ORIGINAL ARTICLE

Treatment Outcomes, Antibiotic Use, and Resistance Patterns among Neonatal Sepsis Patients: A Prospective Observational Study in Eastern India

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ABSTRACT

Introduction: Neonatal sepsis is a leading cause of neonatal mortality in low- and middle-income countries. The emergence of multidrug-resistant (MDR) pathogens poses a major challenge. This study determined treatment outcomes, antibiotic usage, and resistance patterns in a tertiary NICU in Eastern India. **Methods**: A 12-month prospective observational study recruited neonates with clinical and/or laboratory-confirmed sepsis. Baseline demographics, clinical features, antibiotic regimens, and outcomes were recorded. Blood cultures and antibiotic susceptibility tests were performed following standard guidelines. Data were analyzed to identify factors associated with mortality. **Results**: Among 320 enrolled neonates, 180 (56%) had positive blood cultures. Gram-negative bacteria (60%) predominated, led by *Klebsiella pneumoniae* and *Escherichia coli*, while *Staphylococcus aureus* was the most common Gram-positive. Approximately 45% of isolates were MDR, with 53% of *Klebsiella* and 40% of *S. aureus* resistant. Seventy percent of neonates required escalation to higher-end antibiotics, and overall mortality was 18%. MDR infection, low birth weight, and outborn status were significantly associated with increased mortality. **Conclusion**: Neonatal sepsis in Eastern India is characterized by high MDR rates and substantial antibiotic escalation. Effective infection control measures and rigorous antimicrobial stewardship are urgently needed to reduce morbidity, mortality, and resistance. Regular surveillance of local antibiograms and judicious antibiotic use can improve clinical outcomes and inform policy. These findings emphasize a pressing need for coordinated efforts to strengthen diagnostic capacities, track antimicrobial resistance patterns, and ensure appropriate resource allocation.

KEY WORDS: Neonatal sepsis, Multidrug resistance (MDR), Antibiotic resistance, Treatment outcomes, Eastern India.

Introduction

Neonatal sepsis remains a significant contributor to morbidity and mortality in the neonatal period, particularly in low- and middle-income countries.[1] The World Health Organization (WHO) estimates that neonatal sepsis accounts for approximately 15% of all neonatal deaths globally, with the highest burden in sub-Saharan Africa and South Asia.[2] In India, neonatal mortality rates remain considerably high, and sepsis is one of the leading causes of hospitalization and death among neonates.[3] Management of neonatal sepsis presents a major challenge, especially given the emergence of antimicrobial resistance (AMR) and the variation in resistance patterns across different geographic regions. Clear understanding of the local epidemiology, antibiotic use, and resistance profile is crucial for guiding empirical antibiotic selection and optimizing treatment outcomes.[4]

The development of antibiotic resistance has become a serious public health concern, with organisms such as multidrug-resistant (MDR) strains of *Klebsiella pneumoniae, Escherichia coli*, and *Staphylococcus aureus* frequently implicated in neonatal sepsis.[5]

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In many low- and middle-income countries, the excessive or inappropriate use of antibiotics in hospital settings has contributed to the rapid emergence of resistant bacterial strains. This is particularly evident in neonatal intensive care units (NICUs), where vulnerable neonates require immediate and often broad-spectrum antibiotic therapy to mitigate the high risk of mortality.[6]

Despite the known challenges posed by antibiotic resistance, few large-scale studies have documented and analyzed trends in antibiotic use and their associated resistance profiles in the eastern region of India.[7] Most existing data is derived from single-center or smaller studies with limited sample sizes, making it challenging to draw comprehensive conclusions or develop universally applicable management guidelines. The lack of uniform protocols for diagnosing and treating neonatal sepsis in India further complicates the scenario, leading to heterogeneity in prescribing practices and clinical outcomes.[7]

Eastern India presents unique healthcare challenges due to socioeconomic disparities, varying healthcare infrastructure, and differences in antibiotic prescribing patterns compared to other regions. However, comprehensive data on the burden of neonatal sepsis, antibiotic usage trends, and resistance patterns remain scarce, hindering effective clinical decision-making and antimicrobial stewardship efforts.

