Section: Pediatrics

Original Research Article

BACTERIOLOGICAL PROFILE AND ANTIBIOTIC SENSITIVITY PATTERN OF NEONATAL SEPSIS IN A TERTIARY CARE NEONATAL INTENSIVE CARE UNIT: A RETROSPECTIVE STUDY

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ABSTRACT

Background: Neonatal sepsis remains a major cause of morbidity and mortality in neonatal intensive care units (NICUs) worldwide. Timely identification of causative organisms and their antimicrobial susceptibility is essential to guide appropriate treatment. The objective is to identify the predominant microbial isolates and their antibiotic sensitivity patterns among neonates admitted to the NICU of a tertiary care hospital.

Materials and Methods: This retrospective observational study was conducted in the NICU of Lata Mangeshkar Hospital, Nagpur, over a 12-month period from January to December 2025. All neonates with culture-confirmed bloodstream infection were included. Organism identification and antibiotic-susceptibility data were extracted from microbiology records and analyzed descriptively.

Results: Sixty-two culture-positive blood samples were analyzed. The predominant isolates were methicillin-resistant Staphylococcus aureus (MRSA, 25.8%), Klebsiella spp. (14.5%), and coagulase-negative staphylococci (CoNS, 12.9%), followed by Escherichia coli, non-fermenting Gram-negative bacilli, Enterococcus spp., and Pseudomonas aeruginosa. High resistance was observed to ampicillin (30%) and third-generation cephalosporins (48%). Vancomycin and linezolid retained complete sensitivity (100%) against Gram-positive isolates, while amikacin (82%), meropenem (75%), and piperacillin-tazobactam (70%) remained effective against Gram-negative isolates. Multidrug resistance (MDR) was noted in 75% of Klebsiella spp., 60% of E. coli, and 40% of MRSA isolates.

Conclusion: MRSA and Klebsiella spp. were the predominant NICU pathogens, with substantial multidrug resistance among Gram-negative organisms. These findings emphasize the need for continuous microbiological surveillance and robust antibiotic stewardship to guide empirical therapy in NICU settings.

Keywords: Neonatal sepsis; Blood culture; Antibiotic resistance; Multidrug resistance; Neonatal intensive care unit; MRSA.

INTRODUCTION

Neonatal sepsis remains a significant cause of morbidity and mortality worldwide, particularly in low- and middle-income countries. In India, neonatal infections account for up to 30–40% of all neonatal deaths, with bloodstream infections being a major contributor.[1]. The clinical presentation of sepsis in neonates is often non-specific, and delays in appropriate treatment can result in rapid clinical deterioration.[2].

One of the cornerstone investigations in diagnosing neonatal sepsis is blood culture, which allows identification of the causative organism and guides targeted antimicrobial therapy. The pattern of microorganisms responsible for neonatal sepsis and their antimicrobial susceptibility profiles vary significantly across regions and over time, influenced by factors such as hospital infection-control practices, antibiotic usage patterns, and patient demographics.

In neonatal intensive care units (NICUs), infections may be classified as early-onset sepsis (EOS), occurring within the first 72 hours of life, or late-onset sepsis (LOS), developing thereafter.[3]. EOS is frequently caused by vertical transmission from the mother, while LOS is commonly associated with nosocomial or community-acquired pathogens. Gram-positive organisms such as Staphylococcus aureus.[4], coagulase-negative staphylococci (CoNS), and Enterococcus spp., as well as Gram-negative bacteria such as Klebsiella spp., E. coli, and Pseudomonas spp., are commonly implicated in LOS, especially in NICU settings.

Of growing concern is the emergence of multidrug-resistant (MDR) organisms, particularly extended-spectrum beta-lactamase (ESBL)-producing Klebsiella spp. and E. coli, and methicillin-resistant Staphylococcus aureus (MRSA)5. These resistant strains limit therapeutic options and increase hospital stay, healthcare costs, and neonatal mortality. Periodic surveillance of the microbial profile and antibiotic resistance pattern is therefore essential to guide effective empirical antibiotic therapy and to develop robust antibiotic stewardship policies.

While several multicentric and tertiary-care-based studies have addressed neonatal sepsis, localized data from individual NICUs remain critical for understanding hospital-specific trends. This is particularly important for tailoring empirical antibiotic policies, especially in high-risk populations such as preterm and low-birth-weight neonates.

