DOI: https://doi.org/10.53350/pjmhs2023174239

ORIGINAL ARTICLE

Isolated Bacterial Resistance to Antibiotics and Common Pathogens that Cause Neonatal Sepsis

AROOBA IDRIS[1], ASAD SHABBIR[2], FAIQA NAZIR[3], COL.(R) MUHAMMAD AFZAL[4], ASMA YAQUB[5], ANUM RAUF[6]
[1]*Senior Registrar, Rawal Institute of Health Sciences*
[2]*Assistant Professor Pediatrics, Rawalpindi Medical University*
[3]*Assistant Professor, Rawal Institute of Health Sciences*
[4]*Professor/HOD Rawal Institute of Health Sciences*
[5]*Professor, Rawal Institute of Health Sciences*
[6]*Senior Registrar, Rawal Institute of Health Sciences*
Correspondence to: Arooba Idris, Email: a4arooba@hotmail.com, Cell: 0345 5234455

ABSTRACT

Objective: to record the frequency of isolating organisms causing early-onset neonatal sepsis resistant to common antibiotics,

Methodology: This research was a cross-sectional study that was conducted in 2017 and 2018 at the Neonatal Intensive Care Unit of the Benazir Bhutto Hospital in Rawalpindi on 180 children who satisfied the following criteria and were delivered between 34 and 40 weeks of gestation either Spontaneous Vertex Delivery or Cesarean Section. The study was conducted on infants who were born between 34 and 40 weeks of gestation. Those newborns who had congenital defects, had had surgical intervention, had hypoxic ischemic encephalopathy, or had been treated with antibiotics were not included in the research. Every infant had three millilitres of its blood collected and stored in a sterile environment. The blood was placed into the Bactec 9240 blood culture bottles in a direct inoculation. After being transported to the BBH children's hospital pathology lab, the injection bottles were placed in a Bactee 9240 continuous monitoring blood culture system. The temperature inside the incubator was 37 degrees Celsius. The use of catalase, coagulase, and Dnase in the examination of Gram-positive bacteria was carried out. Using the Kirby-Bauer method, the isolated organism was put through a battery of tests to determine whether or not it was resistant to ampicillin, amikacin, tobramycin, cefotaxime, ceftazidime, ceftriaxone, meropenem, sulbactam, and vancomycin. Meuller Hinton Agar was the medium that was employed.

Results: In our study, out of 140 cases, 65.71%(n=92) were between 1-48 hours and 34.29%(n=48) were between 49-72 hours, mean+sd was calculated as 35.96+16.65 hours, 47.86%(n=67) were male and 52.14%(n=73) were females. Frequency of isolated organisms shows that 27.86%(n=39) had S.Aureus, 22.14%(n=31) had E.Coli, 15%(n=21) had Staphylococcus epidermidis, 12.86%(n=18) had Klebseila and 22.14%(n=31) had others.

Practical Implication: determination of the isolated bacterial resistance to antibiotics and common pathogens that cause neonatal sepsis would help in the appropriate and effective management of neonatal sepsis in our population

Conclusion: Our results reveal that S.Aureus and E.Coli were the common showing 27.86% S.Aureus, 22.14% E.Coli, 15%, Staphylococcus epidermidis, 12.86% Klebseila and 22.14% had others isolates. organisms in EOS cases followed by Staphylococcus epidermidis and Klebseila whereas Ampicillin, Cefotaxime, Ceftriaxon, and Ceftazidime are also resistant in these cases.

