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Original Research Article

Blood culture patterns and antibiotic resistance in preterm low birth weight babies with neonatal sepsis

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ABSTRACT

Background: Neonatal sepsis remains a leading cause of morbidity and mortality among preterm infants and rising antimicrobial resistance (AMR) among Gram-negative pathogens poses a significant challenge to treatment. This study evaluated blood culture patterns, antibiotic resistance and clinical outcomes in neonates with sepsis admitted to a tertiary care center in Bangladesh.

Methods: A retrospective observational study was conducted in the NICU of Bangladeshi Specialized Hospital, Dhaka, from January 2023 to December 2024. A total of 144 neonates (0–28 days) with suspected or confirmed bacterial sepsis were analyzed. Demographic, clinical, laboratory and treatment data were extracted and blood culture isolates were tested for antibiotic susceptibility using the VITEK 2 system per CLSI guidelines.

Results: The majority of neonates were male (59.7%) and early preterm (50%), with a mean admission age of 2.92±2.21 days. Late-onset sepsis predominated (73.61%) and respiratory distress was the most common clinical sign (66.67%). Blood culture positivity was 22.92%, with *Klebsiella pneumoniae* (51.52%) and *Acinetobacter* spp. (33.33%) being the most common isolates. *Klebsiella pneumoniae* was 100% sensitive to tigecycline and 94.1% to colistin but showed <6% sensitivity to first-line antibiotics. Supportive care included phototherapy (75%), NG feeding (70.14%) and CPAP (63.89%). Survival at discharge was 96.53%, with a mean NICU stay of 14.82±8.24 days.

Conclusions: MDR Gram-negative sepsis is a growing threat in NICUs, with declining efficacy of first-line antibiotics. The reliance on colistin and tigecycline raises concerns for future treatment options if resistance to these agents develops. Strong infection control, antimicrobial stewardship and updated antibiogram surveillance are essential to combat this trend.

Keywords: Antibiotic resistance, *Acinetobacter* spp., Blood culture, *Klebsiella pneumoniae*, Multidrug resistance, NICU, Neonatal sepsis

INTRODUCTION

Neonatal sepsis remains a major global health challenge, particularly in low- and middle-income countries (LMICs), where it contributes significantly to neonatal morbidity and mortality. It is defined as a systemic inflammatory response to bacterial infection occurring in the first 28 days of life and is typically classified into early-onset sepsis (EONS, <72 hours of life) and late-onset sepsis (LONS, ≥72 hours).[1] Preterm and low birth weight neonates are especially vulnerable to sepsis due to their immature immune systems, frequent exposure to invasive procedures and prolonged hospital stays in neonatal intensive care units (NICUs).[2]

Globally, sepsis accounts for approximately 23% of neonatal deaths, with nosocomial infections further amplifying this burden in resource-limited settings.[3] The emergence of multidrug-resistant organisms (MDROs) such as *Klebsiella pneumoniae*, Acinetobacter species

and Pseudomonas aeruginosa complicates therapeutic strategies, often necessitating the use of broad-spectrum or last-resort antibiotics.[4,5] In particular, the overuse of third-generation cephalosporins and carbapenems has led to the proliferation of extended-spectrum beta-lactamase (ESBL)-producing organisms and plasmid-mediated resistance, severely limiting effective treatment options.[6,7]

Although blood culture remains the gold standard for diagnosing neonatal sepsis, it is hampered by low sensitivity and delayed reporting, often necessitating the empirical initiation of antibiotic therapy based on clinical suspicion.[8] However, this practice increases the risk of inappropriate therapy and contributes to the selection pressure for antimicrobial resistance.[9] Particularly in NICUs across LMICs, empirical protocols often lack regular updates based on local antibiograms and infection control measures are inconsistently enforced.[10]

In Bangladesh, the scenario is further complicated by limited surveillance systems and inconsistent microbiological diagnostics, making it difficult to track resistance patterns systematically. Consequently, treatment failures, prolonged NICU admissions and recurrent infections are increasingly reported.[11] This study aims to explore the spectrum of bacterial pathogens and their antibiotic resistance profiles among preterm, low birth weight neonates diagnosed with sepsis in a tertiary care hospital setting.

