Bacteriological profile and associated risk factors of neonatal sepsis in Paropakar Maternity and Women's Hospital Thapathali, Kathmandu

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ABSTRACT

Neonatal Sepsis is one of the most common reasons for admission to neonatal units in developing countries. It is also a major cause of mortality in both developed and developing countries. Identification of the common bacteria and risk factors causing such infections and their susceptibility patterns will provide necessary information for timely intervention. This study was carried out to determine the bacteriological profile and associated risk factors of neonatal sepsis in Paropakar Maternity and Women's hospital. A cross – sectional descriptive study was conducted among neonates suspected of neonatal sepsis. Blood culture was performed and organisms were identified and antibiotic susceptibility was carried out with standard microbiological methods. Data were analysed by using SPSS. Ver. 16 software. The positive yield of blood culture was 21%. The most common isolates were *Staphylococcus epidermidis, E. coli,Staphylococcus aureus, Klebsiella pneumoniae* and *Pseudomonas* spp. In Antibiotic susceptibility pattern Gentamycin showed the highest sensitivity to all types of isolated organisms. Vancomycin sensitivity was highest for Gram positive organism and Ciprofloxacin was most effective for Gram negative organisms isolated. Ampicillin and Amoxycillin were the least effective drug. Multiple drug resistance was observed in 77.15% of isolates. Prematurity, low birth weight and maternal pyrexia before delivery were found to be strongly associated with neonatal sepsis. Gram positive organisms were more prevalent than gram negative organisms.

Keywords: Neonatal sepsis, Antimicrobial susceptibility pattern, multiple drugs resistant

INTRODUCITON

Neonatal sepsis is one of the commonest causes of neonatal mortality in the developing world accounting for 30-50 percent of total neonatal deaths per year.[1,2] About 99% of the approximate 1 million annual neonatal deaths from life-threatening invasive bacterial infections occur in developing countries, at least 50% of which are from home births or community settings.[3] Neonatal septicemia is a clinical syndrome of bacteremia characterized by systemic signs and symptoms of in first month of life. It encompasses systemic infections of newborn including meningitis, pneumonia, arthritis, osteomyelitis and urinary tract infections of the newborn.[4] Neonatal sepsis may be classified according to the time of onset of the disease: early onset sepsis (EOS) and late onset sepsis (LOS).[5,6] The distinction has clinical relevance, as EOS disease is mainly due to bacteria acquired before and during delivery, and LOS disease to bacteria acquired after delivery (nosocomial or community sources).[7] Early onset sepsis usually presents within the first 72 hours of life and Late onset sepsis usually presents after 72 hours of age.[8] This study was carried out to determine the bacterial etiological agents of neonatal sepsis and the antibiotic sensitivity pattern of the etiological agents as well as the clinical profile of the culture proven sepsis.

MATERIALS AND METHODS

The descriptive cross sectional study, of the cases admitted to the neonatal intensive care unit (NICU) and Premature baby unit (PBU) of Paropakar Maternity and Women's Hospital, Thapathali, Kathmandu, Nepal, was carried out during the period starting from August 2011 to March 2012. The total of 452 neonates who presented with signs and symptoms of septicaemia (Complement reactive protein (CRP) positive i.e ≥6mg/L, Birth Asphyxia, Fever, Grunting, Poor sucking, Respiratory distress, Tachypnoea), with/ without pneumonia and/or meningitis were included in this study. The neonatal and maternal data were obtained by filling the questionnaire designed for the study from

patient record file with the help of medical officers and the staff nurses after taking oral consent from the parents/ guardians. One milliliter of blood was drawn aseptically before starting antimicrobial treatment and inoculated directly into 5ml Brain Heart Infusion broth (BHI) in a ratio of blood: BHI of 1:6, followed by inoculation in the appropriate culture media from Hi-media which includes Mac Conkey Agar, Nutrient Agar, Blood Agar, Chocolate Agar and Muller Hinton Agar. Growth, if any, was identified by the standard bacteriological techniques, including Gram staining, colony characteristics, biochemical properties, and slide agglutination where appropriate.[9] Antibiotic sensitivity was performed by Kirby-Bauer disk diffusion technique that is recommended by Clinical Laboratory Standards Institute (CLSI) recommendations.[10] The various antibiotics used for susceptibility testing are as follows: Amoxycillin, Ampicillin, Gentamycin, Tobramycin, Amikacin, Cefotaxime, Ciprofloxacin, Vancomycin and Cloxacillin manufactured by Hi-media. The cases with suspected sepsis were started empirically on antibiotics, which were changed according to the sensitivity pattern (Kirby-Bauer method) once the culture report was available. The following aspects were studied:

1. The prevalence of different organisms causing neonatal sepsis in the hospital.
2. The antibiotic susceptibility pattern.
3. The risk factors of sepsis in case of culture proven sepsis.
4. Statistical analysis (Chi-square test) of the data was performed by using SPSS ver. 16 software and the P-value was determined.

