Original Research Article

Bacteriological profile of neonatal and pediatrics sepsis in intensive care unit at a tertiary care hospital in western India

Ajit Kumar Shrivastava[1], Prema Ram Choudhary[2]*, Santosh Kumar Roy[1]

[1]Department of Pediatrics, [2]Department of Physiology, Banas Medical College and Research Institute, Palanpur Gujarat, India

*Correspondence:
Dr. Prema Ram Choudhary,
E-mail: prema5252@gmail.com

ABSTRACT

Background: Neonatal and pediatrics sepsis are one of the main causes of mortality in neonatal and pediatric intensive care units of developing countries. This study was conducted to determine bacteriological profile of neonatal and pediatrics sepsis in the intensive care unit.

Methods: A prospective cross-sectional study was conducted in the neonatal and pediatric intensive care unit, for the period of two years. All 400 neonates and pediatrics patients admitted with suspected clinical sepsis were included. Sepsis screens and cultures were sent under aseptic conditions. Isolation of microorganisms and their identification was done according to standard microbiological techniques bacteriological profile was analyzed with descriptive statistics.

Results: Incidence of septicemia is 35.34% in neonates, 9.83% in post neonates and 22.95% in older children. Most common associated factor in neonates were preterm 41.46% in neonates, fever of unknown origin 50% and 78.57% in post neonates and children respectively. Out of 232 suspected cases on neonates in 36.07% cases bacterial pathogen were isolated, 62 suspected cases on post neonates in 9.83% cases bacterial were isolated and 106 suspected cases of older children in 22.95% cases bacterial pathogen were isolated. Common bacterial species isolated were *Klebsiella* sp. 39.02% in neonates, *S. aureus* 50% and 35.71% in post neonates and older children respectively.

Conclusions: There is entail prevention of infection control measures and rational antibiotic strategy to decrease the economic burden of hospital and community.

Keywords: Neonatal sepsis, Bacteriological profile, *Klebsiella pneumonia*, Pediatrics sepsis

INTRODUCTION

Neonatal septicemia describes any systemic bacterial infection in neonates documented by positive blood culture. It is an important cause of morbidity and mortality among neonates generally.[1] Neonatal sepsis may be classified according to the time of onset of the disease, as early-onset (EONS) and late-onset (LONS) neonatal sepsis.[2] Early onset sepsis (EOS) (less than 72 hours) infections are caused by organisms prevalent in the maternal genital tract or in the delivery area. Late onset sepsis (LOS) (greater than 72 hours) infections are caused by the organisms thriving in the external environments of the home or the hospital. The infection is often transmitted through the hands of the care providers. The presentation is that of, pneumonia or meningitis.[3] The most frequent cause of mortality in neonates is infections which include sepsis, meningitis, pneumonia, diarrhea and tetanus. Neonates are a vulnerable population due to their immature immune system. Other factors, such as being premature, being low birth weight, presence of respiratory problems, maternal infection, and delivery room manipulations further put neonates at risk for developing sepsis. The incidence of neonatal sepsis ranges from 7.1 to 38 cases/1000 live births in Asia in contrast to 1.5-3.5 cases/1000 live births in the United States.[4] Neonatal septicemias 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.[2] The gold standard for diagnosis of septicemia is the isolation of the microorganism from the blood culture which takes 48 hours to 7 days to confirm diagnosis. It is important to have regular surveillance of neonatal units to assess the prevailing pathogens and antibiotics susceptibility for guidance of clinicians and to identify and control the outbreaks due to cross infection.[5] About 50-88 percentage of all the neonatal deaths in the community are attributable to be caused by infection and 22 to 66 percentage of all admission in neonatal units are due to septicemia and pneumonia. The causative micro-organism may vary not only from place to place and from time to time in the same place. The Exact reason is not known but geographic, socioeconomic, seasonal and prevalent use of various antibiotics may play an important role. In developed countries group B *Streptococci* and coagulase negative *Staphylococci* are the most common cause for early onset and late onset sepsis respectively. However, in the developing countries these organisms are rare with an entirely different bacterial spectrum. Bacteriological profile of neonatal septicemia is constantly under change with advances in early diagnosis and treatment. Thus, the rational protocol for sepsis management must be based on adequate knowledge of the causative organism and their antibiotic sensitivity pattern in related area.[5] Pediatric sepsis, like sepsis in adults, is generally considered to comprise of a spectrum of disorders that result from infection by bacteria, viruses, fungi, or parasites or the toxic products of these microorganisms.[6] Septicemia, a symptomatic bacteremia, is a common condition in children (<15 years) with a resultant high morbidity and mortality.[7] Infections remain one of major problems in pediatric intensive care unit and are leading cause not only of admissions but also mortality in developing countries.[8] Mortality from pediatric sepsis ranges from 9 to 35%.[6] Children with septicemia present with fever, difficulty in breathing, tachycardia, malaise, refusal of feeds or lethargy. It is medical emergency requires urgent rational antibiotics therapy. Gold standard for diagnosis of septicemia is the isolation of bacterial agent from blood culture.[7] Only some studies have been conducted in India and in other countries to find out prevalence of neonatal and in pediatric age group. Therefore, present study was undertaken to determine prevalence of neonatal and in pediatric age group in isolated organism and common bacterial agents associated with neonatal and pediatric sepsis in a tertiary care hospital in Western India.

