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**Original Research Article**

# Blood culture positive sepsis and sensitivity pattern in NICU at a tertiary care neonatal centre

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## ABSTRACT

**Background:** This study aims to study blood culture positive neonatal sepsis and the sensitivity pattern of pathogenic organisms in a tertiary neonatal care hospital.
**Methods:** This retrospective observational cohort study was done in a tertiary care hospital. All the blood culture positive neonatal sepsis cases, excluding neonates with multiple congenital malformations, diagnosed during March 2023 to August 2023 (6 months) were analyzed.
**Results:** A total of 73 neonatal sepsis cases were diagnosed, among them 40 (54.7%) cases of gram-positive sepsis. In present study 61.6% cases were early onset sepsis, 41 (56%) babies were preterm and 49 (67%) babies were inborn. *Staphylococcus aureus* and Acinetobacter were the most common organisms in early onset and late onset sepsis respectively. The sensitivity of Gram-negative bacilli to colistin, minocycline and levofloxacin was 90%, 84% and 78% respectively. The sensitivity of gram-positive organisms to linezolid, vancomycin and teicoplanin were 92%, 73% and 70% respectively. Mortality rate among culture positive sepsis was 16.4%.
**Conclusions:** In the practice of modern neonatal care, multidrug organisms are emerging. By practicing antibiotic stewardship, infants can be protected from future multidrug resistance organisms.

**Keywords:** Antibiotic stewardship, Neonate, Sepsis, Sensitivity

## INTRODUCTION

In India, 20% of neonatal deaths are related to sepsis.[1] Sepsis occurs in 30 out of every 1000 live births in hospital-based research and in 2.7–17% of all live births in community-based studies.[2,3] Neonatal sepsis is divided into two groups based on when the illness first manifested: early onset and late onset. Most early-onset sepsis-causing bacteria come from the vaginal tract of mothers. Nosocomial infections or community-acquired infections are the culprits of late-onset sepsis.

Nosocomial infections result from a very low birth weight neonate's prolonged hospital stay, the usage of central lines, catheters and breathing support for their life. The therapy of newborn sepsis has significant problems due to the growing incidence of multidrug resistance bacteria in the neonatal intensive care unit (NICU) and the gradual introduction of newer medications. Strict adherence to the antibiotic stewardship program will aid in addressing the current and upcoming sepsis management issue. Monitoring the profile of antibiotic resistance and the pattern of causative organisms on a regular basis is one step towards antibiotic stewardship.[5]

Early empiric antibiotic treatment of neonates suspected of having septicemia is the standard practice. Nonetheless, the problem of unnecessary exposure to antibiotics in this vulnerable population remains, creating an environment for emerging bacterial resistance and the potential for poor outcomes.[3] Also definitive culture results takes at least 48-72 hours resulting in treatment

delays. Hence, a battery of tests called sepsis screen which includes acute phase reactants, X-ray chest, urine examination and blood culture, is used to diagnose septicaemia early and initiate a presumptive treatment. Isolation of bacteria from a central body fluid (usually blood) is the standard and the most specific method to diagnose neonatal septicaemia.[4]

In our nation, the prevalence of organisms in both early and late onset newborn sepsis is different from that of the western world. A unit's bacterial ecology and resistance pattern can fluctuate over time. The goal of the current study was to assess the sensitivity pattern of the causal organisms for newborn sepsis in a tertiary care NICU.[5]

## METHODS

This retrospective observational cohort study was done in a tertiary care neonatal center (SMIMER medical college, Surat), in western India.

Patient case records of blood culture confirmed sepsis cases admitted to the new-born intensive care unit during March 2023 to August 2023 (total duration 6 months) were examined using a pre-defined protocol for assessment. Purposive Sampling technique was used for this study.

