DOI: https://doi.org/10.53350/pjmhs020241811033

ORIGINAL ARTICLE

Trends of Microorganisms and Antibiotic Sensitivity Patterns in Neonatal Sepsis at a Tertiary Care Hospital in Islamabad

NAILA RAFIQUE[1], NIDA BASHARAT KHAN[2], NOSHEEN NABI[3], SIDRA SHEIKH[4], SIDRA NOSHEEN KIYANI[5], SABAH KALEEM[6]
[1]*Senior Registrar Microbiology, Rawal Institute of Health Sciences, Islamabad*
[2]*Assistant Professor Chemical Pathology, Rawal Institute of Health Sciences, Islamabad*
[3]*Assistant Professor Histopathology, Rawal Institute of Health Sciences, Islamabad*
[4]*Senior Registrar Chemical Pathology, Rawal Institute of Health Sciences, Islamabad*
[5]*Assistant Professor Haematology, Rawal Institute of Health Sciences, Islamabad*
[6]*Associate Professor of Histopathology, Rawal Institute of Health Sciences, Islamabad*
Correspondence to: Naila Rafique, Email: nailarafiquemughal@gmail.com

ABSTRACT

Background: Neonatal sepsis remains a leading cause of morbidity and mortality worldwide. Early identification of causative organisms and their antibiotic susceptibility is essential for effective management, particularly in settings with rising antimicrobial resistance.

Objective: To determine the distribution of pathogens causing neonatal sepsis and assess their antibiotic sensitivity patterns at Rawal General Hospital, Islamabad.

Material and Methods: This retrospective study analyzed 350 clinical samples from neonates aged 0–28 days with suspected sepsis, collected between August 2022 and August 2023. Standard culture methods were employed for bacterial isolation, and organisms were identified using Gram staining, colony morphology, and biochemical tests. Antibiotic susceptibility testing was performed using the Kirby–Bauer disk diffusion method following CLSI guidelines. Tested antibiotics included ampicillin, gentamicin, amikacin, cefotaxime, ceftazidime, meropenem, piperacillin-tazobactam, ciprofloxacin, and vancomycin.

Results: Culture positivity was observed in 23 (6.6%) samples. Gram-negative bacteria predominated (73.9%), with Escherichia coli (30%) and Klebsiella pneumoniae (26%) being most frequent. Gram-positive isolates included Staphylococcus aureus (17%) and coagulase-negative staphylococci (13%). High resistance was noted to ampicillin and third-generation cephalosporins, while amikacin and meropenem were largely effective.

Conclusion: Gram-negative bacteria are the primary pathogens in neonatal sepsis at this hospital, with significant resistance to first-line antibiotics. Continuous surveillance of antimicrobial susceptibility is essential to guide empirical therapy and improve neonatal outcomes.

Keywords: Neonatal sepsis; Gram-negative bacteria; Antibiotic resistance; Antimicrobial susceptibility

INTRODUCTION

Neonatal sepsis is a major health complication in the world and one of the major causes of neonatal morbidity and mortality, especially in the low and middle-income nations. It is a systemic infection that develops during the first 28 days of life, which can be either early-onset or late-onset disease depending on the time taken to develop. The clinical picture is usually mild and unspecific, such as poor nutrition, miosis, sporadic temperature changes, dyspnea, and sleep apnea that makes it harder to identify it early and provide timely care[1, 2].

The burden of neonatal sepsis is high even in spite of the development of neonatal intensive care and antimicrobial treatment. The gold standard of diagnosis remains the blood culture, though, the positivity rate of the cultures is often low, such as the reasons being exposures to antibiotics, poor volume sampling of the blood, and low bacterial load of the neonate[3]. Empirical antibiotic treatment is also often started and hence the need to comprehend the local pathogen profile and antimicrobial resistance pattern.

Neonatal sepsis etiological agents vary in different regions and healthcare facilities. Gram- Negative organisms like Escherichia coli and Klebsiella pneumoniae are always found to be the most frequent pathogens, particularly in studies which have been conducted in hospitals in the developing countries[4,5]. These organisms are usually related to serious disease, high rate of progression and increased mortality. Gram-positive bacteria, especially Staphylococcus aureus and coagulase-negative staphylococci also play an important role in neonatal sepsis, particularly in late-onset and healthcare-associated infections[6].