By analyzing blood culture results, laboratory parameters and clinical outcomes, we aim to generate critical insights to guide empirical antibiotic selection and promote targeted infection control strategies. Understanding these patterns is essential to optimize neonatal care, reduce antimicrobial resistance and improve survival outcomes among high-risk neonates.

METHODS

This retrospective observational study was conducted in the NICU of Bangladeshi Specialized Hospital in Dhaka, Bangladesh over a two-year period, from January 2023 to December 2024.

A total of 144 neonates aged 0 to 28 days with suspected or confirmed bacterial sepsis were included. Inclusion criteria encompassed both early-onset sepsis (EONS, <72 hours) and late-onset sepsis (LONS, ≥72 hours) with clinical or laboratory evidence of infection. Neonates with perinatal asphyxia (PNA), congenital heart disease (CHD), congenital anomalies, incomplete medical records or those transferred out before treatment completion were excluded.

Data were extracted from electronic medical records and included demographic details (sex, gestational age, birth weight, mode of delivery, APGAR scores), clinical features (admitting diagnosis, sepsis-related signs, complications), laboratory findings (complete blood count, CRP, serum electrolytes, procalcitonin, blood culture), treatment details (antibiotics used, duration, supportive interventions) and outcomes (length of NICU stay, survival, re-admission and follow-up status). Anemia was defined as a hemoglobin (Hb) level of <14 g/dl for preterm neonates, in line with standard neonatal hematology references.

Leukopenia was defined as a total white blood cell (WBC) count of $<5\times10^9$/l, while leukocytosis was considered at counts exceeding $>30\times10^9$/l. Clinical diagnosis of sepsis was based on WHO criteria and assessed using the SOAP (Subjective, Objective, Assessment, Plan) framework. Subjective symptoms such as lethargy and poor feeding were paired with objective signs like respiratory distress, abnormal vital parameters and elevated inflammatory markers. Blood samples for culture were collected aseptically prior to antibiotic initiation and processed using the BACTEC automated culture system.

Organism identification and antibiotic susceptibility testing were performed using the VITEK 2 system, with interpretation according to CLSI guidelines. All patients received empirical first-line treatment with ampicillin and gentamicin and subsequent antibiotic therapy was guided by the sensitivity patterns of isolated organisms, which were predominantly tested against ceftriaxone, ceftazidime, amikacin, meropenem, colistin, imipenem, linezolid, ciprofloxacin, cloxacillin and tobramycin.

Descriptive statistics were used to analyze the data. Continuous variables were presented as means with standard deviations and ranges, while categorical variables were summarized as frequencies and percentages. All analyses were performed using SPSS version 27.0.

RESULTS

Out of the 144 neonates included in the study, 59.7% (n=86) were male, while 40.3% (n=58) were female. Regarding the mode of delivery, 52.8% (n=76) were delivered via lower uterine cesarean section (LUCS), whereas 47.2% (n=68) were born through normal vaginal delivery (NVD). In terms of prematurity classification, 50% (n=72) were early preterm, 30.56% (n=44) were preterm and 19.44% (n=28) were late preterm.

The mean age at admission among the neonates was 2.92±2.21 days, ranging from 0 to 9 days. Regarding the diagnosis at admission, respiratory distress syndrome (RDS) was the most common diagnosis, accounting for 52.08% (n=75) of cases.

This was followed by hypoglycemia in 26.39% (n=38), infants of diabetic mothers (IDM) in 15.28% (n=22), meconium aspiration syndrome (MAS) in 4.86% (n=7), congenital pneumonia in 4.17% (n=6) and transient tachypnea of the newborn (TTN) in 2.78% (n=4). On

clinical assessment at admission, respiratory distress was the predominant sign, observed in 66.67% (n=96) of neonates. Other notable findings included lethargy in 33.33% (n=48), sclerema neonatorum in 31.25% (n=45), hypothermia in 20.83% (n=30), feeding intolerance in 19.44% (n=28), hyperthermia in 16.67% (n=24) and recurrent apnea in 7.64% (n=11).