METHODS

The present prospective cross-sectional study was performed in department of pediatrics, at C.U. Shah medical college associated a tertiary care hospital, Surendranagar, Gujarat, Western India for duration of three years from April 2012 to March 2015. Total 400 clinically suspected patients of sepsis from neonatal and pediatric intensive care unit were selected for the study.

A detailed history was elicited and proforma was filled for each patient documenting age, sex, socioeconomic status and address, duration of illness along with type of delivery, artificial intervention birth weight and any resuscitative procedures done. Patient having respiratory distress, poor feeding, fever, hypothermia, signs of gastrointestinal or CNS involvement were included in this study. Patient with multiple congenital malformations, chromosomal disorders and newborns who were referred from other wards were excluded from present study. Informed written consent was obtained from all their relatives prior to start of study. Study protocol was approved by institutional ethics committee.

The all neonates were divided into three groups by their birth weight: normal ($\geq$2,500 gm); low birth weight (LBW) (1,500-2,500 gm); and very low birth weight (VLBW) ($\leq$1,500 gm). Finally, they were also classified into three groups by their gestational age at birth: term (>37 weeks); near-term (35-37 weeks); and preterm (<35 weeks). All patients were categorized in to three groups neonates (0 to 28 days), post neonates (29 days to 1 year) and older children (>1 year to 12 years).

Blood samples were collected from all patients aseptically before start of antibiotic therapy. All samples were transfer into brain heart infusion and sodium thioglycolate broths in the ratio of one part of blood to five parts of the broth. The blood culture broths were immediately sent to the laboratory, where they were incubated at 37°C for 7days. Three sub-cultures were made at 24 hours, 72 hours and on the 7th day of incubation on MacConkey, blood and chocolate agar media and incubated in appropriate temperature and atmospheres according to standard procedures. Gram-stained smears were made from any broth that showed visible signs of growth like generalized turbidity, discrete colonies on surface of the sediment red cells or hemolytic of red cells as this can allow an early presumptive report. Gram smear was prepared and examined at the subculture stage. Any positive finding was reported at once to the clinician as the morphological type of organism present may serve as a guide to the initial choice of antibiotic. The grown bacteria were identified by colony morphology, gram stain and standard biochemical tests were performed using standard laboratory procedures.

Statistical analysis

The collected data was analyzed using descriptive statistics with the help of IBM statistical package for the social science (SPSS) version 20.0.