Objectives of the Study

The present study aims to:

- Identify the spectrum of bacterial pathogens isolated from blood cultures in neonates admitted to the NICU.
- Evaluate their antimicrobial susceptibility patterns.
- Analyze monthly trends in isolate distribution over a six-month period.
- Inform empirical antibiotic policy and reinforce infection-control strategies within the NICU.

MATERIALS AND METHODS

Study Design and Setting: This was a retrospective observational study conducted in the Neonatal Intensive Care Unit (NICU) of Lata Mangeshkar Hospital, a tertiary care teaching hospital affiliated with NKP Salve Institute of Medical Sciences & Research Centre, Nagpur, India. The NICU provides level II and III care for both inborn and outborn neonates.

Study Period: The study was conducted over a twelve -month period, from January 1 to 30th December, 2025.

Study Population: All neonates (aged 0–28 days) admitted to the NICU with a culture-confirmed bloodstream infection during the study period were included. Neonates with incomplete records or isolates considered probable contaminants (e.g., a single CoNS isolate without accompanying clinical signs of sepsis) were excluded from the final analysis.

Data Collection: Relevant data were extracted from NICU case records and the microbiology laboratory register. For each neonate, the following details were recorded:

- Date of admission and culture report.
- Microorganism isolated from blood culture.
- Antibiotic susceptibility pattern (Kirby–Bauer disc diffusion or automated system, interpreted as per CLSI guidelines).
- Resistance or sensitivity to individual antibiotics.
- No patient identifiers were used, and all data were anonymized prior to analysis.

Microbiological Processing: Blood samples for culture were collected aseptically prior to initiation of antibiotic therapy and processed using standard microbiological techniques. Organism identification was performed by colony morphology, Gram staining, and biochemical testing. Antibiotic susceptibility testing was performed using the Kirby–Bauer disc diffusion method and interpreted according to Clinical and Laboratory Standards Institute (CLSI) 2024 guidelines.

Antibiotics Tested: The antibiotic panel tested included the following classes:

- Beta-lactams: ampicillin, cefotaxime, ceftriaxone, cefoperazone
- Beta-lactamase inhibitor combinations: piperacillin–tazobactam
- Aminoglycosides: gentamicin, amikacin, tobramycin
- Carbapenems: meropenem, imipenem
- Fluoroquinolones: ciprofloxacin
- Glycopeptides: vancomycin
- Oxazolidinones: linezolid
- Others: clindamycin, cotrimoxazole, doxycycline, fosfomycin, tigecycline

Statistical Analysis: Descriptive statistics (frequencies and percentages) were used to summarize organism distribution and antibiotic-susceptibility profiles. Data were entered and tabulated in Microsoft Excel.

RESULTS

A total of 62 culture-positive blood samples were analyzed from neonates admitted to the NICU between January and December 2025. The distribution of organisms and their antibiotic-sensitivity profiles are summarized below.

Organism-Wise Distribution of Isolates

Table 1: Organism-wise distribution of blood-culture isolates (N = 62)

Organism	Number of Isolates	Percentage (%)
Methicillin-resistant Staphylococcus aureus (MRSA)	16	25.8
Klebsiella spp.	9	14.5
Coagulase-negative staphylococci (CoNS)	8	12.9
Escherichia coli	5	8.1
Non-fermenting Gram-negative bacilli	5	8.1
Enterococcus spp.	3	4.8
Pseudomonas aeruginosa	2	3.2
Others / mixed flora	14	22.6
Total	62	100.0

MRSA was the most frequently isolated pathogen, followed by Klebsiella spp. and CoNS. The presence of non-fermenters and Pseudomonas aeruginosa suggests a notable proportion of hospital-acquired infections.

Month-Wise Distribution of Positive Cultures

Table 2: Month-wise distribution of culture-positive cases and predominant isolates

Month	Total Positive Cultures	Predominant Isolates
January	10	MRSA, Klebsiella spp., CoNS
February	8	E. coli, CoNS
March	9	CoNS, MRSA
April	11	Klebsiella spp., Enterococcus spp.
May	12	MRSA, non-fermenters
June	12	MRSA, Klebsiella spp., Pseudomonas
Total	62	—

A qualitative rise in Gram-negative isolates was noted in the latter half of the study period.

Antibiotic Sensitivity Patterns: Gram-Positive Isolates (MRSA, CoNS, Enterococcus spp.)

