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Original Article

Culture-proven neonatal sepsis in preterm infants in a neonatal intensive care unit over a 7 year period: Coagulase-negative *Staphylococcus* as the predominant pathogen

Hilal Ozkan,[1] Merih Cetinkaya,[1] Nilgün Koksal,[1] Solmaz Celebi[2] and Mustafa Hacımustafaoglu[2]

Divisions of [1]Neonatology and [2]Pediatric Infectious Disease, Department of Pediatrics, Faculty of Medicine, Uludag University, Bursa, Turkey

Abstract ***Background***: The aim of this study was to determine the causative agents in early, late- and very late-onset sepsis in preterm infants. The demographic features, risk factors, clinical and laboratory findings in sepsis types were also defined.

Methods: A total of 151 preterm infants with culture-proven neonatal sepsis were enrolled in this prospective study. The infants were classified into three groups with regard to the onset of sepsis: early onset sepsis (EOS), late-onset sepsis (LOS) and very late-onset sepsis (VLOS). A sepsis screen including whole blood count, blood smear, infection markers and cultures was performed before initiating antibiotic therapy.

Results: EOS, LOS and VLOS groups consisted of 23, 86 and 42 infants, respectively. Coagulase-negative staphylococci (CONS) was the most common organism in all sepsis groups. The main factors associated with EOS included presence of premature rupture of membranes, antibiotic use in pregnancy and choriamnionitis. Previous antibiotic use was the main factor associated with LOS, while low birthweight was the main factor in infants with VLOS. Although mortality rate due to Gram-negative bacteria and fungi was higher, CONS was an important cause of mortality in infants with LOS and VLOS.

Conclusions: CONS was found to be the most common causative organism in three sepsis types in preterm neonates. Although the mortality rate due to CONS was lower in EOS, it was an important cause of mortality in LOS and VLOS. CONS seems to be the main pathogen in neonatal sepsis in developing countries, as in developed countries.

Key words coagulase-negative *Staphylococcus*, early onset sepsis, late-onset sepsis, neonatal sepsis, preterm infant.

Neonatal sepsis is a clinical syndrome characterized by signs and symptoms of infection with or without accompanying bacteremia in the first month of life that continues to be an important cause of morbidity and mortality.[1,2] Neonatal sepsis, which has an incidence that varies between 1 and 8 neonates per 1000 live births, may be classified with regard to its onset into three groups as follows: early onset sepsis (EOS), late-onset sepsis (LOS) and very late-onset sepsis (VLOS).[3]

Early onset sepsis, which occurs within the first 72 h of life, is generally associated with the acquisition of microorganisms from the mother and usually presents with respiratory distress and pneumonia.[1,4] The source of the infection is commonly the maternal genital tract and the microorganisms most commonly associated with EOS are group B *Streptococcus* (GBS), *Escherichia coli, Haemophilus influenzae,* and *Listeria monocytogenes*.[1,5] The main risk factors for EOS include prematurity, low birthweight, febrile illness in the mother within 2 weeks of delivery, foul smelling and/or meconium stained liquor, premature rupture of membranes (PROM), prolonged labor, and perinatal asphyxia.[1,6,7]

Late-onset sepsis usually presents after 72 h of birth.[1] It may either be caused by peri- or postnatally acquired organisms, while it usually occurs as a consequence of nosocomial transmission.[8] Organisms that have been implicated in LOS include coagulase-negative staphylococci (CONS), *Staphylococcus aureus, E. coli, Klebsiella* spp., *Pseudomonas* spp., *Enterobacter* spp., *Candida* spp., GBS, *Serratia* spp., *Acinetobacter* spp., and anaerobes.[5,9] The affected neonates usually present with septicemia and meningitis.[1] The main risk factors for LOS include prematurity and prolonged neonatal intensive care unit (NICU) stay. Other risk factors are central vascular access, invasive procedures, and the use of broad-spectrum antibiotics.[1,9]

Very late-onset sepsis is usually diagnosed in extremely low-birthweight (ELBW) infants who remain hospitalized for several weeks after birth. The major factors increasing the sepsis risk in these infants include the intravascular catheters required for their care, prolonged exposure to antimicrobial agents, and ongoing immature host defense mechanisms.[10]

