



E-ISSN: 2706-9575
P-ISSN: 2706-9567
IJARM 2021; 3 (1): 510-514
Received: 04-01-2021
Accepted: 06-03-2021

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A study of bacteriological profile in neonatal sepsis in neonatal intensive care unit in a tertiary care centre

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DOI: <https://doi.org/10.22271/27069567.2021.v3.i1i.193>

Abstract

Background: Septicaemia in neonates refers to a generalized bacterial infection in the first four weeks of life that is confirmed by a positive blood sample. It is one of India's four leading causes of neonatal death and morbidity.

Aim: To study the bacteria responsible for neonatal sepsis in Neonatal Intensive Care Unit in a tertiary care centre.

Methods: 100 neonates with symptoms and signs suggestive of sepsis were included in this study. A written informed consent was taken from either of parents of the babies who were included in the study. All babies included in the study were subjected to detailed maternal and neonatal history taking. A detailed clinical examination of the new born was done and gestational age was assessed by New Ballard's score chart and the findings recorded in the pre-structured proforma.

Results: In the present study, out of the 100 cases with symptoms and signs suggestive of sepsis, 23 cases (23%) were culture positive. *Klebsiella pneumoniae* was isolated in 2 (28.58%) cases and *Enterococcus* in 2 (28.58%) of cases, *Staph. aureus*-Coagulase +ve in 1 (14.28%) cases, *Pseudomonas* spp. in 1 (14.28%) cases, *Enterobacter* spp. in 1 (14.28%) cases isolated indicating that *Klebsiella pneumoniae* was the most frequent organism isolated in late onset sepsis. *Enterobacter* and *Enterococcus* were isolated only late onset sepsis and not seen in early onset sepsis.

Conclusion: *Klebsiella pneumoniae* and *Staph. aureus*-Coagulase +ve are the leading cause of neonatal sepsis in this study and most of them are resistant to multiple antibiotics. Therefore the results of this study suggest that, surveillance of antimicrobial resistance in our hospital is necessary.

Keywords: sepsis, septicaemia, *Klebsiella pneumoniae*, *Staph. aureus*

Introduction

Neonatal sepsis is a clinical syndrome characterized by signs and symptoms of infection with or without accompanying bacteraemia in the first month of life. It encompasses various systemic infections of the new born such as septicaemia, meningitis, pneumonia, arthritis, osteomyelitis, and urinary tract infections^[1].

According to pooled hospital data based on National Neonatal Perinatal Database 2002-2003 (NNPD) survey, the incidence of neonatal sepsis around is 30/1000 live births. Neonatal infections are estimated to cause, about 1.6 million deaths worldwide and 40% of all neonatal deaths due to sepsis occur in developing countries^[2].

The Mortality rate due to neonatal sepsis in developing countries is 26%^[1]. Neonatal sepsis contributes significantly to morbidity and mortality among young infants. Classically sepsis has been differentiated into early onset and late onset sepsis based on onset before 72 hours or later life up to 28 days. These 2 types of sepsis have different risk factors or settings. Clinical outcome varies in neonatal sepsis. Early onset sepsis is caused by organisms prevalent in the genital tract or in the labour room and maternity operation theatre. Mainly early onset sepsis was caused by *E. coli*, Group B *Streptococci* (in west), *Klebsiella* sp., *Staph. aureus*. (NNPD Report 2002-2003).^[2] Late onset sepsis was caused by of gram-negative bacilli (*Klebsiella pneumoniae*, *E. coli*, *Pseudomonas aeruginosa*, *Proteus* spp., *Citrobacter*, Enterobacteria) in two-thirds of cases^[3-9].

Bacterial responsible for neonatal sepsis varies from place to place and also from time to time. Antibiotics have been used extensively in the management of sepsis. On many occasions, antibiotics have been used empirically without identifying the causative organisms or knowing the antibiotic sensitivity, leading to development of resistance.

It is therefore necessary to note which are the common organisms causing sepsis in our area

and their sensitivity to antibiotics. This will help us to use appropriate antibiotics and reduce the development of antibiotic resistance. Hence this study has been conducted to identify the organisms causing sepsis in the new born and to determine the antibiotic sensitivity pattern of common bacteria isolated in our area.