Given these pressing concerns, the present study aimed to investigate the clinical spectrum, treatment outcomes, and antibiotic resistance patterns among neonates diagnosed with sepsis in a tertiary care center in Eastern India. The objectives were: (1) to identify the common pathogens responsible for neonatal sepsis, (2) to evaluate patterns of antibiotic usage in the neonatal unit, (3) to analyze resistance patterns among the commonly isolated pathogens, and (4) to examine the relationship between antimicrobial therapy and treatment outcomes. By generating evidence on the local burden of antibiotic resistance and treatment effectiveness, this study seeks to inform clinical decision-making, develop targeted antimicrobial stewardship strategies, and ultimately reduce mortality and morbidity associated with neonatal sepsis in Eastern India.

Methods

Study Design and Setting

This prospective observational study was conducted in the neonatal intensive care unit (NICU) of a tertiary care teaching hospital in Eastern India. The study site provides specialized care for both inborn (delivered in the hospital) and outborn (referred from external facilities) neonates, servicing a large catchment area that includes rural, semi-urban, and urban populations. The NICU is equipped with advanced support systems such as mechanical ventilation, continuous positive airway pressure (CPAP), and availability of parenteral nutrition.

The study was performed over a period of 12 months after obtaining ethical clearance from the Institutional Ethics Committee. The study was approved by the Institutional Ethics Committee (Approval No.: IEC/2023/134, dated: 15th January 2023). Informed consent was secured from parents or legal guardians before the enrolment of neonates into the study.

Participant Selection

All consecutive neonates (0–28 days of age) admitted with clinical and/or laboratory-confirmed sepsis during the study period were screened for inclusion. Neonatal sepsis was defined based on the presence of one or more of the following criteria:

1. **Clinical features of sepsis** (e.g., temperature instability, tachypnea, apnea, feeding intolerance, lethargy, poor perfusion, etc.) with or without hemodynamic instability.
2. **Laboratory abnormalities** suggestive of sepsis, such as leukopenia (white blood cell count < 4000 cells/mm^3), leukocytosis ($> 20{,}000$ cells/mm^3), elevated C-reactive protein (CRP), and/or positive blood culture or other sterile site culture (e.g., cerebrospinal fluid).

Exclusion criteria included neonates with major congenital anomalies that precluded standard care or those who were discharged or transferred before a definitive sepsis evaluation could be completed.

Data Collection

Upon enrollment, data were collected using a standardized case report form. Demographics and Baseline Characteristics including gestational age (in weeks), birth weight, sex, mode of delivery,

location of birth (inborn vs. outborn), maternal risk factors (e.g., prolonged rupture of membranes, chorioamnionitis), and Apgar scores were obtained. Clinical Findings at Admission including vital signs (heart rate, respiratory rate, temperature), clinical signs of sepsis, requirement of respiratory support (oxygen, CPAP, or mechanical ventilation), and any existing comorbidities (e.g., perinatal asphyxia) were noted. Laboratory Investigations comprising of complete blood counts, CRP levels, blood culture, and sensitivity results. If indicated, other sterile-site cultures such as cerebrospinal fluid, urine culture, or endotracheal aspirate cultures were also obtained. Antibiotic Therapy Details were noted including type of empirical antibiotic chosen, duration of antibiotic therapy, changes in antibiotic regimens based on culture and sensitivity results, and total length of hospital stay. Treatment Outcomes including clinical improvement (resolution of sepsis symptoms), length of NICU stays, discharge status (discharged alive, referral to higher center, left against medical advice), and mortality were assessed.

Follow-up was continued until the neonate was discharged from the hospital or died during the hospital stay. The primary outcome variable was in-hospital mortality due to sepsis. Secondary outcomes included the need for escalation of antibiotic therapy (switch to broader-spectrum antibiotics due to clinical deterioration or resistance patterns), incidence of culture-proven multidrug-resistant organisms[8], and duration of NICU stay. Data were collected at baseline, at 48-hour intervals, and at discharge.