There is a reported alarming increase of antimicrobial resistance of neonatal pathogens across the globe. The resistance rates to first-line antibiotics (ampicillin and third generation cephalosporins) have been found to be high thus reducing their efficacy forcing people to use wider spectrum agents like aminoglycosides and carbapenems[7]. There is not only complication in treatment but also problem of increased hospital stay, healthcare costs and risk of mortality among neonates brought about by the emergence of multidrug-resistant organism.

Local data on surveillance are essential in directing the empirical policies on antibiotics and enhancing antimicrobial stewardship efforts. The trend of resistance can also differ considerably even within the same nation because of the variations in the practice of antibiotic use, infection control, and hospital infrastructure[8]. Hence, it is necessary to use the institution-specific data to optimize treatment schemes and enhance neonatal outcomes.

Neonatal sepsis is one of the most topical public health issues in Pakistan, but there is a lack of up-to-date and centre-specific data on causative organisms and antimicrobial susceptibility profiles. Knowledge of the local epidemiology of neonatal sepsis would be of paramount importance to treatment guidelines based on evidence and the emergent threat of antimicrobial resistance in the neonatal care environment.

Objective: To identify patterns of distribution of microorganisms that cause neonatal sepsis and also to observe the patterns of their antibiotic sensitivity in Rawal General Hospital, Islamabad.

MATERIAL AND METHODS

Study Design and Setting: This was a retrospective observational study that was carried out in the Department of Microbiology at Rawal General Hospital, which is a tertiary care healthcare institution that offers neonatal services, including intensive care services.

Study Period: The research was conducted during one year of the period, between August 2022 and August 2023.

Study Population: The population that was used in the study was a group of neonates (0-28 days old) whose samples were put in the Microbiology Laboratory to undergo a routine culture of bacteria throughout the study.

Received on 21-10-2023
Accepted on 29-12-2023

Sample Size and Sampling Methodology: The study involved 350 clinical samples of suspected cases of sepsis in neonates. The sampling was done consecutively and all the possible samples that were obtained throughout the period of study were analyzed.

Inclusion and Exclusion Criteria: Infants at the age of 0-28 days with clinical suspicion of sepsis and samples submitted to bacterial culture within the period of the research were eligible to take part. The neonates with age above 28 days, contaminated or poorly collected samples and recurring or duplicate samples within a single patient were not included into the analysis.

Processing and Collection of Specimens: The collection of blood samples and other relevant clinical samples was done under strict aseptic conditions in line with the protocols of the hospital. The blood samples were put in the right culture media and carried to the Microbiology Laboratory to be processed.

Microbiological Culture and Identification: The samples were grown on blood agar, MacConkey agar, and other suitable selective media according to the general practice of the laboratory. The plates were inoculated with culture and incubated aerobically at 37C and observed to grow after 24 and 48 hours. The bacterial isolates were identified using Gram staining, morphology, and of the standard biochemicals.

The test is known as the antibiotic susceptibility test.

The Kirby-Bauer disk diffusion method of antibiotic susceptibility testing was done on Mueller-Hinton agar. The interpretation of the results was based on the Clinical and Laboratory Standards Institute (CLSI). Antibiotics that were tested were ampicillin, amoxicillin-clavulanate, gentamicin, amikacin, cefotaxime, ceftazidime, ceftriaxone, piperacillin-tazobactam, ciprofloxacin, vancomycin and merpenem. Only gram-positive isolates were subjected to vancomycin susceptibility testing.

Quality Control: The testing of culture media and antibiotic discs was done according to the CLSI recommendations by quality assurance of standard reference strains. All the laboratory works were conducted by competent microbiology staff in accordance with standard operating procedures.

Data collection and analysis Data collection and analysis: The retrospective review of laboratory records was done to obtain information about their patient age, culture outcome, identified organisms and antibiotic susceptibility profile. Statistical software was used in entering and analyzing the data. Frequencies and percentages were calculated and reported with the help of descriptive statistics, and represented in tables and figures.

Ethical Considerations: The institutional ethical review committee of Rawal General Hospital was used to get ethical approval of the study. Since the study was retrospective and laboratory-based, informed consent was not used and the patient confidentiality was also ensured.

RESULTS

The study period involved the processing of 350 clinical samples of suspected neonatal sepsis. The overall culture positivity rate was 6.6% and 23 samples grew whereas 327 samples (93.4%) did not grow. Table 1 summarizes the distribution of culture-positive and culture-negative samples.