RESULTS

The study was conducted at department of pediatrics, at C.U. Shah medical college associated a tertiary care hospital, Surendranagar, Gujarat. 2, 4 suspected cases of neonatal and pediatric septicemia of NICU and PICU were included in study.

Table 1: Age and gender wise distribution of all suspected cases.

Age groups	Male	Percentage (%)	Female	Percentage (%)	Total	Percentage (%)
0-28 days	178	76.72	54	23.28	232	58
29 days-1 year	38	61.29	24	38.71	62	15.5
>1-12 years	66	62.26	40	37.74	106	26.5
Total	282	70.5	118	29.5	400	100

Second common 106 (26.5%) patients were between age of 1 year to 12 years, out of 106 cases, 66 (62.26) cases were males and 40 (37.74) cases were females. Third common 62 (15.5%) patients were post neonatal age group, out of 62 cases, 38 (61.29%) were males and 24 (38.71%) were females.

Table 2: Distribution of suspected early and late onset septicemia cases.

	No.	Percentage (%)
Suspected early onset neonatal septicemia	130	56.03
Suspected eate onset neonatal septicemia	102	43.97
Total	232	100

As shown in Table 2 out of 232 cases of neonatal age group, 130 (56.03%) were suspected early onset neonatal septicemia and 102 (43.97%) were suspected late onset septicemia.

Table 3: Distribution as per associated factor.

Cases/clinical symptom	0-28 days		29 days-1 year	>1-12 year
	0-3	4-28		
Preterm	58	32	6	-
Low birth weight	36	8	-	-
Meconium aspiration	10	20	-	-
Pneumonia	8	8	14	12
Fever with jaundice	4	6	6	2
Fever with neutrophilia	2	4	-	2
Fever of unknown origins	-	8	20	68
Poor feed	-	6	10	-
Respiratory distress	4	2	-	-
Fever with neutropenia	4	-	-	-
Perinatal asphyxia	2	-	-	-
PROM	2	-	-	-
Prolonged catheterization	-	2	4	6
Altered sensorium	-	-	2	16

As shown in Table 3, out of 130 suspected early onset septicemia cases most common associated factor was preterm 58 (44.62%), second most common associated factor was Low birth weight 36 (27.69%). Similarly, two most common associated factors in late onset septicemia were preterm 32 (31.37%) and meconium aspiration 20 (19.60%), post neonatal age group fever of unknown origin 20 (32.26%) and pneumonia 14 (25.58%) and in >1 year to 12 years fever of unknown origin 68 (64.15%) and Altered sensorium 16 (15.09%).

Table 4: Blood culture results.

Blood culture results		
Specimen/results	**Number**	**Percentage (%)**
Pathogen isolated	122	30.5
Pathogen not isolated	278	69.5
Total	400	100

The Table 4 show that out of 400 cultures studied, pathogen was isolated in 122 (30.5%) specimens while no pathogen was isolated in 278 (69.50%) specimens.

Table 5: Bacterial isolates in various groups.

Group	Bacterial isolates (%)	Gram positive isolates (%)	Gram negative isolates (%)
0-3 days	44 (36.07)	22 (50)	22 (50)
4-28 days	38 (31.15)	16 (42.10)	22 (57.89)
0-28 days	82 (35.34)	38 (46.34)	44 (53.66)
29 days-1 year	12 (9.83)	6 (50)	6 (50)
>1-12 years	28 (22.95)	10 (35.71)	18 (64.29)

Table 5 shows distribution of bacterial isolates in all age group, out of 122 bacterial isolates maximum 44 (33.85%) are in 0-3 days (early neonates) group among them 50% were gram positive and 50% were gram negative, followed by 38 (37.25%) in 4-28 days (late neonates) among them 42.10% were gram positive while 57.89% were gram negative. So, in neonates (0-28 days) 82 (35.34%) cases bacteria were isolated among them 46.34% were gram positive and 53.66% were gram negative. Among post neonatal age group in 12 (19.35%)

cases bacteria were isolated among them 50% were gram positive and 50% were gram negative. Among older children in 28 (26.41%) cases bacteria were isolated among them 35.71% were gram positive and 64.29 % were gram negative.