The pattern of bacterial pathogens responsible from neonatal sepsis has changed with time and varies from place to place. There has also been a difference in the causative organisms

Correspondence: Merih Cetinkaya, MD PhD, Division of Neonatology, Department of Pediatrics, Faculty of Medicine, Uludag University, Gorükle, Bursa 16059, Turkey. Email: drmerih@yahoo.com

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of neonatal sepsis between the developed and developing countries.[4,9,11–15] Therefore, EOS, LOS and VLOS differ from each other in terms of causative microorganisms, risk factors, clinical and laboratory findings and treatment. The aim of this study was to determine the causative organisms, demographic features, clinical and laboratory findings in infants with EOS, LOS, and VLOS in a tertiary care NICU over a period of 7 years. The factors associated with the three sepsis types were also evaluated.

Methods

The Pediatric Clinic at Uludag University Hospital in Bursa, Turkey, is a tertiary care pediatric referral unit in the South Marmara region that includes a 15-bed neonatal NICU. The patients were taken care of by the same four nursery personnel in the clinic throughout the study period. A total of 151 preterm infants (<37 gestational weeks) admitted to the NICU of Uludag University, School of Medicine between January 2003 and January 2010 and diagnosed with neonatal sepsis according to clinical findings and positive blood cultures were included in this study. The neonates were classified into the following three groups according to sepsis type: EOS (group 1), LOS (group 2) and VLOS (group 3). The neonates were classified into the EOS, LOS and VLOS groups according to whether sepsis first occurred in the first 3 days of life; between 4 and 30 days after birth; and >30 days after birth, respectively. The study protocol was approved by the Human Ethics Committee of Uludag University, Faculty of Medicine. Signed informed consent was obtained from the legal guardians of all infants. Exclusion criteria included antibiotic therapy on admission or refusal of parental consent. A prospective sepsis screen was performed in all infants who were admitted to NICU with probable sepsis during the study period. All suspected neonates underwent sepsis screen including total leukocyte count, absolute neutrophil count, immature-to-total neutrophil ratio, blood smear evaluation, C-reactive protein (CRP), procalcitonin (PCT) and serum amyloid A (SAA) levels in order to corroborate the diagnosis of neonatal sepsis. Once the infant was diagnosed with neonatal sepsis according to sepsis screen, two separate sets of blood cultures from two different sites were obtained before initiating antibiotics.

Gestational age, birthweight, gender, mode of delivery, Apgar scores, prenatal demographics, history of PROM and chorioamnionitis were all recorded. Temperature instability, apnea, need for supplemented oxygen, need for ventilation, tachycardia/bradycardia, hypotension, feeding intolerance, abdominal distension, necrotizing enterocolitis were considered as the clinical signs of sepsis. Changes in hematologic parameters were processed according to the Manroe *et al*. and Rodwell *et al*. scoring systems.[16,17] Leukopenia was defined as leukocyte count ≤5000/mm^3; leukocytosis was defined as leukocyte count ≥25 000/mm^3 at birth, ≥30 000/mm^3 at 12–24 h and ≥21 000/mm^3 after the second day. Thrombocytopenia was defined as platelet count ≤150 000/mm^3.

Before initiating the antimicrobial therapy, blood samples for whole blood count, CRP, PCT, SAA and culture were obtained. We used requirement of two positive cultures from venous or catheter samples as the definition of culture-proven CONS sepsis.[18] This procedure was repeated three times at 48 h, 7 days and at 10 days. Cultures of cerebrospinal fluid (CSF), urine and tracheal aspirate were also obtained, if necessary. Meningitis was diagnosed according to the cell count, glucose and protein levels in the CSF and CSF culture.

