics in Tuzla. Evaluations for EONS were performed at the age of 72 hours or less in 341 neonates, and 199 (58.4%) of them developed EONS, with an incidence of 16.2 per 1,000 live births. Fifty-two of 199 neonates (26.1%) had culture-proven EONS. The rates of gram-positive and gram-negative isolates were almost equal (51.9% vs 48.1%), and the rate of infection was higher among males (54.3%) than females (45.7%).

The mean of total white blood cell (WBC) count, immature/total (I/T) ratio and immature/mature (I/M) ratio, and serum CRP levels were significantly different between neonates with and without EONS (Table II).

In multivariable regression model, total WBC count, I/T ratio, I/M ratio and CRP levels were found to be independent predictors of EONS (Table III). According to weight factors of predictors from the described analysis, the predictive score for EONS was created (Formula 1).

The mean predictive score for EONS was significantly higher in neonates with EONS than in healthy neonates (0.71 ± 0.32 vs 0.36 ± 0.12, $p<0.001$) (Fig. 1). An increase in predictive score for EONS was directly correlated with possibility of EONS, while values larger than one in almost 100% suggested the presence of EONS (Fig. 2).

Receiver operating characteristic (ROC) analysis was made according to the mean predictive score values in the analyzed neonates, and the best results were determined for a cut-off value of more than 0.503, with area under the curve of 0.867 ± 0.019 ($p<0.0001$), and sensitivity of 73% and specificity of 89% (Fig. 3). Logistic regression analysis of predictive score for EONS was significant with $p<0.0001$. Correct prediction of EONS was found in 78% of all neonates, 80% for positive outcome and

Table II. Mean of Total White Blood Cell Count, I/T and I/M Ratio, and Serum CRP Levels of Neonates With and Without EONS

Parameter		Early-onset neonatal sepsis (EONS)		p-value
		YES	NO	
Total WBC count ($x10^9$/L)	X±SD	22.5±9.5	20.5±5.1	0.0233
I/T ratio	median (range)	0.09 (0-0.48)	0.03 (0-0.20)	< 0.0001
I/M ratio	median (range)	0.10 (0-0.92)	0.03 (0-0.20)	< 0.0001
CRP (mg/L)	median (range)	20.6 (4.4-197.8)	3.1 (0.1-12.4)	< 0.0001

WBC: White blood cell. I/T ratio: Immature/total neutrophil ratio. I/M ratio: Immature/mature neutrophil ratio. CRP: C-reactive protein.

Table III. Multivariable Regression Model of Predictors for Early-Onset Neonatal Sepsis

Predictor	Univariable model	Multivariable model
	% of cases correctly classified (*P* value)	Regression coefficient (*P* value)
Total WBC count	53 (0.0246)	0.01 (<0.0001)
I/T ratio	67 (<0.0001)	5.7 (<0.0001)
I/M ratio	67 (<0.0001)	- 2.9 (0.0036)
CRP level	90 (<0.0001)	0.01 (<0.0001)
Total model	94 (<0.0001)	

WBC: White blood cell. I/T ratio: Immature/total neutrophil ratio. I/M ratio: Immature/mature neutrophil ratio. CRP: C-reactive protein.

Formula 1.

Predictive score = (WBC x 0.01) + (I:T ratio x 5.7) – (I:M ratio x 2.9) + (CRP x 0.01) **for EONS**

EONS: Early-onset neonatal sepsis. WBC: White blood cell. I/T ratio: Immature/total neutrophil ratio. I/M ratio: Immature/mature neutrophil ratio. CRP: C-reactive protein.

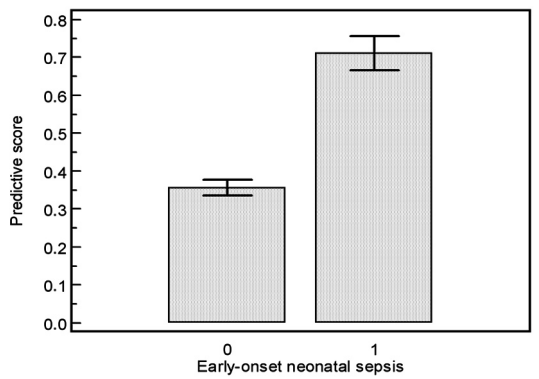

Fig. 1. Comparison of predictive score in neonates with (1) and without (0) early-onset neonatal sepsis.