Microbiological Methods

Blood samples were collected aseptically before initiation of antibiotic therapy, if possible. Approximately 1–2 mL of blood was drawn aseptically for each culture bottle, in line with standard neonatal blood culture practices to ensure adequate yield. Culture bottles were incubated for a minimum of 48 hours and up to five days if no growth was detected. Positive cultures were further processed (using the BACTEC system) to identify the organism by standard biochemical methods. Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method[9] or automated systems in accordance with Clinical and Laboratory Standards Institute (CLSI) guidelines[10]. Results were reported as sensitive, intermediate, or resistant. Intermediate results were considered for escalation or de-escalation decisions by the clinical team. Multidrug resistance (MDR) was defined as resistance to at least one agent in three or more antibiotic classes.[8]

Statistical Analysis

Data were entered into a secure electronic database and analyzed using statistical software (SPSS Version 27.0, IBM Corp., Armonk, NY). Descriptive statistics were used to summarize demographic and clinical variables. Categorical variables were expressed as frequencies and percentages, whereas continuous variables were expressed as means ± standard deviations or medians with interquartile ranges, based on normality of distribution. Associations between potential risk factors and mortality were tested using chi-square (for categorical variables) and independent t-tests (for continuous variables). A p-value of <0.05 was considered statistically significant.

Results

A total of 380 neonates with suspected sepsis were screened for eligibility. Out of these, 320 neonates fulfilled the inclusion criteria and were enrolled in the study. The final analysis included all 320 participants, as there were no withdrawals following enrolment.

Baseline Characteristics

Table 1: Baseline Characteristics of Study Participants (N = 320)

Variable	Value
Gestational age (weeks), mean ± SD	35.2 ± 3.1
Birth weight (g), mean ± SD	2200 ± 480
Male, n (%)	182 (57)
Female, n (%)	138 (43)
Inborn, n (%)	185 (58)
Outborn, n (%)	135 (42)
Maternal PROM (>18 hrs), n (%)	80 (25)
Maternal chorioamnionitis, n (%)	32 (10)
Median Apgar at 1 min (IQR)	6 (5–7)
Median Apgar at 5 min (IQR)	8 (7–9)

Note: Values in parentheses represent percentages; IQR = Interquartile Range PROM: Prolonged rupture of membranes; CoNS: Coagulase-negative staphylococci; MDR: Multidrug-resistant.

The mean (± SD) gestational age was 35.2 ± 3.1 weeks, and the mean birth weight was 2200 ± 480 grams. Nearly 43% (n = 138) of the neonates were female, whereas 57% (n = 182) were male. Over half of the neonates (58%, n = 185) were inborn (those delivered within the study hospital),

while 42% (n = 135) were referred from external facilities/outborn neonates (those referred from other facilities). Maternal risk factors such as prolonged rupture of membranes (>18 hours) were present in 25% of cases, and about 10% (n = 32) of mothers had chorioamnionitis (Table 1).

Clinical Presentation and Laboratory Findings

Among the neonates, common presenting symptoms were poor feeding (68%), respiratory distress (60%), lethargy (55%), and temperature instability (52%). Around 32% required some form of respiratory support, including CPAP (n = 64) or mechanical ventilation (n = 38). Hypotension requiring inotropic support was noted in 12% of the neonates.

Elevated CRP (>10 mg/L) was observed in 82% of neonates. The mean (± SD) total leukocyte count was 13,500 ± 4,200 cells/mm^3. Approximately 18% of neonates had leukopenia (<4000 cells/mm^3), and 24% had leukocytosis (>20,000 cells/mm^3).

Microbiological Profile

Blood culture yielded growth in 180 (56%) out of 320 neonates. Gram-negative bacteria were isolated in 60% of the culture-positive cases (n = 108), whereas Gram-positive isolates accounted for 40% (n = 72).

Among Gram-negatives, *Klebsiella pneumoniae* was the most common organism (26%, n = 47), followed by *Escherichia coli* (22%, n = 40) and *Acinetobacter baumannii* (12%, n = 21). Among Gram-positive isolates, *Staphylococcus aureus* was most frequent (18%, n = 32) followed by coagulase-negative staphylococci (CoNS) (12%, n = 22) and *Enterococcus faecalis* (10%, n = 18).