From the case records, gestational age, onset of disease in days, sex, birth weight, inborn/out born cases, identified organisms with sensitive pattern and survival outcome were recorded for analysis using Microsoft excel. Descriptive summary statistics were used to describe demographic profile of cases, culture identified organisms, sensitive pattern and clinical outcome. Distribution of sensitive pattern by gram positive and gram-negative status were also described. The distribution of multi drug resistance (MDR) among each gram-negative species were tabulated and described. A multidrug resistance is defined as microorganisms, predominantly bacteria, that are resistant to one or more classes of antimicrobial agents.[11]

For blood culture 1.5-2 ml venous blood sample was collected within six hours of birth using aseptic technique, 0.5-1 ml of which was directly transferred into the culture vials (containing blood agar and mac conkey agar). All the samples were transported at room temperature as soon as possible to Microbiology laboratory for further processing, by manual lawn culture method. After incubation at 37ºC the bacterial isolates were identified by gram staining, colony characteristics and a battery of biochemical tests, following a standard protocol.[6]

### *Inclusion criteria*

All the blood culture positive neonatal sepsis cases admitted to NICU.

### *Exclusion criteria*

Neonates with multiple congenital malformations. Neonates with incomplete data Neonates with complex congenital heart disease.

### *Ethical approval*

This study was approved by institutional ethical committee.

## RESULTS

During the study period, total admissions were 1051, 73 (7%) blood culture positive cases of neonatal sepsis were analysed. Out of 73 cases, 40 (54.7%) babies had gram positive sepsis, early onset sepsis was found in 45 (61.6%) cases. In our study, males were more affected, Inborn babies and preterm had higher rate of sepsis positivity (Table 1). Among all neonates, culture positive sepsis was also more common in LBW (Table 1).

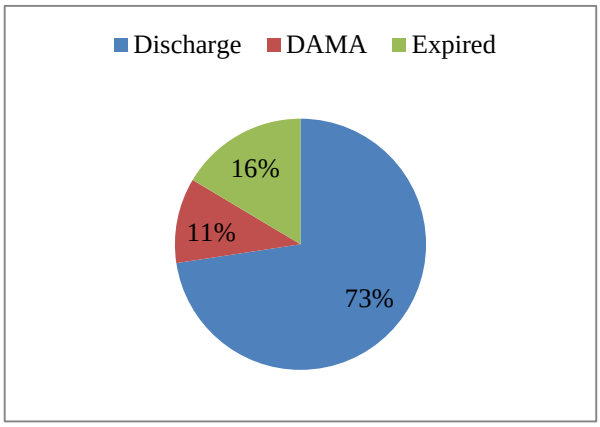


**Figure 1: Clinical outcome.**

Among the culture positive sepsis, rate of gram-positive sepsis was more than gram negative sepsis, incidence of early onset sepsis was higher than late onset sepsis (Table 2). Frequency of isolated organisms was staph aureus (28.7%), Acinetobacter (18%), enterococus (19.1) and kleibsella (5.4%) in our study.

Among gram positive culture, 28.7 were staph aureus followed by enterococcus in 19% (Table 3). Staphylococcus was the predominant organism in early onset sepsis (EOS) and Acinetobacter was the leading cause of late onset sepsis (LOS). Overall gram-positive organism were common isolates in EOS and gram negative in LOS (Table 3).

Sensitivity of gram-negative organisms to colistin, minocycline, levofloxacin was 90%, 84% and 78% respectively. Meropenam was sensitive in only 42% cases (Table 4). Majorities (69%) of gram-negative organisms were resistant to piperacillin and tazobactum and to amikacin 66%. The sensitivity to linezolid, vancomycin

and teicoplanin for gram positive organisms were 92%, 73%, 70% respectively. Resistance of gram-positive organisms was high in amoxicillin-clavulanate (65%) (Table 4). Sensitivity of staph aureus was high for linezolid, levofloxacin and vancomycin. Resistance for amoxicillin-clav was 71% (Table 5).

Sensitivity of enterococcuus was high for linezolid, levofloxacin and tigecycline. High resistance was noted for amoxicillin-clav and cephalosporins (Table 5). *S. aureus* was sensitive to linezolid in 90.4%, levofloxacin in 80.9% and tigecycline. It was resistant to vancomycin (71.4%) in 28.5% (Table 5). Acinetobacter species was sensitive in all cases (100%) to colistin. Kleibsella was sensitive to colistin in 75% and to amikacin in 75% (Table 6). Out of 73 babies, twelve babies died and eight neonates were left the unit before completion of treatment. Seven neonates with gram negative sepsis and 6 babies with gram positive sepsis had died during the study period. Mortality was 16.4%, of these contribution of gram positive and negative organisms was almost equal (Table 7).