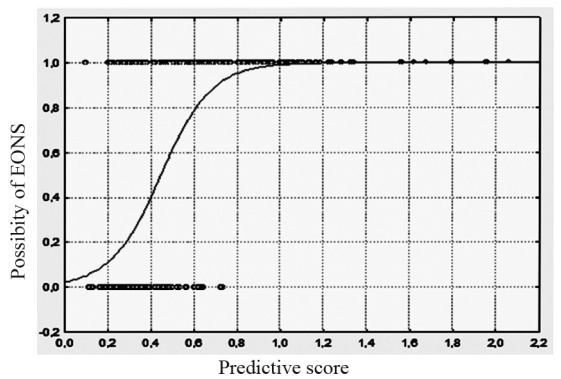

Fig. 2. Logistic regression analysis of predictive score for early-onset neonatal sepsis.

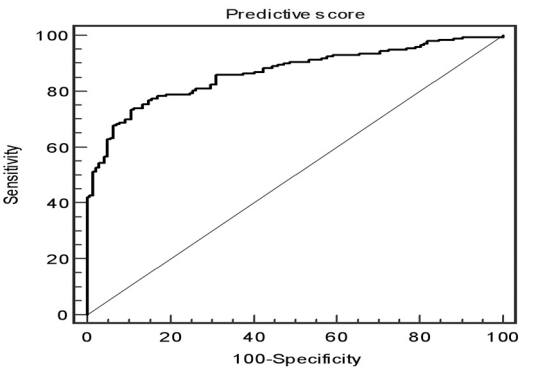

Figure 3.ROC curve analysis of predictive score for early-onset neonatal sepsis.

75% for negative outcome [odds ratio (OR): 4562.18; 95% confidence interval (CI): 675.54-30810.36].

Discussion

Early-onset neonatal sepsis (EONS) is one of the most common diagnostic challenges in neonatal medicine today. Although culture results are the "gold" standard for diagnosis, among the studies[8,16], positive cultures ranged from 8-73%. The isolation of microorganism depends on skin disinfection, sample volume and sampling site. Similar to Arshad et al. [17], we found that 26.1% of neonates had positive cultures.

Ideally, a screening test that is considered reliable and diagnostic must have a high level of sensitivity and specificity in identifying what it is measuring. In other words, a test used to diagnose EONS must always indicate abnormal results in those neonates who have EONS (sensitivity) and indicate normal results in neonates who are not infected (specificity)[18]. Thus, the idea of predictive models based on initial laboratory data is worth trying and seems to be of practical value in evaluation of neonates for EONS. Using multivariable

regression model, we found WBC, I/T ratio, I/M ratio, and CRP levels to be independent predictors of EONS, but this was not found for other hematological parameters. According to weight factors of predictors from the described analysis, the predictive score for EONS was created. Song et al. [19] emphasized superior performance of multivariable regression models in correctly predicting health outcomes. In the present study, an increase of predictive score for EONS was directly correlated with possibility of EONS, while values larger than one in almost 100% suggested the presence of EONS.

Receiver operating characteristic curve analysis determined a cut-off value of predictive score for EONS of more than 0.503, with sensitivity of 73% and specificity of 89%. The ROC curve provides a comprehensive picture of the ability of the test to make the distinction being examined over all decision thresholds. Qualitatively, the closest the curve is to the upper left corner, the higher the overall accuracy of the test[20]. In the current study, the predictive score for EONS with high area under the curve (0.867) showed good accuracy in the diagnosis of EONS.

On the other side, traditional descriptors (sensitivity, specificity, positive and negative predictive values) may not accurately represent test performance, because they are heavily influenced by the prevalence of disease in the sample population. Logistic regression analysis allows us to estimate the relationship between one dichotomous dependent variable and one or more independent variables[21]. Logistic regression analysis of predictive score for EONS was significant with $p<0.0001$. We found correct prediction of EONS in 78% of all neonates, 80% for positive and 75% for negative outcome. Thus, using the constructed predictive score for EONS for levels >0.503 with accuracy of 80% and high OR, we can predict that a neonate had EONS. Hupertan et al.[22] reported the importance of designing prognostic models using their own database, because there is a low accuracy of models from other regions.

Although EONS may have subtle, diverse and non-specific early clinical signs and can easily be confused with other noninfective causes, management of symptomatic neonates is not controversial, as all need evaluation for infection and early treatment. There is the problem, however, with asymptomatic neonates and maternal intrapartum risk factors. Do they really need diagnostic evaluation for infection? In our study, 115/199 (57.8%) newborns with EONS (proven or probable) were asymptomatic at delivery, and 27.8% of them developed symptoms within 24 hours of life. Almost all of them (85.2%) had abnormal predictive score for EONS. It is true that almost all of these neonates would have developed symptoms suggestive of infection at some time after delivery, and be treated with antibiotics, but the consequences of the short delay in evaluation and treatment are not fully known.