Out of the 23 culture-positive isolates, Gram-negative bacteria were most common and they included 17 isolates (73.9%), and Gram-positive organisms included 6 isolates (26.1) as demonstrated in Table 2. Escherichia coli was the most commonly isolated organism (30 out of all isolates) followed by Klebsiella pneumoniae (26 out of all isolates). Other Gram-negative causative agents comprised Pseudomonas aeruginosa (9%), Acinetobacter baumannii (4%), and Serratia marcescens (4%). Staphylococcus aureus was the most frequent isolate with 17% followed by coagulase-negative staphylococci (13) and Streptococcus species (4). Table 3 and Figure 1 show the distribution of isolated organisms in detail.

The antibiotic susceptibility test showed that the majority of isolates had a high level of resistance to ampicillin. The gram-negative organisms showed a significant resistance to the third-generation cephalosporins, which include cefotaxime and ceftazidime. Amikacin and meropenem on the other hand were the most sensitive in Gram-negative isolates. Piperacillin-tazobactam and ciprofloxacin were also effective against a number of Gram-negative pathogens. Gram-positive bacteria were highly susceptible to gentamicin, amikacin, and vancomycin and none were resistant to vancomycin. Table 4 displays the sensitive patterns of individual organisms to antibiotics in detail, whereas the comparison patterns are presented in Figure 1.

Table 1: Culture Results of Neonatal Samples (n = 350)

Culture Result	Number	Percentage (%)
Culture positive	23	6.6
Culture negative	327	93.4
Total	350	100

Table 2: Distribution of Isolates by Gram Reaction (n = 23)

Gram Reaction	Number of Isolates	Percentage (%)
Gram-negative bacteria	17	73.9
Gram-positive bacteria	6	26.1
Total	23	100

Table 3: Distribution of Microorganisms Isolated from Neonatal Sepsis

Organism	Number	Percentage (%)
Escherichia coli	7	30
Klebsiella pneumoniae	6	26
Staphylococcus aureus	4	17
Coagulase-negative staphylococci	3	13
Pseudomonas aeruginosa	2	9
Acinetobacter baumannii	1	4
Serratia marcescens	1	4
Streptococcus spp.	1	4
Total	23	100

Figure 1: Distribution of Microorganisms Causing Neonatal Sepsis

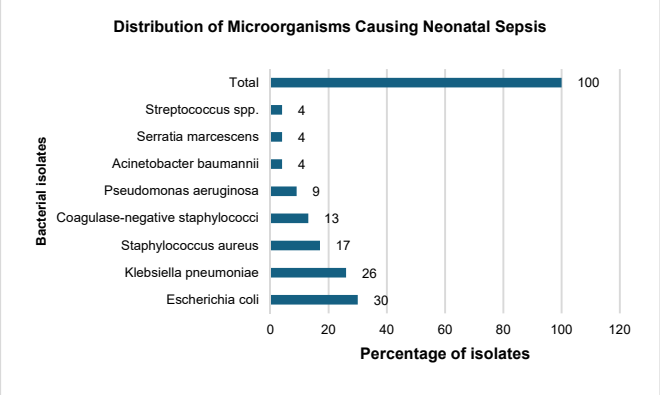

Table 4: Antibiotic Sensitivity Pattern of Bacterial Isolates
R = Resistant, S = Sensitive
Vancomycin tested only for Gram-positive organisms

Organism	Ampicillin	Gentamicin	Amikacin	Cefotaxime	Ceftazidime	Ciprofloxacin	Meropenem	Vancomycin
E. coli	R	S	S	R	R	S	S	–
K. pneumoniae	R	S	S	R	R	S	S	–
S. aureus	R	S	S	S	S	S	S	S
CONS	R	S	S	S	S	S	S	S
P. aeruginosa	R	S	S	S	S	S	S	–
A. baumannii	R	S	S	R				
Serratia spp.	R	S	S	R				
Streptococcus spp.	S	S	S	S				

DISCUSSION

It is shown that the Gram-negative bacteria were the major causative agent of neonatal sepsis and close to three-quarters of all culture-positive isolates in the current study. The most common pathogens were Escherichia coli and Klebsiella pneumoniae. The given finding is also in line with several recent studies that have revealed a similar prevalence of Gram-negative organisms in neonatal sepsis, especially in hospital-based conditions, in which these pathogens are linked to serious illness and poor outcomes[9,10].

The rate of culture positivity in this research was not that high. Similar studies have also found the presence of variable culture positivity rates with low to moderate level of culture positivity. These discrepancies are often explained by early exposure to empirical antibiotics prior to blood sampling, little bacteria in the circulation of newborns, and issues connected to specimen size and the time and place of collection[11,12]. These aspects still restrain microbiological validation of neonatal sepsis and emphasize the use of empirical treatment in clinical practice.

