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The Open Microbiology Journal, 2016, *10*, 133-139 133

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The Open Microbiology Journal

Content list available at: www.benthamopen.com/TOMICROJ/

DOI: 10.2174/1874285801610010133

RESEARCH ARTICLE

Comparison Between Pathogen Associated Laboratory and Clinical Parameters in Early-Onset Sepsis of the Newborn

Bernhard Resch[1,2,*], Renoldner B[1] and Hofer N[1]

[1]*Research Unit for Neonatal Infectious Diseases and Epidemiology, Medical University of Graz, Graz, Austria*
[2]*Division of Neonatology, Department of Pediatrics, Medical University of Graz, Graz, Austria*

Received: December 12, 2015 Revised: June 10, 2016 Accepted: June 14, 2016

Abstract:

Objectives:

To identify laboratory and clinical characteristics of different pathogens associated with early-onset sepsis (EOS) of the newborn.

Methods:

Newborns with EOS were retrospectively analyzed regarding laboratory and clinical parameters associated with the identified pathogen.

Results:

We identified 125 newborns having diagnosis of culture proven EOS between 1993 and 2011. One hundred cases had diagnosis of group B streptococci (GBS) infection (80%), 11 had *Escherichia coli* (8.8%), eight enterococci (6.4%), and six other pathogens (4.8%). White blood cell count (WBC), immature to total neutrophil (IT) ratio, and C-reactive protein (CRP) values did not differ between groups within the first 72 hours of life. Presence of high (>30000/μL) and low (<9000/μl) WBC was significantly less found compared with IT-ratio >0.2 in GBS and *E.coli* EOS. High WBC were more common found than low WBC in all groups. Gram positive pathogens were more common found in late preterm and term infants (84%), and gram negative pathogens more common in very low birth weight infants (64%). *E. coli* was significantly associated with lower gestational age and birth weight, respectively.

Conclusion:

An abnormal IT-ratio was a more common finding than an abnormal WBC in GBS and *E. coli* EOS. *E. coli* was significantly associated with prematurity.

Keywords: Early-onset neonatal sepsis, *Escherichia coli*, Group B Streptococcus, IT-ratio, White blood cell count.

INTRODUCTION

Early onset sepsis (EOS) of the newborn is a severe disease associated with high morbidity and mortality [1]. At present *group B streptococci* (GBS) and *Escherichia coli* (*E.coli*) are the most common pathogens in developing countries [2, 3]. The natural incidence of GBS EOS, before preventive intervention, ranged from 0.5 to 4 or even more cases per 1000 live births with substantial geographical variations [4]. Neonatal GBS diseases remain a global public health concern despite a 70% reduction following successful implementation of intrapartum prophylaxis [4 - 8]. On the other hand there are raised concerns suggesting that the widespread use of antibiotics might increase the frequencies of non-GBS or antimicrobial-resistant pathogens [9]. In the USA the case-fatality ratio for EOS ranged from below 5% among cases of bacteremia to over 15% among cases of Gram negative meningitis [9, 10].

* Address correspondence to this author at the Division of Neonatology, Department of Pediatrics Medical University of Graz Auenbruggerplatz 34/2, 8036 Graz, Austria; Tel: ++43 316 385 81134; Fax: ++43 316 385 12678; E-mail: Bernhard.resch@medunigraz.at

Recently, Murphy and Weiner defined normal values for evaluation of EOS as white blood cell counts (WBC) ranging from 6000 to 30000/μL and an immature to total neutrophil ratio (IT-ratio) below 0.2 [11]. Two negative IT-ratios and a negative blood culture at 24 hours demonstrated to be indicative for a non-infected neonate. Twenty-three of 3154 neonates of this study were diagnosed as having culture proven EOS and all of them had at least one abnormal WBC counts except one *E.coli* positive neonate. These findings are in contrast to results of our group reporting abnormal WBC and IT-ratio as part of the systemic inflammatory response syndrome (SIRS) in only 43% of all neonates with culture-proven EOS and in 39% when defining normal WBC counts as between 9000 and 34000/μL [12, 13]. This raises the question whether there are pathogen-associated laboratory findings that significantly influence interpretation of routine laboratory marker.