Neonatal convulsions were relatively uncommon, occurring in 2.78% (n=4) of cases. With respect to the timing of sepsis, late-onset sepsis (LONS) was predominant in 73.61% (n=106) of the neonates, while early-onset sepsis (EONS) accounted for 26.39% (n=38). Among the clinical complications observed in the study population, retinopathy of prematurity was the most common, affecting 38.19% (n=55) of neonates.

Pneumothorax occurred in 8.33% (n=12), while anemia of prematurity was reported in 6.94% (n=10). Other complications included necrotizing enterocolitis (NEC) in 5.56% (n=8), intraventricular hemorrhage (IVH) in 4.86% (n=7) and both sepsis-related shock or multiple organ dysfunction syndrome (MODS) and interpulmonary hemorrhage in 2.78% (n=4) each.

Among the laboratory investigations, 26.39% (n=38) of neonates were anemic with hemoglobin levels <14 g/dl, while 73.61% (n=106) had normal hemoglobin levels between 14–16 g/dl. Regarding white blood cell counts, 71.53% (n=103) exhibited leukopenia (<5×10⁹/l), 18.75% (n=27) were within the normal range (5–30×10⁹/l) and 9.72% (n=14) had leukocytosis (>30×10⁹/l). Platelet count abnormalities were also common, with thrombocytopenia (<150×10⁹/l) detected in 63.89% (n=92) of the population, whereas 33.33% (n=48) had normal platelet counts and 2.78% (n=4) showed thrombocytosis (>400×10⁹/l).

Inflammatory markers revealed that CRP levels were elevated (≥5 mg/l) in 73.61% (n=106), while 26.39% (n=38) had baseline CRP levels (<5 mg/l). Blood culture positivity was 22.92% (n=33), with *Klebsiella pneumoniae* being the most frequently isolated organism (51.52%, n=17), followed by Acinetobacter species (33.33%, n=11), *Escherichia coli* (9.09%, n=3), Enterococcus species (3.03%, n=1) and *Pseudomonas aeruginosa* (3.03%, n=1).

In terms of biochemical parameters, hypocalcemia (<8.5 mg/dl) was found in 56.94% (n=82), whereas 43.06% (n=62) had normal calcium levels. For serum sodium, hyponatremia (<135 mmol/l) occurred in 45.83% (n=66) and 54.17% (n=78) had normal sodium levels, with no cases of hypernatremia observed. Serum potassium levels showed that 11.11% (n=16) had hypokalemia (<3.5 mmol/l), while 88.89% (n=128) were within the normal range (3.5–5.5 mmol/l). Among the 33 culture-positive isolates, *Klebsiella pneumoniae* (n=17) and *Acinetobacter* species (n=11) exhibited high resistance to most first-line antibiotics. *Klebsiella pneumoniae* showed 100% sensitivity to tigecycline, while colistin retained activity against 94.1% of isolates. However, sensitivity to other agents such as amikacin, gentamicin, meropenem, piperacillin/tazobactam and netilmicin was notably low (all at 5.9%).

Acinetobacter species demonstrated 90.9% sensitivity to tigecycline and 81.8% sensitivity to colistin, but only 18.2–27.3% sensitivity to most aminoglycosides and beta-lactams. *Escherichia coli* (n=3) isolates displayed variable sensitivity, with 66.7% sensitive to gentamicin and netilmicin and 33.3% sensitive to agents such as amikacin, aztreonam, cefepime, ciprofloxacin, linezolid, meropenem, piperacillin/tazobactam and tobramycin.

Enterococcus species (n=1) was 100% sensitive to doxycycline, gentamicin and linezolid, while Pseudomonas aeruginosa (n=1) was 100% sensitive to all tested antibiotics, including amikacin, aztreonam, cefepime, ceftazidime, ciprofloxacin, meropenem, piperacillin/tazobactam, gentamicin and tobramycin.