Patterns of resistance within Gram-negative isolates are of great concern. Ampicillin resistance as well as third-generation cephalosporin was high especially against E. coli and Klebsiella pneumoniae. There is a similar pattern in resistance to beta-lactam antibiotics in recent multicentre studies that reported extensive resistance to standard beta-lactam antibiotics in the neonatal sepsis pathogens[13,14]. This resistance significantly decreases the potency of the standard first-line treatment regimens.

AMI and meropenem on the other hand were active against the majority of Gram-negative isolates in the current study. Recent studies also have stated maintained vulnerability of the neonatal sepsis pathogens to the aminoglycosides and carbapenems, rendering them essential choices in high cephalosporin resistance settings[15,16]. Nonetheless, the growing reliance on carbapenems brings a question about the possibility of the coming up with carbapenem-resistant organisms in the future.

Staphylococcus aureus and coagulase-negative staphylococci were found in more than a quarter of the isolates (gram-positive organisms). They continue to play a significant role in the neonatal sepsis, particularly in healthcare-associated infections. It was highly susceptible to vancomycin, gentamicin, and amikacin, which is in line with the recent research publications that have indicated the continued use of glycopeptides on Gram-positive neonatal pathogens[17]. The lack of vancomycin resistance in the study is an encouraging situation that will have to undergo further monitoring.

The continued resistance to first-line antibiotics underscores the pressing need to upgrade the local antibiograms as a basis to inform the empirical treatment. Some of the recent reports have highlighted that the presence of empirical antibiotic policies that do not agree with local resistance trends have led to poor clinical outcomes and enhancements in antimicrobial resistance[18,19]. It is thus critical that the neonatal care is optimized by regular monitoring and updating of treatment guidelines.

Comprehensively, the results of this research support the current concern in the world about the dynamic microbiological profile of neonatal sepsis and the increasing problem of antimicrobial resistance. Tightening of infection prevention measures, reasonable antibiotic use, and effective antimicrobial stewardship interventions, is essential to decrease the burden of resistant neonatal infections and enhance survival outcomes[20].

Limitations: There are some limitations of this study. Its retrospective nature restricted the availability of more clinical data, such as when sepsis developed, maternal risk factors and patient outcomes. The low amount of culture-positive cases and the fact that this study was carried out in one center could limit the applicability of the results. Also, the molecular characterization of the resistance mechanisms was not carried out, which would have given more information on the antimicrobial resistance patterns observed.

CONCLUSION

Gram-negative bacteria become the causative organisms of neonatal sepsis at Rawal General Hospital, Escherichia coli and Klebsiella pneumoniae were the most common isolates. It exhibited high resistance to first-line antibiotics, especially ampicillin and the third -generation cephalosporins, but meropenem and amikacin were not resistant. These results underscore the urgency of ongoing local surveillance of pathogens and their pattern of antimicrobial susceptibility to guide empirical treatment, enhance antibiotic stewardship, and have a beneficial clinical outcome in neonates.