Thus, we aimed to identify laboratory and clinical characteristics of different pathogens associated with EOS of the newborn.

MATERIAL AND METHODS

A cohort of newborns with blood-culture proven EOS collected at a level 3 neonatal intensive care unit of a university hospital over a 18-year time period was retrospectively analyzed regarding laboratory and clinical parameters associated with the identified pathogen. Only inborn neonates admitted within 24 hours of age having diagnosis of clinical and blood culture proven EOS were included for analysis Exclusion criteria were missing or incomplete documentation, a culture-negative clinical sepsis and an unknown state of infection.

Definition of EOS

In addition to a positive blood or cerebrospinal fluid culture plausible for causing EOS newborns had to meet the following criteria: clinical signs of sepsis in ≥1 with ≥1 maternal risk factor or ≥2 clinical signs of the following groups of symptoms: a) respiratory symptoms [apnea, tachypnea (>60/min), retractions, cyanosis, respiratory distress], b) cardiocirculatory symptoms [tachycardia (>180/min) or bradycardia (<100/min), arterial hypotension], c) neurological symptoms (irritability, lethargy, seizures), d) poor skin color or prolonged capillary refilling time (>2 s), e) fever or hypothermia (core temperature >38° C or <36° C) [14, 15].

Maternal risk factors included prolonged rupture of membranes (>18 h in term newborns), clinical chorioamnionitis (uterine tenderness or foul-smelling amniotic fluid, maternal leukocytosis >12,000/μL, and maternal or fetal tachycardia) and maternal fever >38° C during labor [15, 16].

Laboratory and Clinical Parameter

For analysis of laboratory marker routine parameter available included WBC count, absolute neutrophil count, C-reactive protein (CRP), and IT-ratio. Values were calculated for day 1, 2, 3 (<24, 24-48, and 48-72 hours) and for maximum and lowest values within the time period of 72 hours of age.

Neonatal variables collected for each study patient included sex, gestational age (GA), birth weight (BW), Apgar scores at 1, 5 and 10 minutes, clinical signs [tachycardia, bradycardia, tachypnea, apnea, hypotension, hypothermia, fever], therapeutic approaches [mechanical ventilation (CPAP included), duration of mechanical ventilation, high frequency oscillation, surfactant, nitrogen oxide, immunoglobulin, catecholamine], neonatal morbidities [respiratory distress syndrome, pneumonia, pneumothorax, persistent pulmonary hypertension of the newborn, seizures, periventricular leukomalacia, intra-/periventricular hemorrhage, hypoxic ischemic encephalopathy, septic shock, multi-organ failure, disseminated intravascular coagulopathy, renal failure, mortality], and length of hospitalization.

Statistical Analysis

Statistical analyses were performed with SPSS version 20 (SPSS, Chicago, IL, USA). Descriptive statistics were obtained for all categorical variables. Statistical significance was determined for unadjusted comparisons by Mann-Whitney-U-test for continuous variables and by Fisher´s exact test for categorical variables. The significance level was set at $p<0.05$.

RESULTS

During the study period 125 of 10,555 hospitalized newborns (1.18%) of a catchment area of approximately 153,000 births were identified with culture-proven EOS (incidence 0.8 per 1,000 live births); of whom 100 had GBS infection (80%), 11 *E. coli* (8.8%), eight enterococci (6.4%), and six other pathogens (4.8%). Gram positive pathogens

were predominant (111/125, 88.8%). Perinatal data of the study population are shown in Table **1**. Pathogens identified are given in Table **2**.

Table 1. Perinatal data of 125 neonates with culture-proven early-onset sepsis.