Whole blood count (WBC), PCT, CRP, SAA levels and cultures were evaluated immediately. Whole blood count was done using an automatic counter, Cell Dyn 3700 (Abbott Diagnostics Division, Santa Clara, CA, USA). CRP and SAA were determined on immunonephelometry using a BN II device (Dade Behring Marburg, Marburg, Germany). Detection limits were 0.5 mg/dL and 6.8 mg/dL for CRP and SAA, respectively. PCT was measured on monoclonal immunoluminometric assay (Lumitest PCT; Brahm Diagnostica, Berlin, Germany) which is specific for the PCT molecule. Abnormal was defined as >0.5 ng/mL. Levels <0.5 ng/mL for PCT, 0.5 mg/dL for CRP and 6.8 mg/dL for SAA were accepted as zero for statistical analysis. Blood and CSF cultures were analyzed using the fully automatic BACTEC method with a BACTEC 9240 device (Becton Dickinson, Heidelberg, Germany). Bacterial isolates were identified and antimicrobial susceptibility test was performed using Kirbey Bauer disc diffusion method.

Infants were treated with appropriate antibiotic regimens including ampicillin in combination with gentamicin as the first-line therapy for EOS. For treatment of LOS, a combination of cefotaxime and amikacin was used as first-line therapy. Neonates who had positive cultures were treated with antibiotics according to the culture antibiogram. The antimicrobial therapy was stopped on observation of clinical and laboratory improvement.

SPSS version 15.0 (SPSS, Chicago, IL, USA) was used for statistical analysis. Descriptive statistics are presented as mean ± SD, and a percentage. The Shapiro–Wilk test was used to test the normal distribution of data. Categorical data were analyzed using chi-squared test. Mann–Whitney test and Student's *t*-test were used for comparisons between groups as appropriate. Kruskall–Wallis test and one-way analysis of variance were used for comparisons of more than two groups. $P < 0.05$ was considered to be significant.

Results

A total of 985 infants, out of 2420 neonates admitted to the NICU between January 2003 and January 2010, were evaluated for neonatal sepsis. During the study period, a total of 274 sepsis episodes were determined in 255 infants. After exclusion of term infants, 151 preterm infants with sepsis were included in the study. Groups 1, 2 and 3 consisted of 23, 86 and 42 infants, respectively. Table 1 lists the demographic features of the infants in three sepsis groups. EOS, LOS and VLOS were diagnosed in 15%, 57%, and 28% of infants, respectively. CONS was the most common organism, accounting for 77 cases (50.9%), followed by *Candida* spp. (15.8%). Table 2 lists the distribution of pathogens among the 151 cases.

The main factors associated with EOS included low birthweight, presence of PROM, antibiotic use in pregnancy, maternal infection, and choriamnionitis. Previous antibiotic use

Table 1 Subject characteristics

	Early onset sepsis $n = 23$ n (%) or mean ± SD	Late-onset sepsis $n = 86$ n (%) or mean ± SD	Very late-onset sepsis $n = 42$ n (%) or mean ± SD
Male	14 (60.9)	49 (57)	21 (50)
Gestational age (weeks)	31 ± 2.4	31 ± 3.3	30 ± 2.4
Birthweight (g)	1615 ± 599	1530 ± 684	1254 ± 328
Cesarean delivery	12 (52)	60 (70)	22 (69)
Apgar 1 min	5.4 ± 2.9	4.9 ± 2.2	4.6 ± 2.1
Apgar 5 min	7.7 ± 1.9	7.2 ± 1.6	7.1 ± 1.1
Multiparity	12 (52.2)	36 (41.9)	16 (38.1)
Twins	3 (13)	15 (17.4)	10 (23.8)
SGA	0 (0)	14 (16.3)	8 (19)
Pre-eclampsia	5 (21.7)	28 (32.6)	22 (52.4)
Maternal age (years)	27.7 ± 4.9	26.9 ± 6.1	26.3 ± 6.1

SGA, small for gestational age.

Table 2 Distribution of pathogens

	Early onset sepsis (group 1) ($n = 23$)	Late-onset sepsis (group 2) ($n = 86$)	Very late-onset sepsis (group 3) ($n = 42$)	Total $n = 151$
Gram-positive bacteria				
Coagulase-negative staphylococci	14	40	23	77
Staphylococcus aureus	1	5	2	8
Group B *Streptococcus*	2	5	2	10
Enterococcus spp.	1	3	1	5
Corynebacterium spp.	0	2	0	2
Listeria monocytogenes	1	0	0	1
Gram-negative bacteria				
Pseudomonas spp.	1	6	2	9
Klebsiella spp.	0	4	1	5
Escherichia coli	1	1	0	2
Acinetobacter spp.	1	4	0	5
Enterobacter spp.	1	2	1	4
Fungus				
Candida parapsilosis	0	8	8	16
Candida albicans	0	6	2	8

(91%) was the main factor associated with LOS, whereas prematurity and low birthweight were other factors. Prematurity and low birthweight were also found to be main factors associated with VLOS. Table 3 lists the factors associated with EOS, and Table 4 presents a comparison of the factors associated with LOS and VLOS, which were different from EOS.