Keywords: Early onset neonatal sepsis, common pathogens, antimicrobial resistance of isolated organism

INTRODUCTION

The occurrence of sepsis in neonates, defined as any sepsis diagnosed within the first 28 days of life, is a prevalent issue and remains a significant cause of mortality among newborns globally, as evidenced by various studies.[1-3] Despite the surge in published research studies investigating the pathophysiology of sepsis and its treatment strategies over the last two decades, current statistics indicate that there has been no significant improvement in neonatal mortality associated with sepsis. Neonatal sepsis is responsible for approximately one million newborn deaths on a yearly basis worldwide.[4] Sepsis is responsible for a significant proportion of neonatal deaths in middle and low-income countries, accounting for approximately 30%-50% of such fatalities.[5] The prevention of mortality related to sepsis can be achieved through prompt identification and swift intervention with efficacious antimicrobial agents.[6] Regrettably, the absence of prompt and resilient diagnostic techniques to identify the intruding pathogens responsible for sepsis is evident.[7] The present diagnostic technique that is considered the benchmark, namely blood culture, exhibits inconsistent sensitivity ranging from 50% to 80% and may require several hours to days to generate outcomes.[8] The commencement of empirical antimicrobial therapy is crucial in suspected cases of sepsis, as stated in reference.[9] The identification of the predominant pathogens responsible for neonatal sepsis and their corresponding susceptibility patterns can assist medical practitioners in administering empirical treatment with the suitable antimicrobial agents.

However, even within a single nation, the pathogens that cause neonatal sepsis vary from place to place, and their distribution patterns change over time. Additionally, a lot of them have become more resistant to a variety of commonly used antimicrobial agents, making treatment very challenging;[10-11] in addition, the pattern of their antimicrobial resistance varies from one geographic location to another.[12-13] In order to update the empirical therapeutic approach for suspected sepsis with efficient and targeted antimicrobial agents and, thus, limit antimicrobial resistance, the current main challenge in sepsis management, periodic surveillance of the causative organisms and their antimicrobial susceptibility profile may be valuable.

My research aims to identify prevalent pathogens associated with neonatal sepsis and to characterise the resistance profile of the causative organism in our population, with the ultimate goal of guiding the selection of a novel, optimal antibiotic regimen for neonates based on the susceptibility profile of the isolated pathogen. Antibiotic resistance may be lowered and the likelihood of their abuse decreased. In routine clinical practice characterization of resistance profile of the causative organisms is ignored, however, this will help us to identify the prevalent pathogens associated with neonatal sepsis it will help in a better way to manage neonatal sepsis effectively.

METHODOLOGY

This cross-sectional research was done in 2017–18 at the Neonatal Intensive Care Unit of Benazir Bhutto Hospital in Rawalpindi on 180 infants who met the following criteria and were delivered through Spontaneous Vertex Delivery or Cesarean Section between 34 and 40 weeks of gestation:

1. Maternal history of fever, >38oC, a positive C-reactive protein, or a protracted membrane rupture during the first 18 hours of birth.
2. Having two or more of the symptoms of newborn sepsis, such as poor eating, lethargy, hyponatremia (axillary temperature

less than 36.5oC), tachypnea (respiratory rate >60), tachycardia(>160), or sclerema.

3. A newborn with a positive C-reactive protein (>10 mg/dl).
4. >20,000 total leukocyte count OR 5000 total leukocyte count OR 50,000 platelets per millilitre
5. Culture positive

were enrolled in the study, neonatal patients with congenital malformations, those who had had surgical intervention for whatever reason, those who had hypoxic ischemic encephalopathy, and those who were already receiving antibiotic treatment were not included in the research. For each infant, specimens of 3 millilitres of blood were drawn and stored in an aseptic environment. The collected blood was immediately placed, by direct inoculation, into blood culture bottles containing Bactec 9240. The inoculation bottles were sent straight to the pathology laboratory at the BBH children's hospital, where they were placed in a Bactee 9240 machine, which is a continuous monitoring blood culture system. The temperature of the incubator was set to 37 degrees Celsius. After determining that the cultures included viable organisms, the samples were inoculated onto blood agar and mac conkey agar before being placed in an incubator set to a temperature of 37 degrees Celsius plus or minus three degrees for a period of 18 to 24 hours. Standard procedures, including as colony morphology, gramme stain, and other biochemical tests, are used to further identify the growth that is occurring on these agar plates as it occurs. For gramme positive bacteria, the API 20 E test, which is an analytical profile index of biochemical test for gramme negative bacteria, was carried out. Other tests, including catalase, coagulase, and Dnase, were carried out for gramme positive bacteria. The isolated organism was then subjected to the standard Kirby-Bauer method for determining its susceptibility to various antibiotics, including ampicillin, amikacin, tobramycin, cefotaxime, ceftazidime, ceftriaxone, meropenem, sulbactam, and vancomycin. These tests were performed on Meuller Hinton Agar. In accordance with CLSI standards, a panel consisting of gram-positive and gram-negative organisms was subjected to testing (Clinical laboratory standard institute).