Among the study group, rate of discharge was 73%, DAMA was 11%. Total death was noted to be 16% (Figure 1).

**Table 1: Demographic data.**

| Sex | N (%) |
|---|---|
| **Male** | 37 (50.6) |
| **Female** | 36 (49.3) |
| **M:F** | 1.03:1 |
| **Inborn** | 49 (67) |
| **Outborn** | 24 (32) |
| **Term** | 32 (43) |
| **Preterm** | 41(56) |
| **Birth weight** | |
| <1 kg | 4 (5.4) |
| 1-1.5 kg | 18 (24.6) |
| 1.5-2.5 kg | 33 (45.2) |
| >2.5 kg | 18 (24.6) |

**Table 2: Distribution of organisms.**

| Total | n=73 |
|---|---|
| **Gram positive organisms** | 40 (54.7) |
| **Gram negative organisms** | 33 (45.2) |
| **EOS** | 45 (61.6) |
| **LOS** | 28 (38.3) |

**Table 3: Organisms in EOS and LOS.**

| Gram negative organisms | Total cases (%) | EOS | LOS | Gram positive organisms | Total cases (%) | EOS | LOS |
|---|---|---|---|---|---|---|---|
| **Acinetobacter** | 18 (24.6) | 10 | 8 | *Staph aureus* | 21 (28.7) | 14 | 7 |
| **Enterbacter** | 6 (8.2) | 2 | 4 | Enterococcus | 14 (19.1) | 12 | 2 |
| **Kleibsella** | 4 (5.4) | 2 | 2 | Cons | 3 (4.1) | 3 | 0 |
| ***E coli*** | 3 (4.1) | 0 | 3 | Streptococcus | 2 (2.7) | 2 | 0 |
| **Peudomonas** | 1 (1.3) | 0 | 1 | | | | |
| **Citrobaccter** | 1 (1.3) | 0 | 1 | | | | |

**Table 4: Sensitivity of organisms.**

| Antibiotic for gram negative organisms | Sensitive (%) | Resistance (%) | Antibiotic for gram positive organisms | Sensitive (%) | Resistance ( %) |
|---|---|---|---|---|---|
| **Meropenem** | 14 (42) | 17 (51) | Vancomycin | 29 (72) | 11 (27) |
| **Piperacillin tazobactam** | 10 (30) | 23 (69) | Teicoplanin | 28 (70) | 9 (22) |
| **Amikacin** | 11 (33) | 22 (66) | Linezolid | 37 (92) | 3 (7.5) |
| **Cefepime** | 10 (30) | 20 (60) | Levoflox | 34 (85) | 6 (15) |
| **Levoflox** | 26 (78) | 7 (21) | Amoxicillin Clav | 14 (35) | 26 (65) |

Continued.

| Antibiotic for gram negative organisms | Sensitive (%) | Resistance (%) | Antibiotic for gram positive organisms | Sensitive (%) | Resistance ( %) |
|---|---|---|---|---|---|
| **Fluoroquinolones** | 14 (42) | 17 (51) | Tigecycline | 32 (80) | 8 (20) |
| **Minocycline** | 28 (84) | 5 (15) | | | |
| **Imipenam** | 14 (42) | 19 (57) | | | |
| **Colistin** | 30 (90) | 2 (6) | | | |

**Table 5: Drug sensitivity of individual organisms.**

| Antibiotic | *Staph aureus* (n=21) | | Enterococcus (n=14) | |
|---|---|---|---|---|
| | Sensitive (%) | Resistance (%) | Sensitive (%) | Resistance (%) |
| **Cephalosporin** | 14 (66.6) | 7 (33.3) | 4 (28.5) | 10 (71.4) |
| **Vancomycin** | 15 (71.4) | 6 (28.5) | 9 (64.2) | 5 (35.7) |
| **Teicoplanin** | 14 (66.6) | 1 (4.7) | 9 (64.2) | 5 (35.7) |
| **Linezolid** | 19 (90.4) | 2 (9.5) | 13 (92.8) | 1 (7.14) |
| **Amoxicillin-clav** | 6 (28) | 15 (71.4) | 4 (28.5) | 10 (71.4) |
| **Teigecycline** | 15 (71.4) | 6 (28.5) | 12 (85.7) | 2 (14) |
| **Levofloxacin** | 17 (80.9) | 4 (19) | 12 (85.7) | 2 (14) |