Gestational age (GA in weeks)	37 (24-42)
Birth weight (BW in grams)	2930 (650-4670)
Term infants (≥ 37 weeks)	67 (53.6)
GA < 28 weeks	10 (8.0)
GA 28 – 32 weeks	17 (13.6)
GA 33-36 weeks	31 (24.8)
BW < 1500 grams	21 (16.8)
BW 1500 – 2500 grams	27 (21.6)
BW > 2500 grams	77 (61.6)
Small for GA (< 10^{th} percentile)	14 (11.2)
Maternal age	28 (16-46)
Twins	5 (4)
Apgar score at 1 minute	8 (1-10)
Apgar score at 5 minutes	9 (1-10)
Apgar score at 10 minutes	9 (4-10)

Data are given as median (range) or n (%)

Table 2. Pathogens identified in 125 neonates with early-onset sepsis.

Group B streptococci	100 (80)
Escherichia coli	11 (8.8)
Enterococci	8 (6.4)
Coagulase-negative staphylococci	3 (1.6)
Haemophilus influenza	1 (0.8)
Proteus mirabilis	1 (0.8)
Citrobacter freundii	1 (0.8)

Data are given as n (%)

Laboratory Findings

WBC, IT-ratio, and CRP values did not differ between groups (GBS *vs.E. coli vs.* other pathogens - enterococci and other pathogens grouped together) within the first 72 hours of life. Median CRP values were 30.2, 28.5, and 27.2 mg/L, respectively. WBC values were 17.7, 11.8, and 18.6 x 10^6/L, and IT-ratio 0.24, 0.21, and 0.24, respectively. Differences between laboratory findings in correlation to pathogens are summarized in Table **3**. Presence of high or low WBC was significantly less found compared to IT-ratio >0.2 in GBS ($p<0.001$ and $p=0.001$, respectively) and *E.coli* cases ($p=0.013$ and 0.014, respectively). In the group of other pathogens no differences were found. High WBC values were found more commonly in all groups compared to low WBC ($p=0.015$, 0.013, and 0.001, respectively).

Table 3. Differences between laboratory findings in 125 neonates with early-onset sepsis in correlation to pathogens.

	GBS n=100	***E. coli* n=11**	**Others n=14**	**p-value**
WBC >30.0 (x 10^6/L)	25	0	21	n.s. (0.055*)
WBC <9.0 (x 10^6/L)	13	36	29	0.014*
IT-ratio >0.2	41	36	43	n.s.
Thrombocytopenia (<150 x 10^6/L)	6	0	14	n.s.

Data are given as percentage; * differences between *E. coli* and GBS

WBC: white blood cell count, IT-ratio: immature-to-total neutrophil ratio, GBS: group B streptococci; n.s. not significant

Clinical Findings

The rate of term infants was 58%, 27%, and 43% in GBS, *E. coli*, and other pathogens EOS, respectively. VLBW infants were found in GBS, *E.coli*, and other pathogens EOS in 16%, 55%, and 46%, respectively. Gram positive

pathogens were common found in late preterm and term infants (84%), and gram negative pathogens in VLBW infants (64%). Gestational age and birth weight were significantly lower in *E. coli* compared to GBS EOS (median 32 *vs.* 38 weeks, p=0.005, and 1836 *vs.* 3095 grams, p=0.031, and between other pathogens and GBS (35 *vs.* 38 weeks, p=0.035, and 2240 *vs.* 3095 grams, p=0.022). Male gender did not differ between groups (GBS, *E.coli*, and other pathogens 57, 55, and 71 percent, respectively).

GBS and *E. coli* EOS differed regarding presence of hypothermia (0 *vs.* 18%, p=.009), duration of mechanical ventilation (median 4 *vs.* 8 days, p=.019), duration of therapy with supplemental oxygen (median 2 *vs.* 9 days, p=.031), length of hospitalization (median 15 *vs.* 22 days, p=.039), presence of chorioamnionitis (17 *vs.* 46%, p=.041) and maternal fever (2 *vs.* 18%, p=.049). Compared with other pathogens differences were not significant. Intrapartum antibiotic treatment was documented in 28 mothers (22%) – 24 GBS and 4 *E. coli* infections. Rate was highest in case of preterm birth of 28 weeks gestation and less (60%).