Among the 33 culture-positive cases requiring second-line antibiotics, colistin was the most frequently administered agent (78.79%, n=26), followed by tigecycline (51.52%, n=17). Other antibiotics used included amikacin (15.15%, n=5), gentamicin (15.15%, n=5), piperacillin/tazobactam (15.15%, n=5), ciprofloxacin (12.12%, n=4) and meropenem (12.12%, n=4).

Less commonly used agents were cefotaxime (6.06%, n=2), cotrimoxazole (6.06%, n=2) and cefepime (3.03%, n=1). The average duration of antibiotic therapy was 9.49±5.46 days, ranging from 2 to 42 days. Notably, all neonates initially received a first-line combination of ampicillin, gentamicin, cefotaxime and piperacillin prior to culture results.

Supportive interventions were widely utilized, with phototherapy being the most common (75%, n=108), followed by nasogastric (NG) feeding (70.14%, n=101) and CPAP respiratory support (63.89%, n=92). Additionally, packed red blood cell (PRBC) transfusion was administered to 33.33% (n=48) of neonates, while penta globin was given to 17.36% (n=25) and platelet transfusion was required in 12.5% (n=18).

The mean length of NICU stay among the neonates was 14.82±8.24 days, with durations ranging from 2 to 35 days. At discharge, 96.53% (n=139) of the neonates survived, while 3.47% (n=5) succumbed to illness despite treatment. Regarding readmissions, 93.75% (n=135) of the neonates were not re-admitted within 7 days of discharge, whereas 4.86% (n=7) required re-admission. Additionally, 95.83% (n=138) of the discharged neonates had a follow-up visit scheduled on the 7th day post-discharge.

Table 1: Distribution of the study population based on patient identification and demographics (n=144).

Patient identification and demographics	Frequency (N)	(%)
Sex		
Male	86	59.7
Female	58	40.3
Mode of delivery		
LUCS	76	52.8
NVD	68	47.2
Preterm category		
Early preterm	72	50.00
Preterm	44	30.56
Late preterm	28	19.44

Table 2: Distribution of the study population based on admission and clinical status (n=144).

Variable	Frequency (N)	(%)
Age at admission	**Mean±SD=2.92±2.21 days**	**Range: 0–9 days**
Diagnosis at admission		
Respiratory distress syndrome (RDS)	75	52.08
Hypglycemia	38	26.39
IDM	22	15.28
Congenital pneumonia	6	4.17
Meconium aspiration syndrome (MAS)	7	4.86
Transient tachypnea of the newborn (TTN)	4	2.78
Clinical signs on admission		
Respiratory distress	96	66.67
Lethargy	48	33.33
Hypothermia	30	20.83
Feeding intolerance	28	19.44
Sclerema neonatorum	45	31.25
Recurrent apnea	11	7.64
Hyperthermia	24	16.67
Neonatal convulsion	4	2.78
Timing of sepsis		
Late onset	106	73.61
Early onset	38	26.39

Table 3: Distribution of the study population based on clinical complications (n=144).

Complication	Frequency (N)	(%)
NEC (necrotizing enterocolitis)	8	5.56
IVH (intraventricular hemorrhage)	7	4.86
Pneumothorax	12	8.33
Sepsis-related shock or MODS	4	2.78
Retinopathy of prematurity	55	38.19
Interpulmonary hemorrhage	4	2.78
Anemia of prematurity	10	6.94

Table 4: Distribution of the study population based on laboratory investigation (n=144).

Parameter	Range/Categories	Frequency (N)	(%)
Hemoglobin (g/dl)			
Anemic	<14 g/dl	38	26.39
Normal	14–16 g/dl	106	73.61
WBC count (cells/cu mm)			
Leukopenia	<5×10⁹/l	103	71.53
Normal	5–30×10⁹/l	27	18.75

Continued.