Table 5 presents the clinical findings according to the three sepsis groups. The incidence of pneumonia was significantly

Table 3 Risk factors associated with early onset sepsis

Risk factors	n (%)	OR (95%CI)	P
Male gender	14 (60.9)	1.28 (0.52–3.1)	0.37
Very low-birthweight (<1500 g)	12 (52)	0.53 (0.2–1.3)	0.12
Multiparity	12 (52)	1.59 (0.65–3.8)	0.21
Premature rupture of membranes	**8 (34.8)**	**3.0 (1.1–8.2)**	**0.02**
Choriamnionitis	**4 (17.4)**	**4.2 (1.1–16.5)**	**0.04**
Pre-eclampsia	5 (21.7)	0.4 (0.15–1.2)	0.08
Antibiotic use in pregnancy	**8 (34.7)**	**2.7 (1.1–7.2)**	**0.04**

CI, confidence interval; OR, odds ratio. **Bold**, statistically signficant.

Table 4 Factors associated with late-onset and very late-onset sepsis

Factors associated with sepsis	Late-onset sepsis n (%) or mean ± SD	Very late-onset sepsis n (%) or mean ± SD	P
Very low-birthweight (<1500 g)	51 (59.3)	35 (83.3)	**0.02**
Extreme low-birthweight (<1000 g)	20 (23.3)	10 (23.8)	0.5
Previous hospitalization	8 (9.3)	3 (7.1)	0.6
Duration of hospitalization (days)	10.4 ± 8.4	35.4 ± 31.5	**0.001**
Neutropenia	15 (17.4)	8 (19)	0.5
Mechanical ventilation (days)	7.5 ± 7.7	42.6 ± 33.2	**0.0001**
Total parenteral nutrition (days)	8.8 ± 8.0	45.8 ± 30.3	**0.0001**
Nasogastric catheter (days)	9.7 ± 8.2	55.6 ± 30.3	**0.0001**
Umbilical catheter (days)	4.2 ± 3.2	5.7 ± 3.1	0.14
Surgery	3 (3.5)	1 (2.4)	0.4

Bold, statistically signficant.

Table 5 Clinical findings

Clinical findings	Early-onset sepsis *n* (%)	Late-onset sepsis *n* (%)	Very late-onset sepsis *n* (%)
Tachypnea	4 (17.4)	26 (30.2)	5 (11.9)
Low activity	10 (43.5)	45 (52.3)	16 (38.1)
Worsening in general condition	7 (30.4)	40 (46.5)	19 (40.5)
Fever*	2 (8.7)	17 (19.8)	**17 (40.5)**
Jaundice	1 (4.3)	6 (7.0)	1 (2.4)
Gastric distention	2 (8.7)	18 (20.9)	12 (28.6)
Gastric residuals	1 (4.3)	20 (23.3)	6 (14.3)
Vomiting	1 (4.3)	11 (12.8)	2 (4.8)
Feeding intolerance	1 (4.3)	8 (9.3)	2 (4.8)
Apnea	1 (4.3)	9 (10.5)	4 (9.5)

*P = 0.004 (odds ratio, 3.2; 95% confidence interval: 1.4–7.0). **Bold,** statistically signficant.