Tachycardia and bradycardia were found in 13% and 5% of cases, tachypnea and apneas in 71% and 24%, arterial hypotension in 44%, and fever in 11%, respectively. Differences between pathogens were not significant. Median temperature in case of fever was 38.7 °C. The median duration of antibiotic treatment was 10 days.

Complications of EOS

Septic shock was diagnosed in 4.0%, multi-organ failure in 8.0%, disseminated intravascular coagulopathy in 1.6%, and renal failure in 5.6%, differences between pathogens not significant. Neurological complications including seizures, intra-/periventricular hemorrhage, periventricular leukomalacia, and hypoxic-ischemic encephalopathy were diagnosed in 5.6%, 18%, 2.4%, and 4.0%. Hemorrhages were significantly more common found in *E. coli* and other pathogens compared with GBS EOS (36 and 36 *vs.* 13%, p=0.015 and 0.045, respectively). Periventricular leukomalacia and seizures were significantly more often diagnosed in other pathogens compared with GBS EOS (1.0 *vs.* 14%, p=0.039, and 4.0 *vs.* 21%, p=0.039, respectively). Overall mortality rate was 7.2% with the highest rate found in VLBW infants (29%). Differences were not significant between pathogens.

Changes Over Time

The rate of EOS was 0.26 percent during the study period (125 out of 48,600 live births). Over the years 1993 to 2011 there was a significant decrease of GBS and gram positive pathogens (p=0.014 and 0.006, Pearson´s correlation coefficient -0.601 and -0.553, respectively).

DISCUSSION

Comparing different pathogens of EOS regarding WBC, IT-ratio, and CRP values did not reveal significant differences. Pathogen-associated characteristics included significantly less low WBC counts in *E. coli* compared to GBS EOS. An elevated IT-ratio was a more common finding compared to high or low WBC counts in *E. coli* and GBS EOS. For all pathogens high WBC counts were more commonly found than low counts *E. coli* EOS was significantly associated with prematurity, and clinical findings were not found to be pathogen-specific. Overall complication and mortality rates were low, and the latter was not different between pathogens.

An analysis of 2164 neonates with EOS and positive culture revealed 60% having a WBC values between 5000 and 19,000/μL, 82% having normal platelet count (between 150,000 and 400,000/μL), and 31% having an IT-ratio below 0.2 [17]. The first (WBC <5500/μL) through fourth vintiles (WBC <8800/μL) were associated with increased odds of EOS in this study. This lower limit is in accordance to our definition of the lower WBC cut-off value and scrutinizes critically a lower limit of 6.000/μL [11]. Additionally the authors observed a significant association with EOS for IT-ratios above 0.24 and for platelet counts below 147,000/μL [17]. Odds ratios for low WBC counts and high IT-ratios associated with infection were 5.38 and 7.97, respectively with high specificity and negative predictive values (73.7%-99.9% and >99.8%), however, sensitivities were low (0.3%-54.5%) for all complete blood cell count indices analyzed. In another study IT-ratio was not able to distinguish between culture positive and negative neonates (p=0.189) and was found to be normal in 29% of infected and abnormal in 53% of uninfected neonates [18]. Summarizing our findings and recent reports reveals IT-ratio being the most important early parameter for the diagnosis of EOS irrespective of the responsible pathogen [11, 17, 19].

We recently demonstrated that small for gestational age (SGA) neonates had lower median WBC counts, lower median neutrophil, and lower median platelet counts during the first 3 days of life compared with appropriate for

gestational age (AGA) neonates with the same finding when analyzing values separately for preterm and term neonates [20]. Interestingly, the SGA neonates were not at an increased risk for culture-proven and clinical sepsis, and in neonates with culture-proven sepsis, WBC, neutrophil, and platelet counts did not differ between SGA and AGA neonates.