Parameter	Range/Categories	Frequency (N)	(%)
Leukocytosis	>30×10⁹/l	14	9.72
Platelet count (cells/cu mm)			
Thrombocytopenia	<150×10⁹/l	92	63.89
Normal	150–400×10⁹/l	48	33.33
Thrombocytosis	>400×10⁹/l	4	2.78
C-reactive protein (CRP			
Baseline (Normal)	<5 mg/l	38	26.39
Elevated (Acute-phase)	≥5 mg/l	106	73.61
Blood culture			
Positive	-	33	22.92
Negative	-	111	77.08
Organisms isolated (n=33)			
Klebsiella pneumoniae	-	17	51.52
Acinetobacter species	-	11	33.33
Escherichia coli	-	3	9.09
Enterococcus species	-	1	3.03
Pseudomonas aeruginosa	-	1	3.03
Serum calcium			
Hypocalcemia	<8.5 mg/dl	82	56.94
Normal	8.5–10.5 mg/dl	62	43.06
Hypercalcemia	>10.5 mg/dl	0	0.00
Serum sodium			
Hyponatremia	<135 mmol/l	66	45.83
Normal	135–145 mmol/l	78	54.17
Hypernatremia	>145 mmol/l	0	0.00
Serum potassium			
Hypokalemia	<3.5 mmol/l	16	11.11
Normal	3.5–5.5 mmol/l	128	88.89

Table 5: Antimicrobial sensitivity pattern of isolated organisms (n=33).

Organism (N)	*K. pneumoniae* (n=17)	*Acinetobacter* species (n=11)	*E. coli* (n=3)	*Enterococcus* species (n=1)	*P. aeruginosa* (n=1)
Amikacin	1 (5.9%)	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Amoxiclav	0	2 (18.2%)	0	0	0
Aztreonam	0	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Cefepime	0	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Cefixime	0	2 (18.2%)	0	0	0
Cefotaxime	0	2 (18.2%)	0	0	0
Ceftazidime	0	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Ceftriaxone	0	2 (18.2%)	0	0	0
Ciprofloxacin	0	0	1 (33.3%)	0	1 (100.0%)
Colistin	16 (94.1%)	9 (81.8%)	1 (33.3%)	0	0
Cotrimoxazole	1 (5.9%)	2 (18.2%)	0	0	0
Doxycycline	0	0	1 (33.3%)	1 (100.0%)	0
Gentamicin	1 (5.9%)	2 (18.2%)	2 (66.7%)	1 (100.0%)	1 (100.0%)
Linezolid	0	0	1 (33.3%)	1 (100.0%)	0
Meropenem	1 (5.9%)	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Netilmicin	1 (5.9%)	3 (27.3%)	2 (66.7%)	0	1 (100.0%)
Piperacillin/Tazobactam	1 (5.9%)	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Tetracycline	6 (35.3%)	2 (18.2%)	0	0	0
Tigecycline	17 (100.0%)	10 (90.9%)	0	0	0
Tobramycin	1 (5.9%)	2 (18.2%)	1 (33.3%)	0	1 (100.0%)
Vancomycin	0	0	1 (33.3%)	1 (100.0%)	0

Table 6: Distribution of the study population based on the treatment and interventions (n=144).

Treatment and interventions	Frequency (N)	(%)
Antibiotics treatment (second line, n=33)		
Colistin	26	78.79
Tigecycline	17	51.52
Amikacin	5	15.15
Meropenem	4	12.12
Gentamicin	5	15.15
Ciprofloxacin	4	12.12
Piperacillin/Tazobactam	5	15.15
Cefepime	1	3.03
Cefotaxime	2	6.06
Cotrimoxazole	2	6.06
Duration of antibiotics (days)		
Mean±SD	9.49±5.46	
Minimum	2 days	
Maximum	42 days	
Supportive interventions		
CPAP	92	63.89
NG Feeding	101	70.14
Phototherapy	108	75.00
PRBC transfusion	48	33.33
Penta globin	25	17.36
Platelet transfusion	18	12.50

Table 7: Distribution of the study population based on outcome (n=144).