Table 6 Complications

Complications	Early-onset sepsis *n* (%)	Late-onset sepsis *n* (%)	Very late-onset sepsis *n* (%)
Pneumonia*	6 (26.1)	40 (46.5)	**31 (73.8)**
Meningitis	1 (4.5)	19 (22.1)	6 (14.3)
Peritonitis	0 (0)	8 (9.3)	4 (9.5)
Endocarditis	0 (0)	1 (2.3)	0 (0)
Pleural empyema	0 (0)	1 (2.3)	0 (0)
Soft tissue infection	0 (0)	2 (2.3)	0 (0)
Death from infection**	3 (13)	**17 (19.7)**	2 (4.7)

*P = 0.0001 (odd ratio [OR], 3.8; 95% confidence interval [CI]: 1.7–8.4); **P = 0.03 (OR,1.2; 95%CI: 1.0–2.7). **Bold,** statistically signficant.

higher in infants with VLOS compared with those with EOS and/or LOS. The infection-associated mortality was significantly higher in infants with LOS (19.7%) compared with those in group 1 (13%) and group 3 (4.7%). Table 6 lists the complications for the three sepsis types.

Although CONS was the most common organism in all sepsis types, the mortality rate due to CONS was low in infants with EOS. The mortality rate due to CONS, however, was higher in infants with both LOS and VLOS. Gram-negative pathogens (35.2%) and fungi (35.2%) were the most common pathogens associated with mortality in LOS and they were followed by CONS (17.6%). CONS and *Candida* were again the predominant pathogens that led to mortality in VLOS.

Mean leukocyte count was significantly higher in group 1 compared with groups 2 and 3 ($P < 0.05$). In contrast, leukopenia was found to be more common in infants with VLOS. Mean CRP, PCT, and SAA levels were significantly lower in infants with EOS compared with other groups ($P < 0.05$). Table 7 lists laboratory findings.

The infants were subgrouped according to gestational age, and Table 8 presents the incidence of the three sepsis types in terms of gestational age. As shown in Table 8, no significant differences were detected between the three groups. Table 9 lists the antibiotic susceptibility pattern of bacterial isolates.

Discussion

In the present study, LOS caused by CONS was the most common sepsis type in the present tertiary NICU in a 7 year period. Gram-positive infection was found to be more common than Gram-negative infection and fungal sepsis in all sepsis types. Although CONS was the predominant pathogen in all sepsis types, the mortality rate due to CONS was found to be low in EOS. Although Gram-negative bacteria and fungi were more likely to be associated with mortality, CONS was determined to

Table 7 Laboratory findings

Laboratory findings	Early-onset sepsis *n* (%) or mean ± SD	Late-onset sepsis *n* (%) or mean ± SD	Very late-onset sepsis *n* (%) or mean ± SD
WBC (mm^3)*	15091 ± 7549	14097 ± 8067	10106 ± 4933
Leukocytosis (>20000/mm^3)*	5 (21.7)	15(17.4)	2 (4.7)
Leukopenia (<5000/mm^3)*	2 (8.6)	10 (8.6)	6 (14.2)
Platelets (<150 000/mm^3)	6 (26)	30 (34.8)	14 (33.3)
CRP(mg/dL)*	1.7 ± 3.1	3.9 ± 4.7	4.4 ± 5.3
CRP positivity (>0.5 mg/dL)*	10 (43.4)	60 (70)	34 (81)
PCT(mg/dL)*	3.1 ± 5.7	5.4 ± 15.9	5.3 ± 7.6
PCT positivity (>0.5 mg/dL)	17 (75)	65 (75)	31 (71)
SAA(mg/dL)*	25 ± 37.8	47 ± 53.7	50 ± 76.2
SAA positivity (>6.8 mg/dL)*	10 (45)	61 (71)	34 (81)

*$P < 0.05$. CRP, C-reactive protein; PCT, procalcitonin; SAA, serum amyloid A; WBC, white blood cells.

Table 8 Gestational age and incidence of sepsis

Gestational week	Early-onset sepsis (group 1) (n = 23), n (%)	Late-onset sepsis (group 2) (n = 86), n (%)	Very late-onset sepsis (group 3) (n = 42), n (%)	Total (n = 151)
<28	2/23 (8.6)	12/86 (13.9)	5/42 (11.9)	19/151 (12.5)
28–32	18/23 (78.2)	47/86 (54.6)	28/42 (66.6)	93/151 (61.5)
32–37	3/23 (13)	27/86 (31.3)	9/42 (21.4)	39/151 (25.8)

Table 9 Antibiotic susceptibility pattern of blood culture isolates (% susceptible)