In conclusion, for infection, a neonate is more likely to suffer if infection is underdiagnosed and not treated than if the infection is overdiagnosed and the neonate is treated unnecessarily. While awaiting culture results, for its high sensitivity and prediction rates, the predictive score for EONS is useful in evaluation of neonates suspected for EONS. It is a cheap and cost-effective method in developing countries. Of course, validation of the designed predictive model, using larger databases, is necessary to improve its accuracy.

REFERENCES

1. Aggarval R, Sarkar N, Deorari AK, Paul VK. Sepsis in the newborn. Indian J Pediatr 2001; 68: 1143-1147.
2. Ojukwu JU, Abonyi LE, Ugwu J, Orji IK. Neonatal septicemia in high risk babies in South-Eastern Nigeria. J Perinat Med 2005; 34: 166-172.
3. Adams-Chapman I, Stoll BJ. Neonatal infection and long-term neurodevelopment outcome in the preterm infant. Curr Opin Infect Dis 2006; 19: 290-297.
4. Scuchat A, Zywicki SS, Dinsmoor MJ, et al. Risk factors and opportunities for prevention of early-onset neonatal sepsis: a multicenter case control study. Pediatrics 2000; 105: 21-26.
5. Chacko B, Sohi I. Early onset neonatal sepsis. Indian J Pediatr 2005; 72: 23-26.
6. The WHO Multicenter Study Group. Clinical prediction of serious bacterial infections in young infants in developing countries. Pediatr Infect Dis J 1999; 18: S23.
7. Stoll BJ. Infection of the neonatal infant. In: Behrman RE, Kliegman RM, Janson HB (eds). Nelson Textbook of Pediatrics (17th ed). Philadelphia: WB Saunders; 2004: 623-640.
8. Buttery JP. Blood cultures in newborns and children: optimising an everyday test. Arch Dis Child Fetal Neonatal Ed 2002; 87: 25-28.

9. Vieira RC, Procianov RS, Mule LD, Prado CH. The influence of intrapartum antibiotic therapy on the diagnosis of early-onset neonatal sepsis. J Pediatr 1997; 73: 171-175.

10. Ng PC. Diagnostic markers of infection in neonates. Arch Dis Child Fetal Neonatal Ed 2004; 89: 229-235.

11. Weinberg G, Powell K. Laboratory aids for diagnosis of neonatal sepsis. In: Remington J, Klein J (eds). Infectious Diseases of the Fetus and Newborn Infant (5th ed). Philadelphia, PA: Saunders; 2001: 1327-1344.

12. Zeeshan A, Ghafoor T, Waqar S, et al. Diagnostic value of C-reactive protein and haematological parameters in neonatal sepsis. J Coll Physicians Surg Pak 2005; 15: 152-156.

13. Centers for Disease Control and Prevention. Prevention of perinatal group B streptococcal disease: revised guidelines from CDC. MMWR 2002; 51: 1-22.

14. Gitto E, Karbownik M, Reiter JR, et al. Effects of melatonin treatment in septic newborns. Pediatr Res 2001; 50: 756-760.

15. Hanley JA, McNeil BJ. The meaning and use of the area under a receiver operating characteristic (ROC) curve. Radiology 1982; 143: 29-36.

16. Chiesa C, Pellegrini G, Panero A. C-reactive protein, interleukin-6 and procalcitonin in the immediate postnatal period: influence of illness severity, risk status, antenatal and perinatal complications and infection. Clin Chem 2003; 49: 60-68.

17. Arshad A, Asghar I, Tariq MA. Role of serum C-reactive protein in the rapid diagnosis of neonatal sepsis. Pak Armed Forces Med J 2003; 53: 178-182.

18. Mehr S, Doyle LW. Cytokines as markers of bacterial sepsis in newborn infants: a review. Pediatr Infect Dis J 2000; 19: 879-887.

19. Song X, Mitnitski A, Cox J, Rockwood K. Comparison of machine learning techniques with classical statistical models in predicting health outcomes. Medinfo 2004; 11: 736-740.

20. Zweig MH, Campbell G. Receiver-operating characteristic (ROC) plots: a fundamental evaluation tool in clinical medicine. Clin Chem 1993; 39: 561-577.

21. Pampel FC. Logistic regression: a primer. Sage University Papers Series on Quantitative Applications in the Social Sciences. Thousand Oaks, CA: Sage; 2000: 7-132.

22. Hupertan V, Roupret M, Poisson JF, et al. Low predictive accuracy of the Kattan postoperative nomogram for renal cell carcinoma recurrence in a population of French patients. Cancer 2006; 107: 2604-2608.