The gestational age distribution showing higher rates of term infants in GBS EOS (73 to 80%) is well known and comparable to our data [2, 9]. The finding of lower birth weight and gestational age associated with *E.coli* infection with subsequently longer hospitalization and a higher rate of mechanical ventilation again is again already well documented [21]. In contrast, we found a significantly higher rate of chorioamnionitis, and could not confirm a higher mortality rate in the *E.coli* group after adjustment for gestational age. Differences regarding presence of seizures and intraventricular hemorrhages were markedly higher in the *E.coli* group, but did not reach statistical difference [21]. This is in contrast to our findings and maybe attributable to the higher rate of prematurity. One factor again influenced by the presence of prematurity was the higher rate of hypothermia in the *E.coli* group, a phenomenon of preterm infants´ reaction to infectious stimuli [22].

GBS, diagnosed in 36%, was associated with higher rates of meningitis involvement but lower incidence of mortality compared with *E. coli*, present in 26%, in a 6-years study period analyzing 109 septic episodes of 100 neonates [23]. The sepsis-related mortality rates was higher in EOS (10%) compared to late-onset sepsis (7%). The higher mortality rate (16%) especially associated with *E. coli* sepsis (33%) reported by Stoll *et al.* differs marginally to our findings and might be explained be the particular high rate of preterm infants (81%) [2].

We observed a significant decrease of GBS sepsis over the study period without an increase of EOS caused by *E.coli* neither among term nor preterm infants. In one study, comparing two time periods 1991 to 1993 and 1998 to 2000, a marked reduction in GBS sepsis (from 5.9 to 1.7 per 1000 live births of infants weighing 401 to 1500 g, $p<0.001$) and an increase in *E. coli* sepsis (from 3.2 to 6.8 per 1000 live births, $p=0.004$) was observed without changes of the overall rate of EOS [24]. In another study including 53 cases of *E. coli* EOS over a long time period (between 1979 and 2006) increases in *E. coli* EOS rates over the time, and further increases of intrapartum ampicillin exposure and the observation of ampicillin-resistant *E. coli* were reported [25].

Some limitations of our study have to be mentioned. This is a retrospective analysis performed at a single center that included neonates enrolled over a large time period. Therefore, results can only be extrapolated, with caution, to center-specific characteristics. Positive blood cultures with plausible pathogens are generally accepted as the gold standard as diagnostic criterion for EOS. However, they are known to lack sensitivity, and data are evident that obtaining cultures from neonates might be difficult as sample volumes are small and a substantial number of cultures are contaminated or negative [26, 27]. As a result, the number of culture positive newborns is limited and the true infection rate is probably underestimated.

In conclusion, we observed an abnormal IT-ratio more common than an abnormal WBC count associated both with GBS and *E. coli*. *E. coli* was significantly associated with prematurity. Overall mortality rate was low without differences regarding the causative pathogen.

CONTRIBUTIONS

B. Resch was responsible for study design and writing the manuscript.

B.R. performed data collection and presentation of results.

N.H. was responsible for analysis of data and interpretation of results.

ETHICAL APPROVAL

The study was proven by the local Ethical Committee (Nr. 24-434 ex 11/12).

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGEMENTS

Declared none.

REFERENCES

[1] Cortese F, Scicchitano P, Gesualdo M, *et al.* Early and late infections in Newborns: Where do we stand? A review. Pediatr Neonatol 2015 Dec 2; pii: S1875-9572(15)00175-8

[2] Stoll BJ, Hansen NI, Sánchez PJ, *et al.* Eunice Kennedy Shriver National Institute of Child Health and Human Development Neonatal Research Network. Early onset neonatal sepsis: the burden of group B Streptococcal and *E. coli* disease continues. Pediatrics 2011; 127(5): 817-26.
[http://dx.doi.org/10.1542/peds.2010-2217] [PMID: 21518717]