Outcome measure	Value/Frequency (N)	(%)
Length of NICU stay (days)	Mean±SD: 14.82±8.24	
Minimum	2 days	
Maximum	35 days	
Survival status at discharge		
Survived	139	96.53
Death	5	3.47
Re-admission within 7 days		
Not re-admitted	135	93.75
Re-admitted	7	4.86
Follow-up scheduled on 7th day post-discharge	138	95.83

DISCUSSION

This study provides an overview of neonatal sepsis patterns, antimicrobial resistance and outcomes in a tertiary care center in Bangladesh. The findings reveal an alarming rise in multidrug-resistant (MDR) infections, with culture-positive cases showing limited sensitivity to commonly used antibiotics. This is a serious concern for future management of neonatal sepsis, as the treatment options are narrowing. Most of the neonates in our study were male (59.7%) and early preterm (50%), with over half delivered via cesarean section (52.8%). These patterns are consistent with findings by Saini et al where preterm and male neonates were more frequently affected by sepsis.[12] The mean age at admission was 2.92 days, which is similar to that reported by Sisay et al.[13]

Respiratory distress syndrome (RDS) was the most common diagnosis (52.08%) and respiratory distress was seen in 66.67% of patients. Late-onset sepsis (LONS) dominated, with 73.61% of cases. Similar clinical features and higher LONS rates have been reported by Ogundare et al.[14]

Among complications, retinopathy of prematurity was the most frequent (38.19%), followed by pneumothorax (8.33%) and NEC (5.56%). Studies like Kariniotaki et al have reported comparable rates of complications in septic preterm infants.[15] Blood culture positivity was 22.92% and most isolates were Gram-negative bacteria, mainly *Klebsiella pneumoniae* (51.52%) and Acinetobacter spp. (33.33%). Similar patterns were observed in South Asian NICU studies.[16,17] The antimicrobial sensitivity profile is concerning. *Klebsiella pneumoniae* showed 100%

sensitivity to tigecycline and 94.1% to colistin, but only 5.9% sensitivity to amikacin, gentamicin and meropenem. *Acinetobacter* spp. also showed high sensitivity only to tigecycline (90.9%) and colistin (81.8%). These findings mirror reports by Chatterjee et al which highlighted the limited efficacy of traditional first-line antibiotics.[18] If resistance to colistin or tigecycline emerges, the treatment of neonatal sepsis will become extremely difficult. All neonates initially received ampicillin, gentamicin, cefotaxime and piperacillin as empiric therapy. In culture-positive cases, colistin (78.79%) and tigecycline (51.52%) were most commonly used as second-line treatments, which is consistent with Ipek et al.[19] The mean antibiotic duration was 9.49 days. Supportive measures such as phototherapy (75%), NG feeding (70.14%) and CPAP (63.89%) were widely used, as also noted in other NICU studies.[20]

Outcomes were favorable, with 96.53% survival and a low mortality rate of 3.47%. Early re-admission was 4.86%, while follow-up was arranged for 95.83% of neonates. Similar survival outcomes have been observed in Saini et al.[12] In summary, the study highlights the growing threat of MDR neonatal sepsis, particularly with Gram-negative organisms. The limited sensitivity to first-line antibiotics and dependence on last-line agents like colistin and tigecycline point to a critical need for better antibiotic stewardship, strict infection control measures and ongoing surveillance.

Limitations

This study was limited by its single-center, retrospective design, which may reduce the generalizability of the findings to other settings. Additionally, only culture-positive cases were analyzed for antimicrobial resistance patterns and molecular testing for resistance mechanisms was not performed due to resource constraints.

CONCLUSION

This study highlights the growing challenge of MDR neonatal sepsis in Bangladesh, with Gram-negative organisms such as *Klebsiella pneumoniae* and *Acinetobacter* spp. dominating blood culture isolates. The limited sensitivity to first-line antibiotics and reliance on last-line drugs like colistin and tigecycline underscore the urgent need for effective antibiotic stewardship and enhanced infection control strategies. Although overall survival was high (96.53%), the alarming resistance trends call for robust surveillance, regional antibiogram updates and preventive measures to mitigate the future impact of MDR infections on neonatal outcomes.

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