RESULTS

Age distribution shows that 92(65.71%) were between 1-48 hours and 48(34.29%) were between 49-72 hours, mean age was calculated as 35.96+16.65 hours. Gender distribution shows that 67(47.86%) were male and 73(52.14%) were females, mean gestational age shows that 53(37.86%)were between 34-37 weeks of gestation whereas 87(62.14%) were between 38-40 weeks of gestation.

Frequency of isolated organisms shows that 27.86%(n=39) had S.Aureus, 22.14%(n=31) had E.Coli, 15%(n=21) had Staphylococcus epidermidis, 12.86%(n=18) had Klebseila and 22.14%(n=31) had others. Frequency of antimicrobial resistance of the isolated organism shows that S.Aureus was resistant to 76.92%(n=30) for ampicillin, 2.56%(n=1) for Amikacin, 5.12%(n=2) for Tobramycin, 48.71%(n=19) for Ceftazidime, 69.23%(n=27) for Ceftriaxone, 69.23%(n=27) for Meropenem, 66.67%(n=26) for Sulbactam and 25.64%(n=10) for Vanomycin whereas resistance of others organisms like E.Coli, Staphylococcus epidermidis and Klebseila

Table 1: Frequency of Antimicrobial Resistance of the Isolated Organism (n=140)

Antibiotics	Organisms			
	S.Aureus (n=39)	E.Coli (n=31)	Staphylococcus epidermidis (n=18)	Klebseila (n=31)
Ampicillin	30(76.92%)	23(74.19%)	14(77.79%)	28(90.32%)
Amikacin	1(2.56%)	1(3.22%)	4(22.22%)	11(35.48%)
Tobramycin	2(5.12%)	2(6.44%)	3(16.67%)	9(29.03%)
Cefotaxime	28(71.79%)	2(6.44%)	16(88.89%)	18(58.06%)
Ceftazidime	19(48.71%)	19(61.29%)	14(77.77%)	19(61.29%)
Ceftriaxone	27(69.23%)	18(58.06%)	11(61.11%)	14(45.16%)
Meropenem	27(69.23%)	19(61.29%)	10(55.56%)	20(64.52%)
Sulbactam	26(66.67%)	10(32.26%)	5(27.78%)	11(35.48%)
Vancomycin	10(25.64%)	8(25.81%)	4(22.22%)	9(29.03)

Table 2: Metabolic Complications by Age

Metabolic complications		Age(in hours)		P value
		1-48	49-72	
S.Aureus (n=39)	Yes	31	8	0.03
	No	61	40	
E.Coli(n=31)	Yes	21	18	0.06
	No	71	30	
Staphylococcus epidermidis (n=21)	Yes	11	10	0.16
	No	81	38	
Klebseila(n=18)	Yes	13	5	0.53
	No	79	43	
Others(n=31)	Yes	22	9	0.48
	No	70	39	

Table 3: Metabolic Complications by G.Age

Metabolic complications		G. Age(in weeks)		P value
		34-37	38-40	
S.Aureus (n=39)	Yes	1	38	0.000
	No	52	49	
E.Coli(n=31)	Yes	14	17	0.34
	No	39	70	
Staphylococcus epidermidis (n=21)	Yes	7	14	0.64
	No	46	73	
Klebseila(n=18)	Yes	6	15	0.34
	No	47	72	
Others(n=31)	Yes	11	20	0.75
	No	42	67	