Materials and Methods

The present study was a hospital based prospective observational study conducted in Neonatal Intensive Care Unit, Department of Paediatrics,

Period of study: From January 2019 to December 2020.

Study sample: All babies admitted to Neonatal Intensive Care Unit with the clinical suspicion of sepsis were taken up for the study. Sepsis was clinically suspected if the neonate had symptoms and signs suggestive of sepsis [1] such as – poor feeding, poor activity – respiratory distress, retractions, nasal flaring, apnea. cyanosis. – seizure, lethargy, bulging anterior fontanel – fever, hypothermia – Abdominal distension, vomiting, jaundice.

Sample size: 100 neonates with symptoms and signs suggestive of sepsis.

Inclusion criteria

All babies admitted to the Neonatal Intensive Care Unit, with symptoms and signs suggestive of sepsis.

Exclusion criteria

- Babies who received antibiotics previously.
- Babies with major congenital anomalies.
- Very low birth weight (<1500gms).

Methodology

The present study was approved by the institutional ethics committee, prior to the start of the study.

A written informed consent was taken from either of parents of the babies who were included in the study. All babies included in the study were subjected to detailed maternal and neonatal history taking. A detailed clinical examination of the new born was done and gestational age was assessed by New Ballard’s score chart and the findings recorded in the pre-structured proforma.

All the Data related to history, clinical findings and investigations was entered in pre-structured proforma.

Statistical analysis: The data was managed on the Microsoft Excel spread sheet. All the entries were checked and the data was analysed using SPSS software version 21.

Ethical clearance: Ethical Clearance was taken from the institution ethical clearance committee prior to the commencement of the study.

Observation and Results

Table 1: Distribution based on gender, birth weight, gestational age and age of onset

Gender	Number of babies n (%)
Male	73 (73)
Female	27 (27)
Total	100 (100)
Birth weight	
1500gms – 2500gms	59 (59)
> 2500gms	41 (41)
Gestational age	
Preterm	64 (64)
Term	36 (36)
Age of onset	
Onset <72 hours (Early onset)	83 (83)
Onset >72 hours (Late onset)	17 (17)

In the present study, out of the 100 neonates admitted in the Neonatal Intensive Care Unit with clinical suspicion of sepsis, 73 (73%) of the babies were males and 27 (27%) were female babies, indicating that male babies constituted a majority.

In the present study, out of the 100 new-borns studied, 59 (59%) babies weighed less than 2500gms and 41 (41%)

babies weighed \geq 2500gms, indicating that there is a predominance of low-birth-weight babies weighing <2500gms.

In the present study, 64 (64%) babies admitted with clinical suspicion of sepsis were preterm, and 36 (36%) were term, indicating that majority of the babies studied were preterm and almost double the number of term babies.

Table 2: Distribution based on investigations, bacterial growth and blood and urine culture

Investigations	Number of babies (%)
C-reactive proteins >1mg/dl (positive)	87 (87%)
Abnormal total count	40 (40%)
<5000/mm ³	25 (25%)
>15000/mm ³ leukocyte	15 (15%)
Platelet count <1 lakh/dl	32 (32%)
Bacterial growth	
Culture Positive	23 (23%)
Culture Negative	77 (77%)
Body fluids	
Blood culture positive	21 (91.30%)
Urine culture positive	2 (8.70%)

Haematological investigations done in the babies having symptoms and signs suggestive of sepsis showed that C - reactive protein was found to be positive in 87% cases. Total Leukocyte count was abnormal in 40%, out of these 25% babies had less than 5000/mm³ and 15% babies had more than 15000/mm³. Thrombocytopenia (Platelet count < 1 lakh) was seen in 32% cases.

In the present study, out of the 100 cases with symptoms and signs suggestive of sepsis, 23 cases (23%) were culture

positive and in 77% cases, organisms were not grown.

In the present study, out of 100 cases studied, 23 cases had bacterial growth, in which 21 cases (91.30%) were blood culture positive and 2 cases (8.70%) were urine culture positive for bacterial growth. Cerebrospinal Fluid (CSF) culture was done in all blood culture positive cases and clinically suspected meningitis, but CSF culture revealed no growth of organisms.

