

# A STUDY ON MICROORGANISMS AND THEIR ANTIMICROBIAL SUSCEPTIBILITY PATTERN ISOLATED FROM CLINICALLY SUSPECTED CASES OF NEONATAL SEPSIS IN A TERTIARY CARE CENTRE AT BIHTA

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## ABSTRACT

**Background:** Sepsis is most common cause of neonatal mortality. The profile of microorganism and its susceptibility pattern helps in guiding the empirical therapy and prevent misuse of antibiotics. This study was done with an aim to know the profile of microorganism in clinically suspected cases of neonatal sepsis in the neonatal intensive care units (NICU) setting and their antibiotic susceptibility pattern. **Materials and Methods:** The study was conducted for 1 year period in a tertiary care hospital at Bihta, Patna. All the blood samples received from clinically suspected cases of neonatal sepsis in NICU, were subjected to culture and sensitivity by standard microbiological procedures. **Result:** A total of 199 blood samples were received from NICU. The blood culture positive rate in NICU was 14.6%. Among culture positive samples, 18 (62.07%) were Coagulase negative Staphylococcus species (CONS), followed by 05 (17.24%) Klebsiella pneumoniae, Streptococcus viridans 02 (6.89%), and 01 each (3.45%) species of Enterococcus, Acinetobacter, Enterobacter and Burkholderia cepacia. Among Gram positive cocci (GPC), the most common isolates were CONS. Klebsiella pneumoniae was the most common isolates among gram negative bacilli (GNB) and were multidrug resistance. **Conclusion:** Multi drug resistance was an alarming finding, this highlights the importance of implementing stewardship program in hospital to rationalize the use of antibiotics. Thus preventing the antibiotic resistance.

## INTRODUCTION

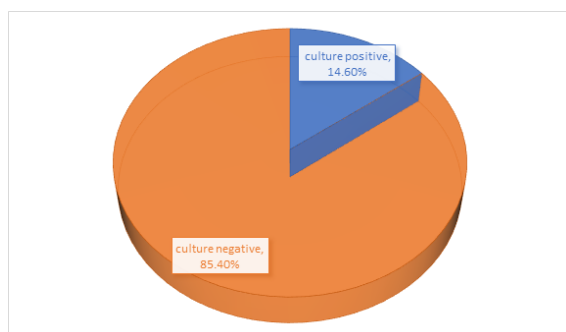
The most common cause of neonatal mortality is sepsis, responsible for one third of neonatal death across the globe.<sup>[1]</sup> The profile of microorganisms that causes neonatal sepsis changes with time and geographical distribution.<sup>[2]</sup> There is frequent emergence of resistant microorganism, suggesting the need for surveillance of neonatal sepsis for optimum therapy.<sup>[3]</sup> The aim of study was to identify microorganisms from clinically suspected cases of neonatal sepsis and its antimicrobial susceptibility pattern.

## MATERIALS AND METHODS

This prospective study was conducted in a tertiary care hospital in Bihta, Bihar over a period of 1 year from March 2024 to February 2025. The study was done after getting approval by the clinical research ethics committee of Netaji Subhas Medical College and Hospital, Bihta, Patna via letter no: CREC/2024/46, dated 28/2/2024. Informed consent were obtained from neonates parents before enrolling into the study. All neonates (0–28 days) clinically suspected of having neonatal sepsis admitted in the NICU of our hospital during the study period were included. Neonates were suspected for sepsis by the clinician on the basis of clinical symptoms based on international recommendation.<sup>[4]</sup> Demographic, clinical data and laboratory parameters were filled for

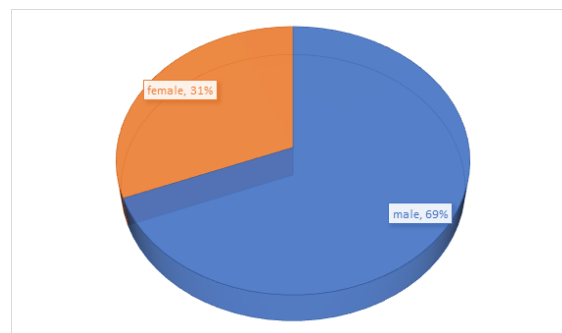
each neonate on a specific proforma. Under aseptic conditions, blood samples were collected. 0.5-1ml of blood were inoculated into BacT/ Alert PF plus culture bottle and incubated. After the bottle were flagged positive, subculture was done on blood agar, chocolate agar and Macconkey agar and incubated at 37°C for overnight incubation. A gram stain was also performed from the broth for presumptive identification of microorganisms. The bacterial isolates were identified based on colony morphology, gram stain, motility and biochemical tests.<sup>[5]</sup> The antimicrobial susceptibility pattern was determined by Kirby Bauer disc diffusion method according to CLSI 2024 guidelines. <sup>[6]</sup> The data were entered in Microsoft Excel and SPSS software was used for statistical analysis. Descriptive data were analyzed statistically and expressed as percentage.