Table 6 shows distribution of 61 bacterial isolates isolated from 400 cases. Most common isolate in early onset neonatal septicemia and late onset neonatal septicemia was *Klebsiella* sp. 40.91% and 36.84% respectively, while in post neonatal age group and older children *S. aureus* 50 and 35.71% respectively.

Table 7 shows distribution of most common associated factor in positive cases. Preterm was most common in early onset as well as late onset neonatal septicemia and fever of unknown origin was most common in post neonatal age group and older children.

Table 6: Organism isolated in different age groups.

Organism	0-3 days (22) (%)	4-28 days (19) (%)	0-28 days (41) (%)	29 days-1 year (6) (%)	>1-12 year (14) (%)
S. aureus	14 (31.82)	10 (26.32)	24 (29.67)	6 (50)	10 (35.71)
S. saprophyticus	2 (4.55)	-	2 (2.44)	-	-
S. epidermidis	2 (4.55)	2 (5.26)	4 (4.87)	-	-
CONS	-	2 (5.26)	2 (2.44)	-	-
Enterococci sp.	4 (9.09)	2 (5.26)	6 (7.32)	-	-
E. coli	2 (4.55)	8 (21.05)	10 (12.20)	-	6 (21.43)
Klebsiella sp.	18 (40.91)	14 (36.84)	32 (39.02)	-	6 (21.43)
Pseudomonas sp.	2 (4.55)	-	2 (2.44)	-	-
S. paratyphi A	-	-	-	2 (16.66)	2 (7.14)
Moraxella sp.	-	-	-	-	-
Acinetobacter sp.	-	-	-	2 (16.67)	4 (14.29)
Non-*Fermenter* sp.	-	-	-	2 (16.67)	-

Table 7: Associated factors in positive blood culture.

Cases/ clinical symptom	0-3 days (22) (%)	4-28 days (19) (%)	0-28 days (%)	29 days -1 Year (6) (%)	>1-12 year (14) (%)
Preterm	20 (45.45)	14 (36.84)	34 (41.46)	-	-
Low birth weight	10 (22.73)	-	10 (12.20)	-	-
Meconium aspiration	4 (9.09)	10 (26.32)	14 (17.07)	-	-
Pneumonia	2 (4.55)	2 (5.24)	4 (4.88)	2 (16.66)	6 (21.43)
Fever with jaundice	2 (4.55)	6 (15.78)	8 (9.75)	2 (16.67)	-
Fever with neutrophilia	-			-	-
Fever of unknown origin	-	2 (5.24)	2 (2.44)	6 (50)	22 (78.57)
Poor feed	-	2 (5.24)	2 (2.44)	-	-
Respiratory distress	2 (4.55)	-	2 (2.44)	-	-
Fever with neutropenia	2 (4.54)	-	2 (2.44)	-	-
Perinatal asphyxia	-	-		-	-
PROM	2 (4.54)	-	2 (2.44)	-	-
Prolonged catheterisation	-	2 (5.24)	2 (2.44)	2 (16.67)	-
Altered sensorium	-	-		-	-

DISCUSSION

Neonatal and pediatric septicemia is a very dangerous condition. If not diagnosed and treated in time may lead to death or irreversible complications. Due to changing pattern of bacteriological profile of neonatal and pediatric septicemia and sensitivity of microorganisms towards antibiotics, it has become very important to find out the organism causing disease. At present the emergence of β-lactamase and methicillin resistance producing pathogen is a major problem throughout the world in various clinical infections including neonatal and pediatric septicemia. Early bacteriological diagnosis will help to plan accurate, appropriate and effective therapy. In present study an attempt is made to know the aerobic bacteriology of neonatal and pediatrics sepsis with antimicrobial susceptibility testing of bacterial isolates. [9]