Table 4: Metabolic Complications by Gender

Metabolic complications		Gender		P value
		Male	Female	
S.Aureus (n=39)	Yes	20	19	0.61
	No	47	54	
E.Coli(n=31)	Yes	17	14	0.37
	No	50	59	
Staphylococcus epidermidis (n=21)	Yes	10	11	1.16
	No	57	62	
Klebseila(n=18)	Yes	13	5	0.02
	No	54	68	
Others(n=31)	Yes	7	24	0.001
	No	60	49	

DISCUSSION

Neonatal sepsis continues to be a significant contributor to morbidity and mortality in both preterm and term infants. Despite the progress made in neonatal care, the incidence of sepsis remains a significant contributor to mortality and morbidity among neonates with a birth weight of less than 1500 g in Neonatal Intensive Care Units.[3]

It is a prevalent cause of newborn morbidity and death, which in turn leads to more longer hospital stay, a more drawn-out treatment session, and a rise in the expense of healthcare. Sepsis is thought to be caused by a number of different factors, one of which is a low birth weight in the newborn, which may be indicative of the health of the mother during pregnancy. A key contributor to the development of septicemia is having a low birth weight. The mothers did not seek prenatal care in order to learn about the state of their unborn child's health. In underdeveloped nations like Pakistan, the greatest significant risk to one's life is posed by inappropriate health care seeking behaviours. The absence of appropriate health knowledge leads to a delay in obtaining immediate treatment for newborns, which is another factor that contributes to the spread of illnesses. People do not get their loved ones evaluated in a timely manner, which leads to a further deterioration of their condition. Septicemia is often associated with poverty, which is a risk factor.[25]

The clinical manifestations of neonatal sepsis lack specificity. The clinical manifestations of the condition encompass a range of symptoms such as fever or hypothermia, respiratory distress characterised by cyanosis and apnea, feeding difficulties, lethargy or irritability, hypotonia, seizures, bulging fontanel, poor perfusion, bleeding problems, abdominal distention, hepatomegaly, gauiac-positive stools, unexplained jaundice, or a general sense of malaise or abnormal appearance. Infants who experience hypoxia-acidosis may exhibit gasping while still in the uterus, which can result in pneumonia and meconium aspiration.[9]

The goal of this research was to identify the most frequently encountered pathogens associated with neonatal sepsis and to

determine the resistance pattern of the causative organism in our population so that new, more effective antibiotic regimens could be chosen for neonates based on the susceptibility of the isolated organism. Antibiotic resistance may be lowered and the likelihood of their abuse decreased.

We compared our result with a local study revealed that Staphylococcus aureus as the commonest bacterial isolate 26.9% followed by Esherichia coli 23.1%, Staphylococcus epidermidis 13.1% and Klebseila 10%.[18] Out of the isolated organisms 70.1% of S.Aureus, 82.3% of S.Epidermidis, 63.4% of E.Coli were resistant to Cefotaxime.[19] Our findings are in agreement with this study.

Alternatively, a recent research conducted in India found that only 33.3% of septicemia patients were classified as late-onset, while 66.7% were classified as EOS (LOS). The most common Gram-negative pathogen was Klebsiellapneumoniae (35.4%), whereas the most common Gram-positive infection was Staphylococcus aureus (22.9%). These findings underscore the importance of Gram-negative germs as a leading cause of newborn sepsis.[20] Differences in population make a difference.

Bacterial resistance to commonly usedantibiotics such as ampicillin and amoxicillin in oursettings was found quite high. These antibiotics showedhigh resistance of 77%, 98.1%, 92.3% and 73.3% incases of Staph aureus, Acinetobacter, Klebseila and E.coli respectively. The study by Waseemetal[21] foundalmost similar resistance pattern being present in 83.3%, 50%, 100% and 83.3% respectively against thesebacteria.