**Table 6: Drug sensitivity of individual organisms.**

| Antibiotic | Acinectobacter (n=18) | | Kleibsella (n=4) | |
|---|---|---|---|---|
| | Sensitive (%) | Resistance (%) | Sensitive (%) | Resistance (%) |
| **Meropenem** | 6 (33) | 12 (66.6) | 1 (25) | 3 (75) |
| **Piperacillin-tazobactam** | 3 (16) | 15 (83.3) | 2 (50) | 2 (50) |
| **Amikacin** | 4 (22.2) | 14 (77.7) | 3 (75) | 1 (25) |
| **Levoflox** | 16 (88.8) | 2 (11) | 2 (50) | 2 (50) |
| **FQ** | 7 (38.8) | 11 (61) | 1 (25) | 3 (75) |
| **Minocycline** | 16 (88.8) | 2 (11) | 4 (100) | 0 |
| **Colistin** | 18 (100) | 0 | 3 (75) | 1 (25) |
| **Cefepime** | 3 (16.6) | 10 (55) | 2 (50) | 2 (50) |
| **Imipenam** | 7 (38.8) | 11 (61) | 2 (50) | 2 (50) |

**Table 7: Clinical outcome.**

| Clinical outcome | N (%) |
|---|---|
| **Discharge** | 53 (72.6) |
| **DAMA** | 8 (10.9) |
| **Expired** | 12 (16.4) |

## DISCUSSION

The gold standard test for diagnosing newborn sepsis is still blood culture and the sensitivity pattern can be used to direct treatment. The current investigation was carried out retrospectively in newborn sepsis with blood culture positive results in order to determine the bacteriological profile and antibiogram. Culture positivity, according to our survey, was 7%.

Gram positive sepsis was more common than gram negative sepsis, however early onsent sepsis was more common. Staph aureus, acinetobacter, enterococci and enterobacter were frequently isolated species. The outcomes were similar to those of other studies and texts.[7-9] In the study undertaken, 50.6% are males and 49.3% are females. The male infants in a study by Khatua et al, constituted 70.7%.[13] Gupta and Sharma et al, reported a male predominance of 64.7 and 74% respectively.[10] The factors regulating the synthesis of gamma globulins are probably situated on the X chromosome. Presence of one X chromosome in the male infant thus confers less immunological protection compared to the female counterpart.[11,12]

In our study, neonatal septicemia was observed more in low-birth-weight neonates, especially lesser than 2 kg. Khatua et al, also reported a higher incidence of septicemia in low-birth-weight infants.[13] Higher incidence in low-birth-weight infants was also observed by other workers.[13] EOS was more common than LOS in our study. Many other studies from developing nations

have also shown similar results.[19-21] High rate of early onset sepsis would be due to early horizontal transmission from delivery rooms and NICU.[22-24] Vertical transmission from maternal genital tract colonized with pathogens due to unhygienic personal and obstetric practices also contribute.[25,26]

In the present study gram positive organism were isolated in 57.3, followed by 45.2%-gram negative sepsis. Gram positive organisms were more commonly isolated in neonates (ratio 40:28) in a study in north india by Agrawal et al, Variable results are seen in different areas regarding gram positive organisms versus gram negative organisms.[21,27] Frequency of organisms isolated from our study in decreasing order Staph aureus, Acinectobacter, Enterococci and Enterobacter. In a study from Nepal by Shrestha P et al, also, Staph aureus was the commonest organism.[28] Most other studies from India show common organisms to be Acinectobacter, Kleibsella and Staph aureus.[19-21,29]

Further analysis to see if organisms causing EOS were different from that causing LOS revealed that in our study staph aureus, enterococcus and acinectobacter were responsible for EOS and Acinetobacter, Kleibsella and Enterobacter for LOS. Staph aureus as commonest organism for EOS has been reported in study done in Nepal by Shreshta et al.[28] Most other studies have found that gram negative organisms predominatly cause EOS.[19,21] A study by Agrawal et al, in a large cohort from 3 tertiary care hospitals in Delhi (India) found no difference in profile for EOS and LOS.[27] This suggests the importance of deciding flora based on individual hospital site studies.