	CONS $n = 77$	*S. aureus* $n = 8$	*Streptococcus* $n = 10$	*Enterococcus* $n = 5$	*Pseudomonas* $n = 9$	*Klebsiella* $n = 5$	*E. coli* $n = 2$	*Acinetobacter* $n = 5$
Ampicillin	28.5	25	32.3	40	NT	20	0	20
Gentamicin	42	62.5	65	60	55.5	80	100	60
Amikacin	76	75	72	NT	77.7	80	100	80
TMP/SMX	26	25	22	NT	NT	NT	NT	NT
Meropenem	40	37.5	32.6	40	100	100	100	100
Ciprofloxacin	48	50	44.5	40	66.6	60	50	80
Piperacillin	NT	NT	NT	40	55.5	60	50	60
Vancomycin	100	100	100	100	NT	NT	NT	NT
Cefotaxime	20	25	43	NT	77.7	80	50	60
Ceftazidime	NT	NT	46	NT	55.5	60	50	60
Erythromycin	44	50	50	NT	NT	NT	NT	NT

NT, not tested; SMX, sulfamethoxzole; TMP, trimethoprim.

cause mortality in infants with LOS and VLOS. The factors, clinical and laboratory findings associated with EOS, LOS and VLOS were also established in the present study. Therefore, we suggest that all of these data can be used for early diagnosis and prompt therapy for neonatal sepsis in NICU practice in developing countries.

Neonatal sepsis is the most common cause of neonatal mortality, which continues to be responsible for up to 50% of neonatal deaths.[19] Many specific risk factors account for the increased risk of bacterial and fungal sepsis in neonates, including use of broad-spectrum antibiotics, parenteral nutrition, and long-lasting use of invasive procedures. Preterm neonates in NICU are at high risk of intestinal disturbances with proliferation of pathogenic microflora, because treatment with antibiotics, total parenteral nutrition, or nursing in incubators may impair the intestinal colonization process. Loss of gut commensals leads to an increased susceptibility to pathogenic gut colonization. Immature or injured skin, and impaired gut barriers may especially allow dissemination of staphylococci and *Candida* spp. from colonizing sites.[20] Prompt diagnosis and effective treatment have been accepted as the milestones of management in neonatal sepsis, given that associated morbidity and attributable and overall mortality are very high.[20] Therefore, it is important to be aware of the most common causative organisms, as well as clinical and laboratory findings associated with neonatal sepsis.

Late-onset sepsis is a common complication of prolonged hospitalization in NICU that usually occurs among premature infants after the third day of life and is more often caused by Gram-positive organisms.[9,21,22] It was reported that Gram-positive organisms, the most prevalent among which is CONS, might account for 45–77% of LOS.[9,23,24] Karlowicz *et al*. reported CONS as the most common pathogen (35%) in a 10 year study that included 825 LOS cases.[22] Similarly, in another 10 year multicenter study from Australia, CONS was again defined as the primary causative agent (57%) in LOS.[13] In recent studies, CONS has also been reported as the primary causative agent, having been isolated in 50–60% of infants with LOS.[25–27] In another recent surveillance network from England, CONS was the most common organism isolated in LOS cases.[28] In agreement with these data, LOS was caused by Gram-positive bacteria including CONS, *S. aureus and Enterococcus* spp., followed by Gram-negative bacteria and *Candida* spp. in the present study. In addition, Gram-negative bacilli have also been implicated in the development of LOS in hospitalized premature infants, and *E. coli, Klebsiella*, *Enterobacter*, *Serratia*, and *Pseudomonas* were reported as the most common agents.[9] In agreement with previous reports, in the present study, the most common Gram-negative organism was *Pseudomonas* spp., which was followed by *Klebsiella* spp. and *Acinetobacter* spp. *Candida albicans* and *C. parapsilosis* were the most common species that caused LOS in the present study, as reported previously.[8] Key risk factors reported for the occurrence of LOS were prematurity, prolonged NICU stay, central vascular access, invasive procedures and the use of broad-spectrum antibiotics, and the present findings are in agreement with the literature.[8,21] In accordance with the present findings, one study performed in Turkey also reported CONS (31%) as the primary causative organism in neonatal nosocomial sepsis, followed by Gram-negative bacteria and fungi (19%).[29]