REFERENCES

1. Ehsan S, Marium R. Bacteriological profile and antibiotic susceptibility pattern of isolates in neonatal sepsis. Pak J Health Sci. 2023;4(03):44-49. doi:10.54393/pjhs.v4i03.608
2. Javed S, Mehdi N, Majeed N, Tahir A, Mir NA, Javed H. Neonatal sepsis by Gram-negative bacteria and antibiotics susceptibility pattern at a tertiary care pediatric hospital in Pakistan. Infect Dis J Pak. 2023;33(3):112-117. doi:10.61529/idjp.v33i3.325
3. Munir S, Hussain M, Rasheed S, Arshad S, Ijaz S, Qamar N. Susceptibility pattern of bacteria isolated from blood cultures of neonates admitted with sepsis at a tertiary care hospital. Infect Dis J Pak. 2023;33(2):79-85. doi:10.61529/idjp.v33i2.237
4. Bibi S, Ashfaq M, Hussain W, et al. Etiology, risk factors and antibiotic resistance in neonatal sepsis. Prof Med J. 2023;31(08):1206-1211. doi:10.29309/TPMJ/2022.31.08.8240
5. Kurma VR, Raju MS, Manchu T, Manchu K. Neonatal sepsis: Clinical spectrum, bacteriological profile and antibiotic sensitivity patterns in neonatal intensive care unit in a tertiary care Hospital. Int J Contemp Med Res. 2019;6(6):F1-48.
6. Li J, Xia S, Liu Y, Zhang S, Jin Z. Bacteriological profile and antibiotic susceptibility pattern of neonatal septicemia and associated factors of ICU hospitalization days. Infection and drug resistance. 2022 Jan 1:427-38.
7. Worku E, Fenta DA, Ali MM. Bacterial etiology and risk factors among newborns suspected of sepsis at Hawassa, Ethiopia. Sci Rep. 2022;12(1):20187. doi:10.1038/s41598-022-24572-0
8. Li J, Shen L, Qian K. Global, regional, and national incidence and mortality of neonatal sepsis and other neonatal infections, 1990–2019. Front Public Health. 2023;11:1139832. doi:10.3389/fpubh.2023.1139832
9. Atif M, Zia R, Malik I, Ahmad N, Sarwar S. Treatment outcomes, antibiotic use and its resistance pattern among neonatal sepsis patients attending Bahawal Victoria Hospital, Pakistan. PLoS One. 2021;16(1):e0244866. doi:10.1371/journal.pone.0244866
10. Sampath S, Baby J, Krishna B, Dendukuri N, Thomas T. Blood cultures and molecular diagnostics in intensive care units to diagnose sepsis. Indian J Crit Care Med. 2021;25(12):1402-1407. doi:10.5005/jpjournals-10071-23924
11. Chan GJ, Lee AC, Baqui AH, Tan J, Black RE. Risk of early-onset neonatal infection with maternal infection or colonization: a global systematic review and meta-analysis. PLoS Med. 2013;10(8):e1001502. doi:10.1371/journal.pmed.1001502
12. Zaidi AK, Thaver D, Ali SA, Khan TA. Pathogens associated with sepsis in newborns and young infants in developing countries. Pediatr Infect Dis J. 2009;28(1 Suppl):S10-S18. doi:10.1097/INF.0b013e3181958769
13. Adefisoye MA, Olaniran AO. Antimicrobial resistance expansion in pathogens: a review of current mitigation strategies and advances towards innovative therapy. JAC Antimicrob Resist. 2023;5(6):dlad127. doi:10.1093/jacamr/dlad127
14. Minotti C, Di Caprio A, Facchini L, Bedetti L, Miselli F, Rossi C, Della Casa Muttini E, Lugli L, Luppi L, Ferrari F, Berardi A. Antimicrobial resistance pattern and empirical antibiotic treatments in neonatal sepsis: a retrospective, single-center, 12-year study. Antibiotics. 2023 Sep 27;12(10):1488.
15. Flannery DD, Puopolo KM, Hansen NI, Gerber JS, Sánchez PJ, Stoll BJ. Antimicrobial susceptibility profiles among neonatal early-onset sepsis pathogens. The Pediatric infectious disease journal. 2022 Mar 1;41(3):263-71.
16. Jadav YJ, Desai JH, Janjrukia YA. Microbial profile and antibiotic resistance patterns in culture-proven neonatal sepsis: a five-year analysis. Eur J Cardiovasc Med. 2023;15(4):219-222. doi:10.61336/ejcm/25-04-37
17. Rafi NA, Miah MMZ, Wadood MA, Hossain MG. Risk factors and etiology of neonatal sepsis after hospital delivery: a case-control study in a tertiary care hospital. PLoS One. 2020;15(11):e0242275. doi:10.1371/journal.pone.0242275
18. Tessema B, Lippmann N, Knüpfer M, Sack U, König B. Antibiotic resistance patterns of bacterial isolates from neonatal sepsis patients. Antibiotics. 2021;10(3):323. doi:10.3390/antibiotics10030323
19. Masanja PP, Kibusi SM, Mkhoi ML. Predictors of early onset neonatal sepsis among neonates in Dodoma. J Trop Pediatr. 2020;66(3):257-266. doi:10.1093/tropej/fmz062
20. Gebremedhin D, Berhe H, Gebrekirstos K. Risk factors for neonatal sepsis in public hospitals. PLoS One. 2016;11(5):e0154798. doi:10.1371/journal.pone.0154798

This article may be cited as: Rafique N., Khan N. B., Nabi N., Sheikh S., Kiyani S. N., Kaleem S., Trends of Microorganisms and Antibiotic Sensitivity Patterns in Neonatal Sepsis at a Tertiary Care Hospital in Islamabad. Pak J Med Health Sci, 2023; 18(1): 1033-1035.