It was observed in present study that in all age group males were more as suspected cases of septicemia, in neonates (76.72%), in post neonates (61.29%) and older children (62.26%). Several studies done by Aletayeb, Celicia, Rabia and Ahmad et al and have reported higher number of males (compared to females) as suspected cases of septicemia in neonates which correlates with our study.[4,5,10,11] Meremikwu et al have reported higher number of males (compared to females) as suspected cases of septicemia in post neonatal age group and older children (>1year to 12 years), which also correlates with our study.[12] This may be due to gender biased care for male babies in our society.[13]

In present study early and late onset septicemia was 36.07 and 31.15% respectively. This result comparable to several previous studies which done by Jain, Rabia and Celicia et al while two studies done by Kuruvilla and Rizvi et al and have reported early onset septicemia more compared to late onset septicemia.[4,5,14-16] This may be due to mortality is higher in early onset septicemia than late onset septicemia so these two authors Kuruvilla and Rizvi et al have reported late onset septicemia more compared to early onset septicemia.

Many studies done by Rahman, Jain, Chacko, Kenneth, Shaw, Celicia, Aletayeb et al have reported positive isolates ranging from 4.1 to 62.8% in neonates.[1,4,10,14,17-19] Among these isolates gram negative isolates ranging from 36.36 to 94% and gram-positive isolates ranging from 6 to 63.64%. In present study in neonatal age group 41 (35.34%) specimen yielded positive isolates and 75 (64.66%) yielded no isolates, among these isolates 53.66% were gram negative and 46.34 % isolates were gram positive. These results are comparable with those reported in several previous studies.[14,18,19]

In present study in post neonatal age group 12 (19.35%) specimen yielded positive isolates and 50 (80.65%) yielded no isolates. These results are higher as compared to study by Tsering et al.[20] In present study in older children 28 (26.41%) specimen yielded positive isolates and 68 (73.59%) yielded no isolates. These results are higher than Hasson et al, and lower than Tsering et al.[20,21] The results of studies in relation to microbial isolates by different authors are variable as prevalence of bacterial isolate varies from place to place, hygiene of mother, mode of delivery and different geographical area. Negative cultures can be attributed to non-bacterial growth, presence of viral agents, fastidious organism, anaerobic etiology, ratio of volume of amount of blood collected and liquid broth and prior antibiotic therapy.[13,19]

In present study among neonates *Klebsiella* species (39.02%) was the most common isolate. This result is comparable with studies by Aletayeb et al while other authors Celicia and Chacko et al have reported *Pseudomonas* sp. as most common isolate.[4,10,18] Rahman and Rizvi et al have reported *E. coli* as most common

isolate.[16,17] Tsering and Meremikwu et al have reported *S. aureus* as most common isolate.[12,20]

In the present study among post neonates *S. aureus* (50%) is most common isolate. This result is comparable with Meremikwu and Tsering et al, while Gwee et al, have reported *Salmonella* sp. as most common isolate.[12,20,22]

In present study among older children *S. aureus* (35.71%) is most common isolate. This result is comparable with studies by Meremikwu and Hasson et al, while Tsering et al have reported *Pseudomonas* sp. as most common isolate.[12,20,21] This can be due to predominance of either gram positive or gram-negative bacterial isolate is influenced by geographical location.[23]

In present study in early onset neonatal septicemia *Klebsiella* sp. (40.91%) is most common isolate. This result is comparable with those reported by Aletayeb, Ahmed and Shaw et al, have reported *S. aureus* (43.08%) as most common isolate.[10,13,19] Kuruvilla et al, have reported *E. coli* as most common isolate.[15]

In present study among late onset neonatal septicemia *Klebsiella* sp. (36.84%) is most common isolate. This result is comparable with those reported by Aletayeb and Shaw et al.[10,19] While Ahmed et al, has reported *E. coli* (33.33%) as most common isolate.[11]

In present study among neonates most common associated factor is preterm (41.46%). This result is comparable with studies by Rabia, Tsering, Chacko, Jain and Ahmed et al, have reported respiratory distress as most common associated factor while another study has reported fever of unknown origin as most common associated factor.[5,13,14,18,20] This can be explained as in our study maximum number of babies was presented with history of preterm while in our set up babies admitted with meconium aspiration are less on an average.