Table 3: Antibiotic sensitivity of Gram-positive isolates

Antibiotic	Sensitivity (%)
Vancomycin	100
Linezolid	100
Clindamycin	85
Gentamicin	78
Ciprofloxacin	60
Cotrimoxazole	55
Ampicillin	30

MRSA isolates showed complete sensitivity to vancomycin and linezolid, while resistance to ampicillin was substantial (sensitivity 30%).

Gram-Negative Isolates (Klebsiella spp., E. coli, Pseudomonas, Non-Fermenters)

Table 4: Antibiotic sensitivity of Gram-negative isolates

Antibiotic	Sensitivity (%)
Amikacin	82
Meropenem	75
Piperacillin–tazobactam	70
Gentamicin	65
Ciprofloxacin	55
Cefotaxime / Ceftriaxone	48
Colistin (selected isolates)	100

A notable proportion of Gram-negative isolates were resistant to third-generation cephalosporins, suggesting the presence of ESBL-producing strains. Carbapenems and aminoglycosides retained better efficacy.

Multidrug Resistance (MDR) Patterns

Table 5: Multidrug resistance rates by organism

Organism	MDR Rate (%)
Klebsiella spp.	75
Escherichia coli	60
MRSA	40

Definition: MDR was defined as resistance to ≥ 3 antibiotic classes. This underscores the substantial burden of resistance in the NICU setting, particularly in late-onset sepsis.

Empirical Treatment Implications: Based on the observed sensitivity trends, empirical coverage with piperacillin–tazobactam and amikacin, combined with vancomycin for suspected Gram-positive sepsis, appears rational in the local context. However, continued surveillance is essential to refine institutional protocols.

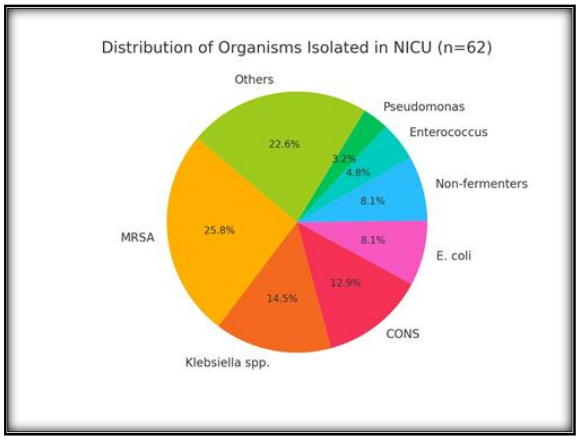

Figure 1: Distribution of organisms isolated in the NICU (N = 62).

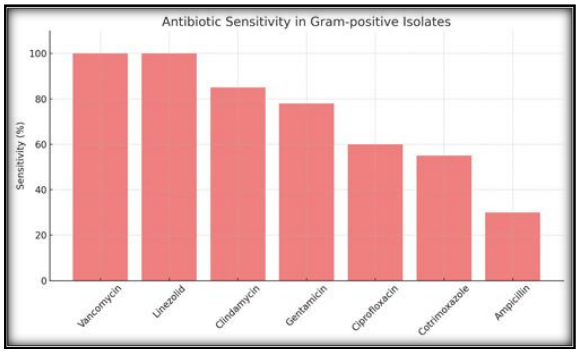

Figure 2: Antibiotic sensitivity pattern of Gram-positive isolates.

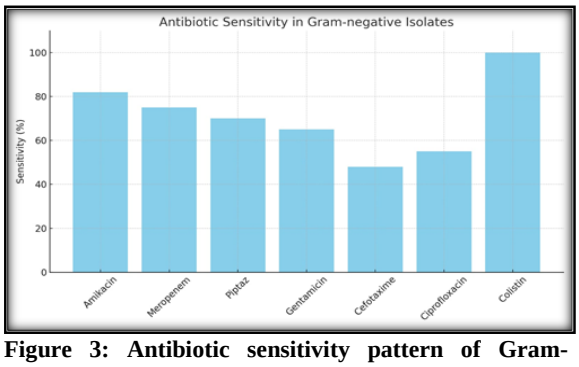

Figure 3: Antibiotic sensitivity pattern of Gram-negative isolates.