Very late-onset sepsis was detected in 28% of the present infants. These premature infants had a lower gestational age and birthweight, and most of them were ELBW. Recently, similar to the present data, Gram-positive organisms were reported to be responsible in VLOS, and also Gram-positive and fungal sepsis were found to be associated with a higher rate of mortality in VLOS.[30]

Although GBS continues to be the most important bacterial pathogen associated with EOS in many developed countries,[4] studies from developing countries report CONS as the most common causative agent in EOS.[31,32] In a recent study from Canada, however, CONS was found to be the most common organism that was identified in EOS in both term and preterm infants.[33] Low rates of invasive GBS disease in some developing countries may be due to infrequent exposure to the organism (i.e. low rates of maternal colonization), exposure to less virulent strains, genetic differences in susceptibility to disease, or high levels of transplacentally acquired protective antibody in serum.[34] In the present study, CONS was the most common agent in EOS and there was no fungal sepsis as expected. Low birthweight, prematurity, PROM, antibiotic use in pregnancy, male gender,

maternal infection, and choriamnionitis were found to be the main risk factors for EOS, which is also in agreement with the literature.[1]

In the present study, although CONS was the most common causative pathogen in all sepsis types, mortality was primarily associated with other microorganisms. We found that Gram-negative bacteria were primarily responsible for mortality in EOS. Although CONS was reported to be the most common cause of nosocomial infections in NICU, the rates of death and focal complications due to CONS were lower than those due to other organisms.[35] One possible explanation is that gentamicin, which was used as first-line therapy, had sufficient activity against CONS or at least prevented overwhelming infection. Another possibility is that CONS are often of sufficiently low pathogenicity that host defenses are able to cope with the low-level bacteremia and prevent the infection from becoming fulminant.[13,22] But CONS was found to be the third leading cause of mortality in preterm infants with LOS. Also, we established CONS as an important cause of mortality in infants with VLOS. Therefore, we suggest that CONS may be an important cause of mortality in infants with LOS and VLOS, and appropriate antibiotic therapy for CONS may be considered in the treatment of preterm infants with LOS and VLOS.

Laboratory data also might be used for diagnosis of neonatal sepsis. The present study shows that leukopenia and thrombocytopenia may be more common in, especially, Gram-negative and fungal infections in neonates. Use of several biomarkers including CRP, PCT and SAA, either alone or in combination might be beneficial in both the diagnosis and follow up of infants with neonatal sepsis. These data are also in agreement with those reported in our previous study.[36]

The microorganisms responsible for neonatal sepsis have changed over time, and they vary markedly from region to region. Prematurity, frequent use of catheters, use of total parenteral nutrition, and frequent antibiotic resistance were all reported as causes of change in the etiology of neonatal sepsis.[37] Therefore, it is necessary to be familiar with the microorganisms, antibacterial agent susceptibilities and general predominant patterns in specific institutions, regions or countries, so that prompt therapy can be targeted for the most likely bacteria when an infection is suspected. CONS is an important causal factor in all neonatal sepsis types because it is part of normal skin flora, making it difficult to avoid in NICU.[26] CONS infections are associated with low birthweight, low gestational age, history of prolonged intravascular catheterization, and longer hospital stay.[38] Although CONS is the leading causative agent of neonatal sepsis, pathogens other than CONS are associated with a higher risk of mortality in infants with EOS; but it is an important cause of mortality in infants with LOS and VLOS.

Conclusion

Neonatal sepsis remains as a significant cause of neonatal morbidity and mortality. Early diagnosis and initiation of appropriate therapy offer the best outcome for all three types of neonatal sepsis. Therefore, it is important to be familiar with the microorganisms, clinical findings and specific factors associated with neonatal sepsis in preterm infants. In the present 7 year study, CONS was found to be the primary causative agent in all sepsis types in preterm infants, and mortality was higher in the presence of sepsis caused by Gram-negative bacteria and fungi. The rate of mortality due to CONS, however, was found to be higher in infants with LOS and VLOS. We suggest that these findings might be useful for other staff working at NICU in similar developing countries. Future epidemiological and clinical studies are also needed to monitor changes in the microorganisms causing neonatal sepsis.

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