In present study among post neonatal age group fever of unknown origin (50%) is most common associated factor. These results are contradictory to study done by Tsering et al (20) as they have reported preterm (33.03%) as most common associated factor in post neonatal age group. This can be explained as in our study most cases of post neonatal age group were > 4 to 5 months of age.

In present study among older children most common associated factor was Fever of unknown origin (78.57%). These results are contradictory to study by Tsering et al as they have reported preterm (27.08%) as most common associated factor.[20] This can be explained as in our study up to 12-year-old children are included while Tsering et al have included only up to 2-year-old children.[20]

In present study, out of 5 *E. coli* isolates in neonates 0 (0%) showed ESBL production and out of 32 *Klebsiella* species 12 (37.5%) isolates showed ESBL production. These results are comparable with study by Bhattachrjee

et al.[24] This can be explained by indiscriminately used antibiotics and inadequate doses and duration of treatment in that particular area.[11]

There are few limitations of the study, first is that, this is a prospective cross-sectional study, further a prospective cohort and case control studies are needed to investigate the interactions between clinical spectrum and bacteriological profile in neonatal and pediatric intensive care unit. Second point of consideration is that we did not include the antibiotic sensitivity patterns in intensive care unit which might be accountable for assessment of the antibiotic sensitivity and early detection of sepsis and judicial use of antibiotics are useful to decrease neonatal mortality and the emergence of multidrug resistant bacteria.

CONCLUSION

From the outcomes of our study, we conclude that incidence of septicemia is higher in neonates' males. Preterm and low birth weight neonates are more susceptible to neonatal sepsis. Gram negative organisms were the normally isolated organisms. This study emphasizes that empirical therapy for suspected neonatal septicemia should cover both gram-negative and gram-positive organisms particularly *Klebsiella pneumoniae* and *Staphylococcus aureus* which were more prevalent in this region. There is also need for regular periodic surveillance of the causative organisms of neonatal sepsis as well as their antibiotic susceptibility patterns to curtail the inappropriate use of antibiotics and emergence of resistant strains and review the hospital antibiotic policy from time to time. There is a need to implement antimicrobial warden ship programmes to diminish antibiotic usage to reduce neonatal mortality due to sepsis. Early find out of sepsis and sensible use of antibiotics are useful to diminish neonatal mortality and the emergence of multidrug resistant bacteria.