Several regional investigations have indicated the changing causes and susceptibility to antibiotics of neonatal sepsis. In 2012, a published study revealed that Staphylococci and E.coli were the most prevalent bacteria, exhibiting intermediate sensitivity to penicillins and third generation cephalosporins. Conversely, aminoglycosides, flouroquinolones, carbapenems and vancomycin demonstrated high levels of sensitivity and low resistance.[22] In 2016, a study conducted in Peshawar revealed a prevalence of E.coli and staphalococcusaureus, which exhibited notable susceptibility to carbapenems and flouroquinolones.[23] Both of these studies demonstrated a reduced occurrence of Klebsiella and Acinetobacter, with no mention of their absence in either investigation. Another study conducted in Karachi has documented a notable prevalence of extensively resistant gram-negative bacteria that exhibit susceptibility solely to colistin.[24-25]

The variability and rapid changes in bacteriological and antimicrobial sensitivity patterns are noteworthy. The emergence of Colomycin resistance has been observed in a gradual manner, as evidenced by the report of a single organism previously. A requirement exists for meticulous and ongoing monitoring of antimicrobial sensitivity, and subsequent revision of treatment protocols in accordance with the findings. The contemporary need of the hour is the advancement of novel antimicrobial agents.

The choice of the empirical antibiotic that is used is determined by a number of parameters, including the age at which symptoms first appeared, the potential pathogens, and antibiotic susceptibility patterns, with a particular emphasis on group B Streptococcus, E coli, other Gram-negative organisms, and Listeria monocytogenes. Because the majority of neonates who are given antibiotics have negative blood cultures, clinical trials that evaluate empirical antibiotics need to take into consideration, in both their design and their analysis, the possibility that neonates with true sepsis will be pooled with non-infected neonates because there are no specific early diagnostic criteria. With the diagnostic capabilities that are now available, it is unavoidable that non-infected newborns will be included in the process of evaluating empirical antibiotics. This has the potential to lead to type II errors since the occurrence rate of clinically important outcomes would be lower in a research sample that included newborns who were healthy.[26]

The findings of our study are not final and needs to be verified through someother local studies.

CONCLUSION

Our results reveal that S.Aureus and E.Coli were the common organisms in EOS cases followed by Staphylococcus epidermidis and Klebseila whereas Ampicillin, Cefotaxime, Ceftriaxon, and Ceftazidime are also resistant in these cases.

Conflict of Interest: Nothing to declare

REFERENCES

1 Bacteriological profile and antibiotic susceptibility pattern of common isolates of neonatal sepsis, Ho Municipality, Ghana-2016. Aku FY, Akweongo P, Nyarko K, et al. Matern Health Neonatol Perinatol. 2018;4:2.

2 Global incidence and mortality of neonatal sepsis: a systematic review and meta-analysis. Fleischmann C, Reichert F, Cassini A, et al. Arch Dis Child. 2021:745–742.

3 Blood culture result profile and antimicrobial resistance pattern: a report from neonatal intensive care unit (NICU), Asella teaching and referral hospital, Asella, south East Ethiopia. Sorsa A, Früh J, Stötter L, Abdissa S. Antimicrob Resist Infect Control. 2019;8:42.

4 Sepsis kills one million newborns a year: WHO. Vogel L. CMAJ. 2017;189:0.

5 Epidemiology of neonatal sepsis and associated factors implicated: observational study at neonatal intensive care unit of Arsi University Teaching and Referral Hospital, South East Ethiopia. Sorsa A. Ethiop J Health Sci. 2019;29:333–342.

6 Sepsis: early recognition and optimized treatment. Kim HI, Park S. Tuberc Respir Dis (Seoul) 2019;82:6–14.

7 Vibrational spectroscopic analysis of blood for diagnosis of infections and sepsis: a review of requirements for a rapid diagnostic test. Confield LR, Black GP, Wilson BC, Lowe DJ, Theakstone AG, Baker MJ. Anal Methods. 2021;13:157–168.

8 Bacteriological profile and antibiotic sensitivity pattern of neonatal septicaemia in a rural tertiary care hospital in North India. Thakur S, Thakur K, Sood A, Chaudhary S. Indian J Med Microbiol. 2016;34:67–71.