Table 3: Distribution of cases based on bacteria isolated and onset of sepsis

Onset of sepsis	Gram positive sepsis n (%)	Gram negative sepsis n (%)	Total cases n (%)
<72 hours Early onset sepsis	5 (21.74)	11 (47.82)	16 (69.56)
>72 hours Late onset sepsis	3 (13.04)	4 (17.40)	7 (30.44)
Total cases	8 (34.78)	15 (65.22)	23 (100)

Chi-square = 0.0038, P Value = 0.47

Out of 23 cases with positive culture, Gram negative organisms were predominantly isolated comprising of 15 cases (65.22%). 8 cases (34.78%) had isolated Gram positive organisms. Culture positivity in early onset sepsis

was 69.56% compared to late onset sepsis, where only 30.44% was culture positive. P value is 0.47 which is not significant.

Table 4: Distribution of cases of type of bacteria isolated and bacterial culture positivity based on the onset of sepsis

Bacteria isolated N = 23	Early onset sepsis N = 16 (%)	Late onset sepsis N = 7 (%)
<i>Staph. aureus</i> - coagulase +ve	3 (18.75)	1 (14.28)
Coagulase -ve <i>Staph. aureus</i>	2 (12.50)	0
<i>Enterococcus</i>	0	2 (28.58)
<i>Esherichia coli</i>	2 (12.50)	0
<i>Pseudomonas</i> spp.	1 (6.25)	1 (14.28)
<i>Klebsiella pneumoniae</i>	5 (31.25)	2 (28.58)
<i>Citrobacter</i>	1 (6.25)	0
<i>Acinetobacter</i>	2 (12.50)	0
<i>Enterobacter</i>	0	1 (14.28)
Total	16 (100)	7 (100)

In the present study, 23 babies had shown growth in the culture. In the 23 babies with culture positivity, the predominant organism isolated was *Klebsiella pneumoniae* (30.43%), followed by *Staphylococcus aureus* (17.39%), *Esherichia coli* (8.7%), *Pseudomonas* spp. (8.7%), *Acinetobacter* (8.7%), Coagulase -ve *Staph. aureus* (8.7%), *Enterococcus* (8.7%), *Citrobacter* (4.34%), *Enterobacter* (4.34%).

In the present study, out of 16 cases of Early onset sepsis, *Klebsiella pneumoniae* was isolated in 5 (31.25%) of cases, *Staph. aureus* - Coagulase in 3 (18.75%) cases, Coagulase negative *Staph. aureus* in 2 (12.50%) cases, *Esherichia coli* in 2 (12.50%) cases, *Pseudomonas* spp. in 1 (6.25%) cases,

Citrobacter in 1 (6.25%) cases and *Acinetobacter* in 2 (12.50%) cases, indicating that *Klebsiella pneumoniae* was the most frequently grown organism in Early onset sepsis.

In the present study, out of 7 cases of late onset sepsis, *Klebsiella pneumoniae* was isolated in 2 (28.58%) cases and *Enterococcus* in 2 (28.58%) of cases, *Staph. Aureus* - Coagulase +ve in 1 (14.28%) cases, *Pseudomonas* spp. in 1 (14.28%) cases, *Enterobacter* spp. in 1 (14.28%) cases isolated indicating that *Klebsiella pneumoniae* was the most frequent organism isolated in late onset sepsis. *Enterobacter* and *Enterococcus* were isolated only late onset sepsis and not seen in early onset sepsis.

Table 5: Antibiotic sensitivity pattern in gram positive and gram negative isolates

Sensitivity of gram positive organisms	
Antibiotic	n (%)
Vancomycin	7 (87.5)
Ciprofloxacin	7 (87.5)
Linezolid	4 (50)
Cefotaxime	4 (50)
Amikacin	4 (50)
Gentamicin	3 (37.5)
Co-trimoxazole	1 (25)
Sensitivity of gram negative organisms	
Antibiotic	Number (%)
Ciprofloxacin	13 (86.66)
Gentamycin	12 (80)
Amikacin	12 (80)

Meropenem	8 (53.3)
Piperacillin/tazobactem	4 (33.3)
Cefotaxime	4 (33.3)
Ceftazidime	3 (26.6)
Vancomycin	2 (13.3)
Amoxicillin clavulanate	2 (6.7)

In the present study, out of the 8 cases which had grown Gram positive organisms, 7 cases (87.5%) were sensitive to vancomycin and ciprofloxacin, 4 cases (50%) were sensitive to linezolid, amikacin and cefataxime, 3 cases (37.5%) to Gentamycin and only 1 case was sensitive to co-trimoxazole (25%).