## RESULTS



**Figure 1: distribution based on blood culture result**

A total of 199 blood samples were received from clinically suspected cases of neonatal sepsis admitted in NICU for culture and sensitivity. Among the clinically suspected cases of neonatal sepsis, culture positive samples were 29 (14.6%) and culture negative sample were 170 (85.4%). [Figure 1]



**Figure 2: distribution based on sex of culture positive sample**

Male predominance 20 (69%) was seen as compared to female 09 (31%) in culture positive samples. [Figure 2]

Most of the culture positive samples 22 (75.9%) were from pre term neonates and remaining 07 (24.1%) were from term neonates. Majority of culture positive samples 24 (82.8%) were from low birth weight (LBW) neonates while 05 (17.2%) were from normal weight neonates. Among culture positive sample in clinically suspected cases of neonatal sepsis, 19 (65.52%) were early onset sepsis (EOS) and 10 (34.48%) were late onset sepsis (LOS). [Table 1]

**Table 1: Distribution of culture positive sample based on demographic details of neonates**

Demographic details	Number	Percentage
Pre term neonates	22	75.9%
Term neonates	07	24.1%
Total	29	100%
Low birth weight	24	82.8%
Normal birth weight	05	17.2%
Total	29	100%
Early neonatal sepsis	19	65.52%
Late neonatal sepsis	10	34.48%
Total	29	100%

Among culture positive samples, CONS 18 (62.07%) followed by *Klebsiella pneumoniae* 05 (17.24%), *Streptococcus viridans* 02 (6.89%), and 01(3.45%)

each species of *Enterococcus*, *Acinetobacter*, *Enterobacter* and *Burkholderia cepacia* were isolated. [Table 2].

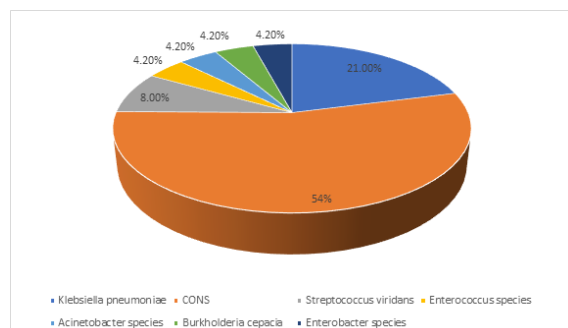
**Table 2: Distribution of bacterial isolates in culture positive samples (N = 29)**

GNB (N =08)		
1.	<i>Klebsiella pneumoniae</i>	05 (17.24%)
2.	<i>Acinetobacter species</i>	01 (3.45%)
3.	<i>Burkholderia cepacia</i>	01 (3.45%)
4.	<i>Enterobacter species</i>	01 (3.45%)
GPC (N =21)		
6.	CONS	18 (62.07%)
7.	<i>Streptococcus viridans</i>	02 (6.89%)
8.	<i>Enterococcus species</i>	01 (3.45%)

**Table 3: Distribution of organism based on LOS (N=5)**

Organism	Percentage
CONS	100%

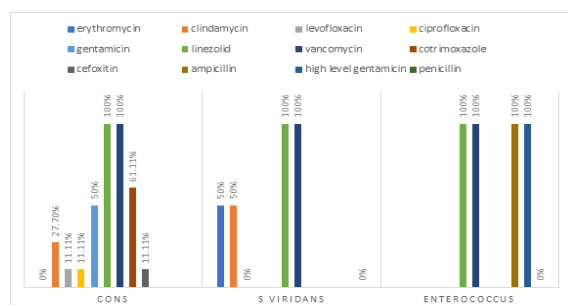
In EOS, the predominant organism was CONS 13 (54.2%), followed by *Klebsiella pneumoniae* 05 (21%), *Streptococcus viridans* 02 (8%), 01 (4.2%) each species of *Enterococcus* species, *Acinetobacter* species, *Burkholderia cepacia* and *Enterobacter* species respectively. [Figure 3].



**Figure 3: distribution of organism on basis of EOS (N=24)**

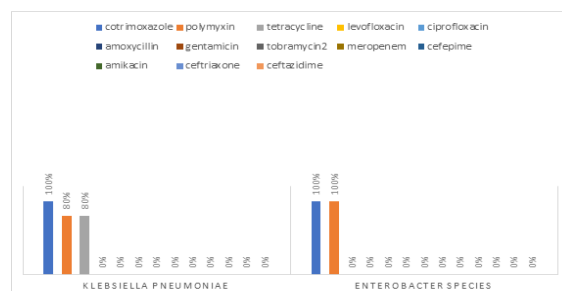
The organism isolated in LOS in this study was CONS 05 (100%). [Table 3].