9 Culture-negative early-onset neonatal sepsis — at the crossroad between efficient sepsis care and antimicrobial stewardship. Klingenberg C, Kornelisse RF, Buonocore G, Maier RF, Stocker M. Front Pediatr. 2018;6:285.

10 Neonatal sepsis in Haiti. Boulos A, Rand K, Johnson JA, Gautier J, Koster M. J Trop Pediatr. 2017;63:70–73.

11 A retrospective cohort study patient chart review of neonatal sepsis investigating responsible microorganisms and their antimicrobial susceptibility. Al-Mouqdad MM, Alaklobi FA, Aljobair FH, Alnizari TM, Taha MY, Asfour SS. J Clin Neonatol. 2018;7:141.

12 Changing trend of neonatal septicemia and antibiotic susceptibility pattern of isolates in Nepal. Thapa S, Sapkota LB. Int J Pediatr. 2019;2019:3784529.

13 Outcome of preterm babies and associated risk factors in a hospital. Shrestha S, Dangol SS, Shrestha M, Shrestha R. https://pubmed.ncbi.nlm.nih.gov/22049892/ JNMA J Nepal Med Assoc. 2010;50:286–290.

14 Half a decade of neonatal sepsis, Riyadh, Saudi Arabia. Haque KN, Chagia AH, Shaheed MM. J Trop Pediatr. 1990;36:20–23.

15 A case control study of neonatal sepsis: experience from Saudi Arabia. Dawodu A, al Umran K, Twum-Danso K. J Trop Pediatr. 1997;43:84–88.

16 Early and late onset neonatal sepsis in very low birth weight infants in a tertiary center in Saudi Arabia. Sobaih BH, Al-Mandeel H. J Neonatal Biol. 2014:3–5.

17 Characteristics of neonatal sepsis at a tertiary care hospital in Saudi Arabia. Al-Matary A, Heena H, AlSarheed AS, et al. J Infect Public Health. 2019;12:666–672.

18 El-Sheikh H, Gaafar M, Yosri M, Hassan DM, Said H. Study of bacteria causing septicemia in neonatal intensive care unit. The Egyptian Journal of Medical Microbiology (EJMM) 2016;27:25.

19 Muhammad Z, Ahmed A, Hatat U, Wazir MS, Rafiyatullah WH. Neonatal sepsis: causative bacteria and their resistance to antibiotics. J Ayub Med Coll Abbottabad 2010;22:33-6.

20 Muley VA, Ghadage DP, Bhore AV. Bacteriological profile of neonatal septicemia in a tertiary care hospital from Western India. Journal of Global Infectious Disease 2015;7:75-7.

21 Waseem R, Khan M, IzharTS. Neonatal sepsis. ProfessionalMed J 2005;12:451–6.

22 Najeeb S, Gillani S, Rizvi SK, Ullah R, Rehman AR. Causative bacteria and antibiotic resistance in neonatal sepsis.J Ayub Med Coll Abbottabad 2012; 24(3-4):131-4.

23 Ullah O, Khan A, Ambreen A, Ahmad I, Akhtar T, Gandapor AJ, et al. Antibiotic Sensitivity pattern of Bacterial Isolates of Neonatal Septicemia in Peshawar, Pakistan.Arch Iran Med 2016; 19(12):866-9.

24 Ambreen G, Salat MS, HussainK,Raza SS, Ali U, Azam I, et al. Efficacy of colistin in multidrug-resistant neonatal sepsis: Experience from a tertiary care center in Karachi, Pakistan. Arch Dis Child 2020; 105(9): 830-6.

25 Bhatti ZI, Nawaz K, Mahmood R, Bhatti SZ. Indicators of Neonatal Sepsis in Pakistan. P J M H S 2021;15:629-30

26 Korang SK, Safi S, Nava C, Gordon A, Gupta M, Greisen G, Lausten-Thomsen U, Jakobsen JC. Antibiotic regimens for early-onset neonatal sepsis. Cochrane Database Syst Rev. 2021 May 17;5(5):CD013837.