OBSERVATION AND RESULTS

In total 452 neonates admitted with suspected cases of sepsis were studied. Of the total 452 patients, 269(59.5%) were males and 183(40.5%) were females resulting in an overall male to female ratio of 1.47:1. A total of 438(96.9%) neonates presented with Early onset sepsis (EOS) and 14(3.1%) presented with Late onset sepsis (LOS). Among the neonates with suspected EOS, 260(59.4%) were males and 178(40.6%) were females. Among neonates with suspected LOS 9(4.3%) were males and 5(35.7%) were females. Common clinical features observed in both EOS and LOS, were, Complement reactive protein (CRP) positive, Birth Asphyxia, Fever, Grunting, Poor sucking, Respiratory distress, Tachypnoea, Thick meconium stained liquor (TMSL) and Jaundice. Among the 452 neonates admitted with suspected cases of sepsis, 94 (21%) had positive blood culture for bacteria. *Staphylococcus epidermidis* accounted for 57.4% of the total isolates followed by *Escherichia coli* (26.6%), *Staphylococcus aureus* (10.6%), *Klebsiella pneumoniae* (4.3%), and *Pseudomonasspp.* (1.06%). The Gram – positive and negative bacteria accounted for 64/94 (68%) and 30/94 (32%) respectively. The two neonates expired due to *E. coli* sepsis during the study period which accounts 0.44% out of total neonates admitted with suspected sepsis. Table 1 shows the bacterial etiologic agents isolated. Neonatal and Maternal Risk factors associated/Not associated with culture proven sepsis are outlined in Table 2.

Table 1. Bacterial etiologic agents isolated from blood culture in neonates with suspected cases early – onset sepsis and late – onset sepsis.

Etiologic agents	Early onset sepsis No. (%)	Late onset sepsis No. (%)	Total No. (%)
Staphylococcus epidermidis	51 (57.3)	3 (60)	54 (57.4)
E. coli	25 (28.1)	-	25(26.6)
Staphylococcus aureus	10 (11.2)	-	10 (10.6)
Klebsiella pneumoniae	2 (2.2)	2(40)	4 (4.3)
Pseudomonas spp.	1 (1.1)	-	1 (1.06)
Total	89 (94.7)	5 (5.3)	94 (100)

Table 2. Neonatal and maternal risk factors associated with culture proven neonatal sepsis

Variables	Culture positive (n=94) No. (%)	Culture Negative (n=358) No. (%)	P - value
Gestational age			
<37 weeks (Pre term)	63 (67)	186 (52)	0.009
37 – 42 weeks (Term)	26 (27.7)	161 (45)	
>42 weeks (Post term)	5 (5.3)	11 (3)	
Weight at birth			
1500 – 2499g (LBW)			
2500 – 4000g	43 (45.7)	106 (29.6)	
(Normal)	50 (53.2)	242 (67.6)	0.01
>4000 (Overweight)	1 (1.1)	10 (2.8)	
Place of delivery			
Hospital	94 (100)	356 (99.4)	0.468
Home	0	2 (0.6)	

LBW: Low Birth Weight
PROM: Premature rupture of membrane.
P-value<0.05 is considered statistically significant

Among these, preterm neonates (p=0.009), neonates with low birth weights (p=0.01), Maternal Pyrexia before delivery (p=0.00) had a statistically significant association with culture proven neonatal sepsis.

The antibiotic sensitivity pattern of bacterial isolates are shown in Table 3.

Table 3. Antibiotic sensitivity patterns of Bacterial Isolates.