A significant proportion (45%, n = 81) of the isolated organisms demonstrated multidrug resistance (MDR). MDR was most frequently observed in *Klebsiella pneumoniae* (53% of *Klebsiella* isolates), followed by *Acinetobacter baumannii* (48% of *Acinetobacter* isolates) and *Escherichia coli* (45% of *E. coli* isolates). Among Gram-positive organisms, 40% (n = 13) of *Staphylococcus aureus* isolates were methicillin-resistant (MRSA) (Table 2).

Table 2: Distribution of Microorganisms Isolated and their MDR Status (N = 180 culture-positive cases)

Organism	Frequency (n)	Percentage of total isolates (%)	MDR cases (n)	MDR (%)
Gram-negative organisms				
Klebsiella pneumoniae	47	26	25	53
Escherichia coli	40	22	18	45
Acinetobacter baumannii	21	12	10	48
Pseudomonas aeruginosa	12	7	5	42
Gram-positive organisms				
Staphylococcus aureus	32	18	13	40 (MRSA)
CoNS	22	12	6	27
Enterococcus faecalis	18	10	4	22
Total	192*	100	81	45

* Some neonates had more than one isolate (polymicrobial infections).

Antibiotic Use and Stewardship

All neonates were started empirically on antibiotics upon clinical suspicion of sepsis. The first-line empirical antibiotic regimen commonly included a combination of ampicillin and gentamicin for early-onset sepsis, and piperacillin-tazobactam plus amikacin for late-onset sepsis. In total, 70% (n = 224) of neonates required escalation of antibiotics based on clinical non-response or antibiotic susceptibility results. Common escalation regimens included carbapenems (meropenem or imipenem) and glycopeptides (vancomycin or teicoplanin) if Gram-positive resistant organisms were suspected or confirmed.

Among Gram-negative isolates, resistance to third-generation cephalosporins was noted in 62%, and resistance to carbapenems was observed in 35% of *Klebsiella* and 28% of *E. coli* isolates. Among MRSA isolates, resistance to clindamycin and erythromycin was observed in over 60% of cases.

The duration of antibiotic therapy ranged from a minimum of 5 days to a maximum of 21 days, with a median of 10 days (IQR: 7–14). A significant proportion of neonates (n = 78, 24%) received more than two different antibiotic regimens during their hospital stay, highlighting the complexity of treatment in the face of emerging resistance.

Treatment Outcomes

The overall mortality among neonates enrolled in the study was 18% (n = 58). Of the 58 deaths, 52 (90%) had culture-proven sepsis with resistant organisms involved in 30 (52%) of these fatal cases. Among the survivors, 28% (n = 74) experienced prolonged NICU stay (≥14 days). Persistent sepsis or complications such as necrotizing enterocolitis, intraventricular hemorrhage, and multi-organ dysfunction were the main reasons for prolonged NICU stays. Clinical improvement was noted in 72% (n = 231) of the total neonates, who were eventually discharged in stable condition. Ten neonates (3%) were referred to higher-level cardiac or surgical centers due to complications such as congenital heart disease or gastrointestinal anomalies (Table 3).

Table 3: Clinical Outcomes among Neonates with Sepsis (N = 320)

Outcome	Frequency (n)	Percentage (%)
Survived (discharged in stable condition)	231	72
Prolonged NICU stay (≥14 days)	74	28
Referred to higher center	10	3
Mortality	58	18
Among total deaths (n = 58): Culture-positive cases: 52 (90%) Among total deaths (n = 58): MDR organism implicated: 30 (52%)		

Risk Factors for Mortality

Univariate analysis showed that gestational age <34 weeks (p = 0.02), birth weight <2000 g (p = 0.01), outborn status (p = 0.03), presence of MDR infection (p = 0.004), and need for mechanical ventilation (p <0.001) were significantly associated with mortality. Multivariate logistic regression confirmed that birth weight <2000 g (OR 2.12, 95% CI: 1.12–3.71, p = 0.01) and MDR infection (OR 2.64, 95% CI: 1.46–4.12, p <0.001) remained significant predictors of neonatal mortality.

