Bacteriological Study of Early Onset and Late Onset Neonatal Septicaemia in a Tertiary Care Hospital in South India

Mamatha P Samaga[1], Keerthi B J[2], Sini Joseph[3]

ABSTRACT

Introduction: Neonatal septicaemia is a clinical syndrome characterised by signs and symptoms of infection with or without accompanying bacteraemia in the first month of life. It is classified into early onset sepsis within 72 hour of life and late onset sepsis after 72 hour. The varying microbiological pattern of septicemia and their high antibiotic resistance needs to be studied.

Material and Methods: This retrospective observational study was conducted for a period of one year. The bacterial isolates and their antibiogram from blood samples of clinically suspected neonatal septicaemic cases were studied from the records of Microbiology Laboratory.

Results:Among 350 blood samples collected from clinically suspected cases of neonatal septicaemia,50(14.2%) were culture positive. Among 50 isolates, 41(82%) were from Early onset septicaemia and 9(18%) were from Late onset septicaemia. The predominant isolate in Early Onset Septicaemia and Late Onset Septicaemia was Klebsiella pneumoniae and Escherichia coli respectively followed by Staphylococcus aureus. Klebsiella showed high resistance to all the antibiotics and was most sensitive to meropenem (82.3%). All other Gram negative bacteria were 100% sensitive to Meropenem, Imipenem and Amikacin.The Gram positive bacteria showed 92% resistance to penicillin and were 100% sensitive to vancomycin and linezolid.

Conclusion: In our study, Klebsiella pneumoniae was predominantly isolated. The susceptibility of the bacteria to the commonly used antibiotics was low and needs increased efforts to ensure rational use of antibiotics.

Keywords: Early Onset Septicaemia, Escherichia coli, Klebsiella Pneumoniae, Late Onset Septicaemia, Neonatal Septicaemia, Staphylococcus Aureus

INTRODUCTION

Neonatal septicaemia is a clinical syndrome characterised by signs and symptoms of infection with or without accompanying bacteraemia in the first month of life.[1] Neonatal sepsis is associated with significant morbidity and mortality throughout the world.[2] Though sepsis is a cause of neonatal deaths in the developed countries the scenario is more serious in developing countries, where neonatal sepsis is responsible for 30-50% of neonatal mortality.[3] Incidence of Neonatal septicaemia in India is 30/1000 live births.[1]

The risk factors for neonatal septicemia include premature rupture of membranes, prolonged rupture, prematurity, Urinary Tract Infection, poor maternal nutrition, Low Birth Weight, birth asphyxia and congenital anomalies.[4]

Depending on the onset of symptoms, it can be classified into early onset sepsis(EOS) within 72 hour of life and late onset sepsis (LOS) after 72 hour of life.[5] The importance of this classification is that it helps to guide the antibiotic therapy by implying differences in the mode of transmission and the predominant causative organisms.[6]

The bacteria most commonly associated with EOS include Group B Streptococcus (GBS), Coagulase negative Staphylococcus (CONS), Escherichia coli, Haemophilus influenzae and Listeria monocytogenes and LOS is caused by CONS, Staphylococcus aureus, Escherichia coli, Klebsiella spp., Pseudomonas spp., Enterobacter spp., Group B Streptococcus, Serratia spp., Acinetobacter spp. and anaerobes.[7]

Neonatal sepsis is difficult to diagnose clinically as it presents with non-specific signs and symptoms.[8] Though various diagnostic modalities exist for neonatal sepsis including c-reactive protein, complete blood count, platelet count and erythrocyte sedimentation rate, yet blood culture is the gold standard.[9]

The varying microbiological pattern of septicemia in children warrants the need for an ongoing review of the causative organisms and their antimicrobial susceptibility pattern. The incidence of bacteremia in children varies widely.[10] The emergence of the resistant bacteria in the NICU leads to failure in the treatment of neonatal sepsis.[8] Multidrug antibiotic resistance is an emerging problem in NICU particularly in developing countries.Also, the spectrum of organisms that cause neonatal sepsis changes from time to time and varies from region to region.[11]

This study was conducted to know the bacteriological profile of early and late onset neonatal septicaemia along with the antibiotic susceptibility patterns and thus help the clinician in the accurate diagnosis and treatment of neonatal septicaemia.