DISCUSSION

The predominance of MRSA and Klebsiella spp. in the present study is consistent with findings reported from several neonatal intensive care units (NICUs) across India, where multidrug-resistant Gram-positive and Gram-negative organisms remain major causes of neonatal morbidity and mortality. The predominance of these organisms may reflect the complex interaction between prolonged hospitalization, frequent exposure to broad-spectrum antibiotics, invasive procedures, and the vulnerability of neonates to healthcare-associated infections. Roy et al,[6] in a tertiary care NICU in northern India, identified Klebsiella pneumoniae as the most common Gram-negative pathogen and reported high levels of resistance to commonly used cephalosporins and aminoglycosides. These observations are particularly important because third-generation cephalosporins and aminoglycosides have traditionally constituted an important component of empirical treatment for neonatal sepsis. Increasing resistance among Klebsiella isolates therefore substantially limits the effectiveness of these agents and may result in delayed initiation of appropriate antimicrobial therapy.

Similarly, Gupta et al,[7] reported MRSA as one of the leading causes of late-onset neonatal sepsis, with considerable resistance to penicillins and cephalosporins. The emergence and persistence of MRSA in NICUs is of particular concern because neonates frequently require prolonged vascular access, respiratory support, and other invasive interventions that may facilitate transmission of resistant organisms. Kumar et al,[5] likewise documented high rates of multidrug resistance among neonatal sepsis isolates in a tertiary care hospital in India. The similarity between these findings and those of the present study suggests that antimicrobial resistance in neonatal units is not an isolated institutional phenomenon but represents a broader and continuing challenge within Indian healthcare settings.

Among Gram-negative organisms, Enterobacteriaceae, particularly E. coli and Klebsiella spp., continue to represent important causes of neonatal sepsis. Shah et al,[8] reported a substantial contribution of ESBL-producing Enterobacteriaceae among neonatal sepsis isolates. ESBL production is clinically significant because it confers resistance to several commonly used beta-lactam antibiotics, particularly third-generation cephalosporins. Consequently, infections caused by ESBL-producing organisms may require the use of broader-spectrum agents, including carbapenems, especially in critically ill neonates. However, increasing reliance on carbapenems may itself exert selective pressure and contribute to the emergence of carbapenem-resistant organisms. Thus, although carbapenems may remain essential for selected severe infections, their use should ideally be guided by microbiological confirmation and institutional susceptibility patterns rather than indiscriminate empirical prescribing.

The presence of non-fermenting Gram-negative organisms and Pseudomonas spp. in the present study further highlights the potential contribution of healthcare-associated transmission. Non-fermenters are particularly important in NICU settings because they can survive in moist environmental reservoirs and may be associated with contaminated equipment,

intravenous fluids, respiratory devices, sinks, and other healthcare surfaces. Their occurrence may also reflect the extensive use of invasive devices and prolonged exposure to antibiotics. In addition, inadequate hand hygiene, improper environmental cleaning, overcrowding, and gaps in adherence to infection-control practices can facilitate transmission between healthcare workers, neonates, and the surrounding environment. Therefore, the identification of these organisms should prompt not only appropriate antimicrobial treatment but also a review of infection-prevention practices within the unit.

The predominance of resistant organisms observed in the present study has important implications for empirical antimicrobial therapy. In neonatal sepsis, treatment decisions frequently need to be made before culture and susceptibility results become available, making the choice of empirical antibiotics particularly challenging. While broad-spectrum therapy may be necessary in critically ill neonates, prolonged or inappropriate use of broad-spectrum antibiotics can further promote antimicrobial resistance. Conversely, inadequate initial antimicrobial coverage may result in treatment failure and increased mortality. This therapeutic dilemma emphasizes the importance of developing institution-specific empirical treatment protocols based on the local microbial profile and regularly updated antibiogram data.

The present findings therefore support the need for strict antimicrobial stewardship within the NICU. Antibiotics should be selected according to the most likely pathogens, local resistance patterns, clinical severity, and subsequently available microbiological results. Wherever possible, antimicrobial therapy should be narrowed or de-escalated once culture and sensitivity results become available. Periodic review of antibiotic consumption and resistance patterns can help identify inappropriate prescribing practices and guide modifications in empirical treatment protocols. Such an approach can help maintain the effectiveness of currently available antibiotics while reducing unnecessary exposure to broad-spectrum agents.

The findings also support the continued role of agents such as vancomycin and linezolid for selected resistant Gram-positive infections, particularly when MRSA is suspected or confirmed. However, these agents should not be used routinely without microbiological or clinical justification because unnecessary exposure may promote further resistance and increase treatment-related concerns. Similarly, carbapenems may remain important for severe infections caused by ESBL-producing Gram-negative organisms, but their use should be carefully restricted to situations where their spectrum is justified. Nagata et al,[9] in a Japanese NICU setting, similarly emphasized the importance of appropriate antimicrobial selection in the management of resistant neonatal infections. Differences in microbial ecology between countries and individual hospitals, however, highlight why treatment recommendations from other settings should be adapted cautiously to local epidemiological conditions.