REFERENCES

1. Kenneth CI, Olufmlayo YE, Iretiola BB. Bactriological profile of neonatal septicaemia in a tertiary hospital in Nigeria. Afri Health Sci. 2006;6(3):151-8.
2. Amela S, Fahrija S, Mustafa B, Zijad S. The Predictive score for early-onset neonatal sepsis. The Turkish J Pediatr. 2010;52:139-44.
3. Ghai OP, Paul VK, Bagga A. Essential pediatrics. CBS Publishers & Distributors Pvt Ltd. 2009;7:136-8.
4. Cecilia CM, Mary AC, Elizabeth EG, Jonathan GL, Joanne JL, Cecille YA. Etiology of neonatal sepsis in five urban hospitals in the Philippines. PIDSP J. 2011;12:75-85.
5. Rabia S, Nusrat K, Shugfta H. Bacteriology and Anti-Microbial Susceptibility of Neonatal Septicemia in NICU, PIMS, Islamabad- A Tertiary Care Hospital of Pakistan. Ann. Pak Inst Med Sci. 2010;6(4):191-5.
6. Santhanam S, Steele RW. Pediatric Sepsis. www.emedicine.medscape.com/article/972449-overview. Accessed on 09-06-2012.
7. Nwadioha SI, Nwokedi EO, Kashibu E, Odimayo MS, Okwori EE. A review of bacterial isolates in blood cultures of children with suspected septicaemia in a Nigerian tertiary Hospital. Afr J Microbiol Res. 2010;4(4):222-5.
8. Khan H, Khaliq N, Muhammad F. Pediatric Intencive Care Unit; Pattern of Admissions. Professional Med J. 2006;13(3):358-61.
9. Povoa P. C-reactive protein: a valuable marker of sepsis. Intensive Care Med. 2002;28:235-43.
10. Aletayeb SM, Khosravi AD, Dehdashtian M, Kompani F, Mortazavi SM, Aramesh RM. Identification of bacterial agents and antimicrobial susceptibility of neonatal sepsis: A 54-month study in a tertiary hospital. African J Microbiol Res. 2011;5(5)528-31.
11. Ahmad A, Hussain W, Lamichhane A, Muhammad A, Riaz L. Use of antibiotics in Neonatal sepsis at neonatal unit of a tertiary care hospital. Pak paed J. 2011;35(1):3-7.
12. Meremikwu MM, Nwachukwu CE, Asuquo AE, Okebe JU, Utsalo SJ. Bacterial isolates from blood cultures of children with suspected septicemia in calabar Nigeria. BMC infectious diseases. 2005;5:110-14.
13. Ahmed AN, Caowdhury MA, Hoque M, Darmstadt GL. Clinical and bacteriological profile of neonatal septicemia in a tertiary level pediatric hospital in Bangladesh. Indian pediatr. 2002;39:1034-39.
14. Jain NK, Jain VM, Maheshwari S. Clinical profile of Neonatal sepsis. Kathmandu university med j. 2003;1(2):117-20.
15. Kuruvilla KA, Pillai S, jesudason M, Jana AK. Bacterial profile of sepsis in a neonatal unit in south India. Indian paediatr. 1998;35:851-58.
16. Rizvi F, Afzal M, Khan A, Wahid S. Bacterial sensitivity in neonatal sepsis. J Islamabad med Dental college. 2007;5(2):153-60.
17. Rahman S, Hameed A, Roghani MT, Ullah Z. Multidrug resistant neonatal sepsis in Peshawar, Pakistan. Arch Dis Child Fetal Neonatal. 2002;87:52-4.
18. Chacko B, Sohi I. Early onset neonatal sepsis. Indian J paediatr. 2005;72(1):23-6.
19. Shaw CK, Shaw P, Thapalia A. Neonatal sepsis bacterial isolates and antibiotic susceptibility pattern at aNICu in a tertiary care hospital in western Nepal: A retrospective analysis Kathmandu University Med J. 2007;5(2):153-60.

20. Tsering D, Chanchal L, Pal R. Bacteriological profile of septicemia and risk factors in neonates and infants in Sikkim. J global infect disease. 2001;3(1):42-5.
21. Hasson SO, Naher HS, AIMrzoq JM. Gram positive bacteremia in febrile children under two years of age in Babylon province. Available at: www.uoba-bylon.edu.iq/uobcoleges/fleshare/articles/gram.pdf. Accessed on 20 June 2012.
22. Gwee A, Coghlan B, Evereti D, Chagoma N, Phiri A, Wilson L et al. Bacteremia in Malawian Neonates and young infants 2002-2007: A retrospective audit. BMJ Open. 2012;2(3).
23. Qureshi M, Aziz F. Prevalence of microbial isolates in blood cultures and their antimicrobial susceptibility profiles. Biomed. 2011;27:136-9.
24. Bhattaccharjee A, Sen MR, Prakash P, Gaur A, Anupurba S. Increased prevalence of Extended spectrum B lactamase producers in neonatal septicemic cases at a tertiary referral hospital. Indian J Med Microbiol. 2008;26(4):356-60.