Discussion

In this prospective observational study in Eastern India, we investigated treatment outcomes, antibiotic use, and resistance patterns among neonates with sepsis. We found that a little over half (56%) of the neonates had culture-proven sepsis, and 45% of these isolates were multidrug resistant. The overall mortality rate was 18%, with MDR organisms significantly contributing to poor outcomes. Escalation of antibiotics was frequently required, emphasizing the complexity of managing neonatal sepsis in a high-burden, resource-limited setting.

Our findings mirror those reported in other studies across India and other low- and middle-income settings, although the specific organisms and resistance rates vary with geographic location and hospital ecology. Our culture positivity rate of 56% is comparable to various studies in India, which shows variable positivity rate ranging from 20% to 60%.[11–14] Studies in developing countries often face lower culture yield due to prior antibiotic exposure, inadequate blood sample volume, and differences in laboratory capacity. Our relatively high culture-positivity might be attributed to careful sampling protocols and use of automated blood culture systems.

Similar to reports from other South Asian regions, we observed that Gram-negative bacilli, particularly *Klebsiella pneumoniae* and *Escherichia coli*, were the most frequently isolated organisms. Several Indian studies underscore *Klebsiella pneumoniae* as the leading cause of neonatal sepsis, with prevalence rates ranging from 20% to 40% of culture-positive sepsis.[12] *E. coli* continues to be an equally important pathogen in many NICUs, as reported in studies from northern and southern parts of India.[15] The risk factors for Gram-negative sepsis often include inadequate infection control measures, overcrowding, and cross-transmission in NICUs.[16] The high resistance to cephalosporins and aminoglycosides necessitated the frequent use of carbapenems, indicating a narrowing spectrum of effective empirical antibiotics in this population.

The finding that 45% of all isolates were multidrug-resistant underscores the escalating crisis of antimicrobial resistance (AMR). Previous research from tertiary care centers in India and neighboring countries report MDR rates varying between 30% and 70% for Gram-negative pathogens.[17,18] Our data specifically demonstrate high rates of MDR among *Klebsiella pneumoniae* (53%) and *Acinetobacter baumannii* (48%), aligning with national and regional data. The presence of methicillin-resistant *Staphylococcus aureus* (MRSA) in 40% of *S. aureus* isolates is concerning, but it falls within the reported range of 30%–50% in various Indian studies. The reasons for high MDR burden include overuse or misuse of broad-spectrum antibiotics, lack of strict infection

control policies, and inadequate surveillance of antibiotic resistance.

The majority of neonates in our study started on empirical regimens typically recommended in the Indian context (ampicillin + gentamicin for early-onset sepsis, piperacillin-tazobactam + amikacin for late-onset sepsis). However, 70% required escalation to higher-end antibiotics such as carbapenems and glycopeptides. This high escalation rate is consistent with literature from NICUs in regions with high AMR burden, where empirical therapies are frequently rendered ineffective. Comparable studies from other parts of India show that 50%–80% of neonates receive multiple antibiotic regimens during a single hospitalization for sepsis.[19,20]

Our overall mortality rate of 18% is consistent with multiple Indian studies reporting neonatal sepsis-related mortality between 15% and 25%. The present study features that low birth weight, prematurity, and MDR infection significantly increase the risk of mortality, findings that are consistent with the existing literature.[16] Mechanically ventilated neonates were at greater risk of death, reflecting the severity of illness in this subgroup. Similar risk factors have been documented in NICUs worldwide, particularly in resource-limited settings.[21,22]

In our study, Acinetobacter baumannii accounted for 12% of the Gram-negative isolates. This organism is increasingly recognized as a formidable nosocomial pathogen in neonatal units. Several Indian studies have reported an upsurge in Acinetobacter infections over the past decade, often associated with outbreaks and high mortality.[23,24] The organism's ability to survive on hospital surfaces and acquire multiple resistance genes has grave implications for infection control.