Thapan et al,[10] also highlighted the dynamic nature of antimicrobial resistance in neonatal units and emphasized the importance of regular surveillance of resistance patterns. This is particularly relevant because the microbial profile of a NICU may change over time in response to antibiotic prescribing practices, infection-control interventions, patient turnover, and local outbreaks. An antibiogram prepared annually or at regular intervals can provide clinicians with practical information regarding the susceptibility of common organisms and assist in the development of rational empirical treatment guidelines. In addition, continuous surveillance may help identify early increases in resistance that could otherwise remain unnoticed until treatment failures become frequent.

Infection prevention remains equally important because antimicrobial therapy alone cannot control the spread of resistant organisms. Strict hand-hygiene compliance before and after every patient contact, appropriate use of personal protective equipment, regular cleaning and disinfection of NICU surfaces and equipment, safe handling of intravenous lines, and proper catheter-care practices should form the foundation of infection-control measures. Particular attention should be given to environmental reservoirs when non-fermenters or Pseudomonas spp. are repeatedly isolated. Regular surveillance cultures may be considered during suspected outbreaks, accompanied by prompt investigation of common sources and reinforcement of infection-control measures.

Overall, the predominance of MRSA and Klebsiella spp., together with the presence of ESBL-producing Enterobacteriaceae and non-fermenting Gram-negative organisms, indicates a substantial antimicrobial resistance burden in the studied NICU. These findings are broadly comparable with reports from other Indian centres,[5–8] and reinforce the need for a coordinated strategy combining early microbiological diagnosis, rational empirical antimicrobial therapy, antimicrobial stewardship, and rigorous infection-control practices. Rather than relying on a fixed antibiotic regimen, empirical treatment protocols should remain dynamic and should be periodically revised according to the prevailing local antibiogram. Such an approach can improve the likelihood of appropriate initial therapy while minimizing unnecessary antibiotic exposure.

In conclusion, the resistance pattern observed in the present study emphasizes that neonatal sepsis management should move beyond antibiotic selection alone toward an integrated infection-control and antimicrobial-stewardship framework. Regular monitoring of causative organisms and their susceptibility profiles, timely review of antibiotic policies, strict hand hygiene, appropriate device care, environmental surveillance, and prompt identification of outbreaks are essential components of this strategy. Continued surveillance and periodic

updating of institutional antibiograms, as recommended by Thapan et al,[10] may help clinicians respond more effectively to changing resistance patterns. Such measures are particularly important in resource-limited settings, where treatment options for multidrug-resistant infections may be restricted and the consequences of delayed appropriate therapy can be severe.

Limitations: This study has several limitations. It was a single-center, retrospective analysis with a relatively modest sample size (n = 62), which may limit generalizability. Clinical correlates such as gestational age, birth weight, and mortality outcomes were not analyzed. Genotypic confirmation of resistance mechanisms (e.g., ESBL, mecA gene) was not performed; susceptibility was inferred phenotypically. Statistical trend analysis was not performed for monthly variation in isolate distribution, so the observed changes should be interpreted as descriptive rather than statistically significant. Prospective, multicentric studies with longer follow-up and clinical-outcome data are warranted to validate and extend these findings.

CONCLUSION

The present study highlights the evolving microbiological landscape of neonatal sepsis in a tertiary care NICU setting. The predominance of MRSA and Klebsiella spp., together with emerging resistance among both Gram-positive and Gram-negative pathogens, reflects the increasing burden of multidrug-resistant infections in neonates. The high resistance rates to commonly used antibiotics such as ampicillin and third-generation cephalosporins underscore the need for an urgent review of empirical treatment protocols.

Our findings support the continued utility of vancomycin, linezolid, and piperacillin–tazobactam in treating resistant infections, though their use should be guided by ongoing local antibiogram data. The isolation of non-fermenters and Pseudomonas spp. also suggests possible nosocomial transmission, potentially linked to invasive procedures and infection-control lapses.

This study underlines the critical importance of regular microbiological surveillance, strict adherence to infection-prevention practices, and implementation of antibiotic stewardship programs in neonatal units. Timely pathogen identification and rational antibiotic use guided by susceptibility data remain cornerstone strategies to improve neonatal outcomes and curb the spread of antimicrobial resistance.

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