MATERIAL AND METHODS

The study was conducted after obtaining approval from the Institutional Ethical Committee. This was a retrospective observational study conducted for a period of one year (Jan-Dec 2015). The bacterial isolates and their antibiogram from the blood samples of clinically suspected neonatal septicaemic cases were studied from the records of the Microbiology Laboratory. The neonates with septicemia were divided into early onset septicemia (EOS, within 72 hour of age) and late onset septicemia (LOS,after 72 hour of age). 2ml blood

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drawn under aseptic precautions and inoculated into 20 ml brain heart infusion broth (Hi-Media, India) were received in the Microbiology laboratory. These were incubated at 37° C under aerobic conditions in the incubator for 7 days. The first subculture was done after 24 hours of incubation, the second on the third day and a final on the seventh day onto Blood agar and MacConkey agar plates. The inoculated plates were incubated aerobically at 37° C for 24 hours, and observed for growth. The growth was identified by Colony morphology, Gram's stain and Standard biochemical tests.[12] Antibiotic sensitivity testing was performed on Mueller-Hinton agar by modified Kirby-Bauer disk diffusion method as per Clinical Laboratory Standard Institute guidelines.[13]

Bacteria	EOS	LOS	Total
Escherichia coli	4	4	8 (16%)
Klebsiella pneumoniae	15	2	17(34%)
Staphylococcus aureus	8	3	11(22%)
Citrobacter spp	5	0	5(10%)
Acinetobacter spp	3	0	3(6%)
Proteus vulgaris	2	0	2(4%)
Proteus mirabilis	1	0	1(2%)
Coagulase negative Staphylococci	3	0	3(6%)
Total	41(82%)	9(18%)	50(100%)

Table-1: Frequency of various bacteria in EOS and LOS.Figure in paranthesis indicate percentage. EOS-Early Onset Septicaemia, LOS- Late Onset Septicaemia.

STATISTICAL ANALYSIS

Descriptive statistics was used to analyse the data and data is presented as percentage.

RESULTS

A total of 350 blood samples were collected from clinically suspected cases of neonatal septicaemia. Among them, 50 samples were culture positive. Thus the culture positivity rate was 14.2%. Among 50 isolates, 41(82%) were from Early onset septicaemia and 9(18%) were from Late onset septicaemia (Figure 1). The predominant isolate in EOS was Klebsiella pneumoniae followed by Staphylococcus aureus, Citrobacter spp, Escherichia coli and others. The predominant isolate in LOS was Escherichia coli followed by Staphylococcus aureus and Klebsiella pneumoniae as shown in Table 1. Antibiotic sensitivity pattern of Gram negative and Gram positive bacteria are shown in the Tables 2 and 3.

DISCUSSION

The blood culture positivity rate in our study was 14.2%. Martin et al.[14] reported bacteriologically proven sepsis in 9.5% of the cases, whereas a lower rate of 4.1% was reported by Aletayeb et al.[15] Higher culture positivity rates of 48%[16] and 64%[17] among neonates with sepsis has been reported.

A low blood culture isolation rate could be due to anaerobic, viral or fungal pathogens. Also it may be due to the various changes that have occurred with increasing awareness of prevention of sepsis like early and more aggressive enteral feeding and better

Antibiotics	E.coli N=8	Klebsiella N=17	Citrobacter N=5	Proteus vulgaris N=2	Proteus mirabilis N=1	Acinetobacter N=3
Amikacin	8(100)	3 (17.6)	5 (100)	2(100)	1(100)	3(100)
Ampicillin	2(25)	1 (5.9)	1 (20)	-	-	1(33.3)
Ciprofloxacin	2(25)	6 (35.3)	2 (40)	2(100)	0	3(100)
Meropenem	8(100)	14 (82.3)	5 (100)	2(100)	1 (100)	3(100)
Cefotaxime	3 (37.5)	2 (11.7)	3 (60)	1(50)	1(100)	2 (66.7)
Piperacillin-Tazobactam	4(50)	6 (35.3)	5 (100)	2(100)	1(100)	3(100)
Imipenem	8(100)	13 (76.5)	5(100)	2(100)	1(100)	3(100)
Gentamicin	1 (12.5)	4 (23.5)	2 (40)	1(50)	0	1 (33.3)
Ceftazidime	2(25)	1 (5.9)	2 (40)	1(50)	0	3(100)
Cefepime	3 (37.5)	1 (5.9)	3 (60)	2(100)	1(100)	3(100)
Ceftriaxone	3 (37.5)	1 (5.9)	2 (40)	1(50)	1(100)	1 (33.3)

Table-2: Antibiotic sensitivity pattern of Gram negative bacteria. Figures in paranthesis indicate percentage.

Antibiotics	Staphylococcus aureus N=11	CONS N=3
Penicillin	1 (9)	0
Erythromycin	5 (45.4)	2 (66.7)
Ciprofloxacin	6 (54.5)	2 (66.7)
Gentamycin	5 (45.4)	1 (33.3)
Vancomycin	11(100%)	3(100%)
Linezolid	11(100%)	3(100%)
Tetracycline	3 (27.2)	0
Oxacillin	5(45.4)	1 (33.3)
Azithromycin	4 (36.7)	1 (33.3)

Table-3: Antibiotic sensitivity pattern of Gram positive bacteria. Figures in paranthesis indicate percentage. CONS-Coagulase negative Staphylococcus.