Among GPC, 16 (88.89%) isolated CONS were methicillin resistant. Highest sensitivity in CONS was seen in vancomycin, linezolid (100%), followed by cotrimoxazole (61.11%) and gentamicin (50%). *Enterococcus* species was 100% sensitive to vancomycin, linezolid, ampicillin and high level gentamicin. *Streptococcus viridans* was 100% sensitive to vancomycin, linezolid followed by erythromycin, clindamycin (50%) respectively. [Figure 4]



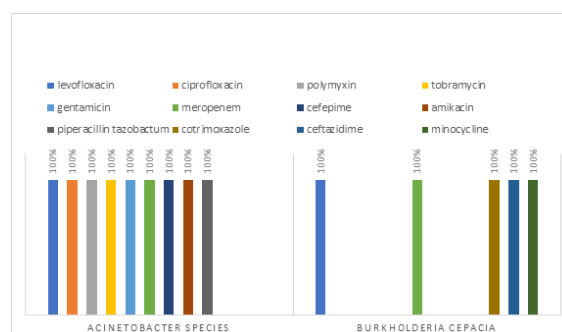
**Figure 4: Percentage sensitivity to antimicrobial agents shown by GPC**

Among GNB, *Klebsiella pneumoniae* was the most resistant isolates. It showed 100% sensitivity towards cotrimoxazole and 80% sensitivity towards polymyxin and tetracycline. *Enterobacter* species was 100% sensitive to cotrimoxazole, polymyxin and piperacillin tazobactam. [Figure 5]



**Figure 5: Percentage sensitivity to antimicrobial agents shown by members of enterobacteriaceae family**

*Acinetobacter* species showed 100% sensitivity towards levofloxacin, ciprofloxacin, polymyxin, tobramycin, gentamicin, meropenem, ceftazidime, amikacin and piperacillin tazobactam. *Burkholderia cepacia* showed 100% sensitivity towards levofloxacin, cotrimoxazole, meropenem, ceftazidime and minocycline. [Figure 6]



**Figure 6: Percentage sensitivity to antimicrobial agents shown by non fermenters**

## DISCUSSION

In this study, the culture positive neonatal sepsis rate was 29 (14.6%). This finding is in accordance with the study of Khadka P *et al*<sup>[7]</sup> who reported 18.7% culture positive neonatal sepsis. Lower blood culture positive rate may be due to sepsis caused by fastidious microorganisms which cannot grow on routine culture media.

Male predominance 20 (69%) was seen as compared to female 09 (31%) in culture positive neonatal sepsis. Siddiqui *et al*<sup>[8]</sup> in their study reported similar findings. In their study they reported culture positive neonatal sepsis sample was higher in male (53.5%) as compared to female (46.5%).

In this study, majority of culture positive sample (82.8%) were from LBW neonates. Similar findings were observed by Yadav NS *et al*<sup>[9]</sup> in their study. They also observed a higher positive blood culture (62.7%) in LBW neonates.

Neonatal sepsis was more common in preterm neonates 22 (75.9%) as compared to term neonates 07 (24.1%). The results in this study is same as in the study of Chauhan H *et al*<sup>[10]</sup> They also reported neonatal sepsis was more common in preterm neonates (60.38%) than in term neonates (39.62%).

Among culture positive, 19 (65.52%) and 10 (34.48%) were diagnosed as EOS and LOS respectively. This finding is in accordance with study of Rafi M.A et al.<sup>[11]</sup> They diagnosed EOS as 61.5% and 38.5% as LOS cases.

The predominant organism isolated was CONS 62.07%. This is in accordance with study of Nazir A et al.<sup>[12]</sup> Sah R et al.<sup>[13]</sup> Jin Z et al.<sup>[14]</sup> They also isolated CONS as the predominant organism in neonatal sepsis. If proper aseptic techniques are not done while collecting blood samples, CONS are considered as a contaminant. In this study, we consider CONS as pathogenic organism as other evidence of sepsis like increased C reactive protein (CRP), procalcitonin level and clinical condition of neonates were looked into while reporting the organism.

*Klebsiella pneumoniae* was the most commonly isolated GNB from clinical suspected cases of neonatal sepsis. This finding was similar with previous studies conducted by Yadav N et al.<sup>[9]</sup> The susceptibility pattern of *Klebsiella pneumoniae* was matter of concern. As it showed only 80% susceptibility towards polymyxin. Similar findings were observed by Pandita N et al.<sup>[15]</sup> They reported polymyxin susceptibility as 87% for *Klebsiella pneumoniae*. All isolates of *Klebsiella pneumoniae* were resistant to cephalosporins, aminoglycosides, fluoroquinolones and carbapenems. This may be due to indiscriminate use of antibiotics and no strict policy to reinforce effective antibiotic stewardship program.

Methicillin resistant CONS was (88.89%) in this study. This is in accordance with study of Nazir A et al.<sup>[12]</sup> They reported 84% isolates of CONS as methicillin resistant. The increased prevalence of methicillin resistant CONS strains, nowadays may be due to overuse of third-generation cephalosporin as suggested by Vanitha et al.<sup>[16]</sup>

#### Limitation

Due to lack of resources, molecular confirmation of drug resistant gene could not be done. Further speciation of CONS could not be done. This study findings is based on a single tertiary care hospital NICU so the findings cannot be generalized to other hospital NICU. This was a cross sectional study so follow up of neonates was not done.

## CONCLUSION

In this study, CONS was the predominant isolate followed by *Klebsiella pneumoniae* in clinically suspected sepsis cases. Multi drug resistance was an alarming finding, this highlight the importance of implementing stewardship program in hospital to rationalize the use of antibiotics. Thus preventing the antibiotic resistance.

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