[3] Vergnano S, Sharland M, Kazembe P, Mwansambo C, Heath PT. Neonatal sepsis: an international perspective. Arch Dis Child Fetal Neonatal Ed 2005; 90(3): F220-4.
[http://dx.doi.org/10.1136/adc.2002.022863] [PMID: 15846011]

[4] Melin P. Neonatal group B streptococcal disease: from pathogenesis to preventive strategies. Clin Microbiol Infect 2011; 17(9): 1294-303.
[http://dx.doi.org/10.1111/j.1469-0691.2011.03576.x] [PMID: 21672083]

[5] Verani JR, McGee L, Schrag SJ. Division of Bacterial Diseases, National Center for Immunization and Respiratory Diseases, Centers for Disease Control and Prevention (CDC). Prevention of perinatal group B streptococcal disease-revised guidelines from CDC, 2010. MMWR Recomm Rep 2010; 59(RR-10): 1-36.
[PMID: 21088663]

[6] Gray KJ, Bennett SL, French N, Phiri AJ, Graham SM. Invasive group B streptococcal infection in infants, Malawi. Emerg Infect Dis 2007; 13(2): 223-9.
[http://dx.doi.org/10.3201/eid1302.060680] [PMID: 17479883]

[7] Luck S, Torny M, d'Agapeyeff K, *et al.* Estimated early-onset group B streptococcal neonatal disease. Lancet 2003; 361(9373): 1953-4.
[http://dx.doi.org/10.1016/S0140-6736(03)13553-2] [PMID: 12801740]

[8] Verani JR, Spina NL, Lynfield R, *et al.* Early-onset group B streptococcal disease in the United States: potential for further reduction. Obstet Gynecol 2014; 123(4): 828-37.
[http://dx.doi.org/10.1097/AOG.0000000000000163] [PMID: 24785612]

[9] Moore MR, Schrag SJ, Schuchat A. Effects of intrapartum antimicrobial prophylaxis for prevention of group-B-streptococcal disease on the incidence and ecology of early-onset neonatal sepsis. Lancet Infect Dis 2003; 3(4): 201-13.
[http://dx.doi.org/10.1016/S1473-3099(03)00577-2] [PMID: 12679263]

[10] Unhanand M, Mustafa MM, McCracken GH Jr, Nelson JD. Gram-negative enteric bacillary meningitis: a twenty-one-year experience. J Pediatr 1993; 122(1): 15-21.
[http://dx.doi.org/10.1016/S0022-3476(05)83480-8] [PMID: 8419603]

[11] Murphy K, Weiner J. Use of leukocyte counts in evaluation of early-onset neonatal sepsis. Pediatr Infect Dis J 2012; 31(1): 16-9.
[http://dx.doi.org/10.1097/INF.0b013e31822ffc17] [PMID: 21860335]

[12] Hofer N, Zacharias E, Müller W, Resch B. Performance of the definitions of the systemic inflammatory response syndrome and sepsis in neonates. J Perinat Med 2012; 40(5): 587-90.
[http://dx.doi.org/10.1515/jpm-2011-0308] [PMID: 23120762]

[13] Resch B, Edlinger S, Müller W. White blood cell counts in neonatal early-onset sepsis. Pediatr Infect Dis J 2012; 31(5): 540-1.
[http://dx.doi.org/10.1097/INF.0b013e31824e4740] [PMID: 22510996]

[14] Hofer N, Müller W, Resch B. Non-infectious conditions and gestational age influence C-reactive protein values in newborns during the first 3 days of life. Clin Chem Lab Med 2011; 49(2): 297-302.
[http://dx.doi.org/10.1515/CCLM.2011.048] [PMID: 21126207]

[15] Hofer N, Kothari R, Morris N, Müller W, Resch B. The fetal inflammatory response syndrome is a risk factor for morbidity in preterm neonates. Am J Obstet Gynecol 2013; 209(6): 542.e1-542.e11.
[http://dx.doi.org/10.1016/j.ajog.2013.08.030] [PMID: 23994220]