In the present study, Gram negative isolates were most sensitive to ciprofloxacin (86.6%), followed by gentamycin (80%) amikacin (80%), meropenam 53.3%, piperacillin/tazobactem 33.3%, cefataxime 33.3%, and 26.6% to ceftazidime. Gram negative isolates had low sensitivity to amoxicillin-clavulanate (6.7%).

Table 6: Distribution of cases in the present study based on outcome

Out come	No. of babies N (%)
Recovered/Discharged	97 (97)
Death	3 (3)

In the present study, out of the 100 neonates admitted with sepsis, 97 (97%) babies recovered, whereas 3 babies i.e. 3% had died.

Discussion

In the study done by Pooja *et al.* [10] the gram-negative organisms were susceptible to Imipenem (86.13%), Meropenem (83.22%), piperacillin/tazobactam (76%) and fluroquinolones (74.5%). These findings are not comparable with this study, where Ciprofloxacin was sensitive in 86.6% of cases and Meropenems are sensitive only in 53.3% cases. In the study done by Bhatt S *et al.* [11] the antibiotic sensitivity pattern revealed that the majority of Gram-negative isolates (*Klebsiella* species and *Escherichia coli*) were sensitive to Meropenem (95% and 97%), Piperacillin and tazobactam (88% and 91%), Levofloxacin (84% and 81%) and Ampicillin and sulbactam (58% and 49%) respectively and these findings are not comparable with this present study in where meropenem sensitivity was only 53.3% and Piperacillin and tazobactam only 33.3%.

In the present study gram negative organisms were sensitive to ciprofloxacin which is comparable with the studies done by Pooja *et al.* [10] and Bhatt S *et al.* [11]. Gram negative organisms were highly sensitive to Amikacin in the present study and was comparable to studies done by Pooja and Bhatt S *et al.* Cefotaxime sensitivity was low in this study comparing with Pooja and Bhatt *et al.* explanation for this could be this drug being used as empirical treatment in neonatal sepsis in our Neonatal Intensive Care Unit.

In the present study gram negative bacteria were sensitive to Ciprofloxacin, Amikacin and Gentamycin which was not comparable with the studies done by Premalatha [12] and Shivakumar [13]. Wherein the gram-negative bacteria were more sensitive to Imipenem's and resistance to Gentamycin and Amikacin.

A study done by Srinivasa *et al.* Gram-positive group had greater susceptibility to higher antibiotics vancomycin 97%, linezolid 91.2%, cephalosporin 84%, quinolones 70.1% in

order and low susceptibility to ampicillin 31.5% these findings were comparable with present study [14].

In study done by Desai *et al.* [15] Gram positive isolates were maximum sensitivity to Vancomycin (100%). Gram negative isolates were maximum sensitivity to Piperacillin-tazobactam (98%). These findings were not comparable to the present study in where Gram negative isolates were more sensitive to Ciprofloxacin, Gentamycin, Amikacin. In our study, *Klebsiella* was the commonest organism grown which was highly sensitive to Amikacin, Gentamycin and Ciprofloxacin.

This study was having limitations with regard to the sample size. A larger sample size would be better to derive the bacteria responsible for sepsis and the antibiotic sensitivity in our area.

Conclusion

In this study, *Klebsiella pneumoniae* and *Staph. aureus* - Coagulase +ve were the most common causes of neonatal sepsis. The majority of them are immune to a variety of antibiotics. As a result, the authors recommend that antimicrobial resistance be monitored. In addition, the hospital should develop an antibiotic policy. Antibiotics should be used based on the isolates' antibiotic susceptibility pattern. Furthermore, we recommend that the public be educated about the risks of indiscriminate antibiotic usage, which is still considered a threat in our community and has been linked to the ineffectiveness of most widely used antibiotics like penicillin and ampicillin, as found in our research.

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