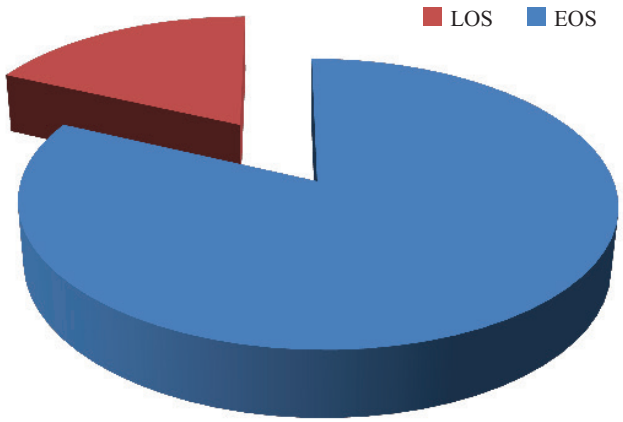

Figure-1: Prevalence of bacteria in Early and Late onset septicaemia.

hand hygiene practices.

In our study, we noted a predominance of Gram negative bacteria (72%).A study conducted in Karnataka reported 70.5% neonatal septicemia cases caused by Gram-negative bacteria.[18] Gram-negative bacteria were predominantly isolated in other studies also.[19,20]

We found that EOS (82%) was more in this study compared to LOS(18%) which is consistent with other reports from Nepal[21] and Bangladesh.[22]

In our study, Klebsiella pneumoniae (36.6%) was the major isolate in EOS followed by Staphylococcus aureus (19.5%), Citrobacter (12.1%), Escherichia coli (9.8%) and others. Escherichia coli (44.4%) was the major isolate in LOS followed by Staphylococcus aureus (33.3%) and Klebsiella pneumoniae (22.2%).

EOS is caused mainly by bacteria transmitted from mothers to neonates during the intrapartum period, these are the bacteria prevalent either in the maternal genital tract or in the area of delivery. LOS is caused by postnatal acquisition of the pathogens, caused by the bacteria which thrive in the external environment of the hospital or home.[6]

Zakariya et al in their study reported Klebsiella pneumoniae as the commonest (74.4%) isolate in EOS and CONS were the second common isolate.[23] Klebsiella pneumoniae is commonly found in the environment of the neonatal intensive care units and nursery. It can also be present as colonizers on the hands of the health care workers. There are also frequent reports of neonatal septicemia outbreaks due to Klebsiella pneumoniae in nursery and NICUs.[24]

We noted that Acinetobacter spp. was isolated in 3 cases of EOS. Acinetobacter spp. causing septicemia in neonates is reported by Arora *et al.*[25] and Vinodkumar *et al.*[26]

Among gram positive bacteria, we noted that Staphylococcus aureus was predominantly isolated (22%).It was the second most common isolate in EOS(19.51%) and LOS (33.3%).CONS was isolatedin 3 cases of EOS. Staphylococcus aureus as a major pathogen of neonatal septicemia has been reported by Karthikeyan *et al.*[27]

In our study, Klebsiella showed high resistance to almost all the antibiotics and was most sensitive to meropenem (82.3%). All other Gram negative bacteria were 100% sensitive to Meropenem, Imipenem and Amikacin. Except Klebsiella and Escherichia coli, all other gram negative bacteria were 100% sensitive to Piperacillin-Tazobactum combination.High resistance was seen for ampicillin and gentamycin which are most commonly used first line antibiotics.

The gram positive bacteria showed 92% resistance to penicillin. Kumhar etal and Iregbu etal also reported a high level of penicillin resistance.[28,29] 100% sensitivity was noted for vancomycin and linezolid. Vancomycin was also found as the most effective antibiotic in studies of Desai and Malek and Rajendraprasad BP etal.[30,18]

Neonatal septicemia remains as an important and challenging problem even with modern and advanced diagnostics and drug therapy. Hospital data should be generated regularly about the spectrum of bacteria and their antibiotioc susceptibility pattern to enable accurate diagnosis and empirical treatment.

Limitation of the study

This was a retrospective study and sample size was small. We could not correlate with neonatal morbidity and mortality.A more extensive research should be conducted to study the bacterial spectrum, antibiotic resistance pattern and treatment outcome in neonatal septicaemia.

CONCLUSION

In our study, Klebsiella pneumoniae was predominantly isolated. The prevalence of Early Onset Septicaemia was more than Late Onset Septicaemia. The most effective antibiotic for gram negative bacteria was imipenem and the most effective antibiotic for gram positive bacteria was vancomycin.The susceptibility of the pathogens to the commonly used antibiotics was low and needs increased efforts to ensure rational use of antibiotics. Antibiotic resistance can cause difficulties in the treatment of sepsis, such as increase in the mortality rate, duration of hospitalization and treatment expenses.A regular antibiotic susceptibility surveillance and periodic review of the antibiotic policy of the hospital will reduce the development of antibiotic resistance.

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