[16] Chan GJ, Lee AC, Baqui AH, Tan J, Black RE. Risk of early-onset neonatal infection with maternal infection or colonization: a global systematic review and meta-analysis. PLoS Med 2013; 10(8): e1001502.
[http://dx.doi.org/10.1371/journal.pmed.1001502] [PMID: 23976885]

[17] Hornik CP, Benjamin DK, Becker KC, *et al.* Use of the complete blood cell count in early-onset neonatal sepsis. Pediatr Infect Dis J 2012; 31(8): 799-802.
[http://dx.doi.org/10.1097/INF.0b013e318256905c] [PMID: 22531231]

[18] Soni S, Wadhwa N, Kumar R, Faridi MM. Use of immature-to-total-neutrophil ratio in early neonatal sepsis. Pediatr Infect Dis J 2012; 31(10): 1101-2.
[http://dx.doi.org/10.1097/INF.0b013e31826108da] [PMID: 22982953]

[19] Makkar M, Gupta C, Pathak R, Garg S, Mahajan NC. Performance evaluation of hematologic scoring system in early diagnosis of neonatal sepsis. J Clin Neonatol 2013; 2(1): 25-9.
[http://dx.doi.org/10.4103/2249-4847.109243] [PMID: 24027741]

[20] Hofer N, Edlinger S, Resch B. Comparison of risk for early-onset sepsis in small-for-gestational-age neonates and appropriate-for-gestational-age neonates based on lower levels of white blood cell, neutrophil, and platelet counts. Pediatr Neonatol 2014; 55(4): 323-5.

[http://dx.doi.org/10.1016/j.pedneo.2013.12.006] [PMID: 24751401]

[21] Mayor-Lynn K, González-Quintero VH, O'Sullivan MJ, Hartstein AI, Roger S, Tamayo M. Comparison of early-onset neonatal sepsis caused by *Escherichia coli* and group B Streptococcus. Am J Obstet Gynecol 2005; 192(5): 1437-9.
[http://dx.doi.org/10.1016/j.ajog.2004.12.031] [PMID: 15902130]

[22] Weisman LE, Stoll BJ, Cruess DF, *et al.* Early-onset group B streptococcal sepsis: a current assessment. J Pediatr 1992; 121(3): 428-33.
[http://dx.doi.org/10.1016/S0022-3476(05)81801-3] [PMID: 1517922]

[23] Wu JH, Chen CY, Tsao PN, Hsieh WS, Chou HC. Neonatal sepsis: a 6-year analysis in a neonatal care unit in Taiwan. Pediatr Neonatol 2009; 50(3): 88-95.
[http://dx.doi.org/10.1016/S1875-9572(09)60042-5] [PMID: 19579754]

[24] Stoll BJ, Hansen N, Fanaroff AA, *et al.* Changes in pathogens causing early-onset sepsis in very-low-birth-weight infants. N Engl J Med 2002; 347(4): 240-7.
[http://dx.doi.org/10.1056/NEJMoa012657] [PMID: 12140299]

[25] Bizzarro MJ, Dembry LM, Baltimore RS, Gallagher PG. Changing patterns in neonatal *Escherichia coli* sepsis and ampicillin resistance in the era of intrapartum antibiotic prophylaxis. Pediatrics 2008; 121(4): 689-96.
[http://dx.doi.org/10.1542/peds.2007-2171] [PMID: 18381532]

[26] Volante E, Moretti S, Pisani F, Bevilacqua G. Early diagnosis of bacterial infection in the neonate. J Matern Fetal Neonatal Med 2004; 16(Suppl. 2): 13-6.
[http://dx.doi.org/10.1080/jmf.16.2.13.16] [PMID: 15590427]

[27] Modi N, Doré CJ, Saraswatula A, *et al.* A case definition for national and international neonatal bloodstream infection surveillance. Arch Dis Child Fetal Neonatal Ed 2009; 94(1): F8-F12.
[http://dx.doi.org/10.1136/adc.2007.126458] [PMID: 18499771]