In our study, among gram negative organisms, Acinetobacter was 24.6%, Kleibsella (5.4%) and Escherichia coli (4.1%). In a study Agrawal et al, 64% isolates were gram-negative, frequency being Acinetobacter (22%), Klebsiella (17%) and *Escherichia coli* (14%).[15,27] Now a days there is increasing incidence of Acinetobacter spp infections in tertiary neonatal units of India.[15] Among gram positive organisms, most common organism was Staphylococcus aureus in 28.7%, followed by Enterococcus (19.1%) and CONS was seen in 4.1 % cases in our study. Another study conducted in a tertiary care centre of western India also found that Staphylococcus aureus (61%) was the predominant among gram-positive pathogens.[17]

In our study Staph aureus showed high resistance to amoxyclav and cephalosporins and sensitivity was higher to linezolid, levoflox, vancomycin, tigecycline. Most studies from north India and western India have shown that all Staph aureus (100%) were sensitive to linezolid, tigecycline, teicoplanin and vancomycin.[27,29]

In our study Acinectobacter was resistant to piptaz (69%), amikacin (66%) and carbapenams (51%) and was sensitive to colistin (100%), minocycline (84%) and levoflox (78%). In study by Agrawal et al, Acinectobacter species was resistant to carbapenem in 78% and MDR in 82% cases.[27] In Shrestha P et al, acinectobacter was sensitive to 1st generation antibiotic (3rd generation cephalosporins and gentamycin) only in 5.8% cases.[28] In study by Shah et al, Acinetobacter showed 100% resistance to piptaz/ cefoperazone sulbactum and aminoglycoside and sensitivity was good for colistin and tigecycline.[29] Sensitivity to cephalosporin and aminoglycosides and piptaz is low for Acinetobacter species in overall studies.

In our study, enterococci showed resistance to vancomycin in 35.7% and 85.7% sensitivity to tigecycline. Similar results were seen in Agrawal et al, where Enterococci showed resistance to vancomycin in 27 %.[27] Sensitivity of tigecycline 80% cases in study by Shah et al.[29] VRE are emerging in India. The mortality in neonatal sepsis in advanced intensive care era is secondary to emergence of multi drug resistance bugs. Early isolation of organism and antibiogram guided treatment can help in survival of the septic neonates.

Our study revealed different patterns in antibiotic sensitivity, resistance and trends as compared to other regions of India. Similarly, various studies are reporting the changing trends as per the area, the organism and the antibiotics used.[7,8] Traditional infection control practices aimed at decreasing hospital-acquired infections need to be extended to all health care facilities because health care-associated infections including bacteremia occur in diverse settings and not only during inpatient stays. Also, the importance of early diagnosis and intervention with effective antimicrobial therapy will likely be crucial to achieve further decreases in mortality associated with bloodstream infection.

As study was carried out for a short period, further studies will be required to know about emerging drug resistance and antibiotic sensitivity of organisms.

## CONCLUSION

Multi drug resistant organisms are emerging in modern neonatal care practice. Practice of antibiotic stewardship may save the babies from multidrug resistance organisms in future.

As the cultures showed variable antibiogram with complicated patterns of resistance, culture and sensitivity test should be performed in all cases of septicemia. The pattern of antibiotic sensitivity has changed and differs in various regions. Major antibiotics that were once sensitive are starting to show signs of resistance. Gram negative bacteria that are resistant to carbapenem may be treated with antibiotics such as colistin, levofloxacin and minocycline. Adhering to stringent antibiotic stewardship practices could protect infants from future exposure to multidrug-resistant organisms. Since tigecycline and colistin appear to be the new focus medications, great

care is taken to avoid resistance to the drugs. These could assist the doctor in responding quickly while taking evolving patterns into consideration.

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