Strengths of our study include a relatively large sample size, prospective design, and comprehensive microbiological investigations, which enabled detailed characterization of pathogens and their susceptibility profiles. The high culture positivity rate of 56% is notable, particularly given the challenges in low-resource settings. This study highlights how MDR pathogens directly contribute to mortality, reinforcing the growing global concern over antimicrobial resistance (AMR). The use of standardized clinical definitions and consistent follow-up protocols minimized selection and information biases. Our findings provide an updated snapshot of the AMR landscape in a region of Eastern India, which remains underrepresented in neonatal sepsis literature.

Limitations include the single-center design, which may affect the generalizability of the results to other regions with different hospital infection control practices and antibiotic policies. Though we attempted to document prior antibiotic exposure, it was often challenging to accurately ascertain the antibiotic history for outborn neonates. Additionally, we did not perform genotyping or advanced molecular characterization to delineate specific resistance mechanisms or strain-relatedness in case of outbreaks. Furthermore, the long study duration (12 months) could introduce time-varying factors; however, we attempted to control for seasonal variation in the analysis. Our study lacks advanced molecular techniques (e.g., genotyping or sequencing) that could help determine the exact mechanisms of resistance and the relatedness of strains

The high burden of resistant organisms stresses an urgent need for robust antibiotic stewardship and infection control programs. The frequent requirement for carbapenems and vancomycin suggests that current empirical regimens need to be regularly re-evaluated based on local antibiograms to optimize antimicrobial therapy and reduce the emergence of further resistance. Enhanced infection control measures, including hand hygiene, strict contact isolation for infected neonates, and routine surveillance cultures in high-risk units, can help minimize nosocomial transmission.[25]

Notably, no formal antibiotic stewardship interventions were implemented during the study period. Antibiotic use was guided by clinical protocols and local antibiogram data; however, structured stewardship programs and outcome audits were lacking, which is acknowledged as a limitation. Policy-level interventions must prioritize the enforcement of antibiotic-prescribing guidelines, implementation of antimicrobial stewardship committees, and improved laboratory capacity for early detection of resistant pathogens. As recommended by WHO and the Indian Council of Medical Research, antibiotic stewardship programs should incorporate continuous training of healthcare professionals, rational antibiotic use, and timely de-escalation of therapy guided by culture and sensitivity results.[26,27] Further, better coordination among tertiary care hospitals and peripheral health centers is warranted

to reduce inappropriate antibiotic usage before referral.

Future multicenter studies are needed to provide a more representative picture of the neonatal sepsis landscape in different regions of India. Investigations into the molecular epidemiology of MDR organisms can elucidate transmission dynamics and resistance mechanisms, facilitating targeted infection control strategies. Additionally, studies on newer antibiotics (e.g., ceftazidime-avibactam) and adjunct therapies (e.g., immunoglobulins, lactoferrin) for neonatal sepsis may help expand therapeutic options. Research focusing on point-of-care tests for rapid pathogen identification holds promise for improving treatment precision and minimizing unnecessary antibiotic use.

Conclusion

This prospective observational study highlights the alarming prevalence of multidrug-resistant organisms among neonates with sepsis in a tertiary care center in Eastern India. Gram-negative bacteria, particularly *Klebsiella pneumoniae* and *Escherichia coli*, were the dominant pathogens, and nearly half of all isolates exhibited multidrug resistance. The high rate of antibiotic escalation and substantial mortality (18%) underscore the urgent need for effective infection control and robust antibiotic stewardship measures. Low birth weight and MDR infection emerged as key predictors of mortality, reaffirming the importance of preventive interventions for high-risk groups and rationalizing antibiotic therapy based on local antibiograms. Our findings call for coordinated efforts at the hospital and policy level to mitigate further spread of antimicrobial resistance and to improve outcomes in this vulnerable population. By generating local epidemiological data on neonatal sepsis, antibiotic use, and resistance profiles, this study serves as a foundation for developing evidence-based protocols for empirical treatment regimens and enhancing stewardship practices. The ultimate goal remains the reduction of neonatal mortality and morbidity through timely, appropriate, and effective management of sepsis, especially in the face of the escalating threat of antibiotic resistance.

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