Antimicrobial agents		Organisms				
		E.coli **(n=25) No. (%)**	*Klebsiella pneumoniae* **(n=4) No. (%)**	*Pseudomonas* **spp. (n=1) No. (%)**	*Staphylococcus epidermidis* **(n=54) No. (%)**	*Staphylococcus aureus* **(n=10) No. (%)**
Ampicillin	S*	4(16)	-	-	15(27.7)	4(40)
Amoxycillin	S*	4(16)	-	-	14(26)	4(40)
Gentamycin	S*	17(68)	3(75)	1	47(87)	5(50)
Tobramycin	S*	8(32)	2(50)	1	22(40.7)	4(40)
Amikacin	S*	12(48)	2(50)	-	44(81.5)	7(70)
Cefotaxime	S*	13(52)	3(75)	1	31(57.4)	3(30)
Ciprofloxacin	S*	21(84)	3(75)	1	32(59.3)	5(50)
Vancomycin	S*	-	-	-	54(100)	10(100)
Cloxacillin	S*	-	-	-	35(64.8)	6(6 0)

***S**: Sensitive

Among Gram- negative isolates, low level (<60%) of resistance were observed with Gentamycin, Amikacin, Cefotaxime and Ciprofloxacin. *E.coli* showed intermediate level (60% – 80%) of resistance to Amoxycillin and Ampicillin whereas other isolates were 100% resistant to these antibiotics. In general Ciprofloxacin was found to be most effective drug against Gram – negative bacteria followed by Gentamycin. Among Gram-positive isolates, low level (<60%) of resistance were observed to most antibiotics tested: Gentamycin,Tobramycin, Amikacin, Vancomycin, Cloxacillin, Cefotaxime and Ciprofloxacin. All isolates showed intermediate level (60 – 80%) of resistance to Ampicillin and Amoxycillin. In general Vancomycin was found to be most effective drug against Gram – positive bacteria followed by Gentamycin and Amikacin. Multiple resistances (resistance to two or more drugs) was observed in 54/64 (84.3%) of Gram – positive bacteria and 21/30 (70%) of Gram-negative bacteria.

DISCUSSION

The causative organisms in neonatal sepsis vary from place to place and the frequency of the causative organisms is different in different hospitals and even in the same hospital at different time. In the present study, 21% of neonates had microbiologically confirmed sepsis. This finding was in agreement with the findings (19.56%, 20%, 22%) reported by previous studies done in Nepal. [11-13] However, variable results have been reported by various studies from different parts of Nepal.[14,15,19] Administration of prior antibiotics from primary centre, infection with anaerobes or effective control in spread of nosocomial infection, might be the reasons for the variable results in different studies. [14]

In the present study most common isolates after Coagulase negative staphylocci (CoNS) were *E.coli* followed by *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Pseudomonas* spp. The other published data in Nepal on the subject shows *E.coli* as the most common isolate.[16,17] The study carried out in western Nepal showed *Staphylococcus aureus* to be the most common isolate.[18] Coagulase negative staphylococci (CoNS) was the most common isolate in the NICU of a University hospital in Turkey and in neonatal ward of teaching hospital in Urmia, Iran.[19,20] In our study *Staphylococcus epidermidis* (CoNS) were the most common isolates (57.4%) causing neonatal sepsis followed by *E.coli* (26.6%)and *Staphylococcus aureus* (10.6%). This result is different from the other studies in Nepal where *E.coli* was the leading cause.[16,17] *E. coli* was the second most common isolate as reported from Uganda.[21] *Staphylococcus aureus* was the third most common isolate as in India.[22] *Klebsiellapneumoniae* was found to be the fourth most common isolate but the other report in Nepal and India showed *Klebsiellapneumoniae*as the second most common cause,[16,22] and in West Indies showed as the most common cause.[23] *Pseudomonas* spp.was isolated from one case in this study but the reports from Iran and India showed *Pseudomonas* spp. to be the most common cause of neonatal sepsis.[24,25] Both gram positive and gram negative bacteria showed high level of resistance to Amoxycillin and Ampicillin as in the other studies from Nepal .[17,18] Vancomycin was the most effective drug against Gram positive isolate. Ciprofloxacin was the most effective drug against gram negative. 77.15% of the isolates were multiple drugs resistant. In this study pre-term neonate, neonates with low birth weight were at risk in developing sepsis as reported in previous studies in Nepal.[26,27] Maternal pyrexia before delivery had statistically significant association with culture proven sepsis($p<0.05$).

The causative organisms of neonatal sepsis vary with

time and place. There is increasing trend of antibiotic resistance to the commonly used and available drugs. Continuous surveillance is needed to monitor changing epidemiology of pathogens and antibiotic susceptibility pattern.

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