The Research of Medical Science Review
ISSN: 3007-1208 & 3007-1216 Volume 3, Issue 3, 2025

FREQUENCY OF CULTURE PROVEN SEPSIS AND COMMON ISOLATES AMONG NEONATES ADMITTED IN NICU OF TERTIARY CARE HOSPITAL

Dr. Rameeha Rauf[*1], Dr. Shehla Choudhry[2]

[*1]*Resident FCPS Pediatrics, Shifa International hospital Islamabad*
[2]*FCPS, Consultant Pediatrician, Shifa International hospital Islamabad*

[*1]raza.data01@gmail.com, [2]rameeharauf@gmail.com

DOI: https://doi.org/10.5281/zenodo.15113253

Keywords
Culture positive sepsis, neonates, microbial spectrum.

Article History
Received on 22 February 2025
Accepted on 22 March 2025
Published on 31 March 2025

Copyright @Author

Corresponding Author: *

Abstract

Objective:

To determine the frequency of culture proven sepsis (CPS) and to investigate the occurrence of various bacterial pathogens that lead to neonatal sepsis.

Methods:

This study is a descriptive cross-sectional analysis that focused on 124 neonates, ranging from birth to 28 days old, who were suspected to have neonatal sepsis. Samples of body fluids were sent to the hospital microbiology laboratory for determination of CPS and microbial spectrum.

Results:

The average age of the neonates was 6.5±3.6 days. Among the participants, there were 69 males (55.6%) and 55 females (44.4%). CPS was identified in 18 (14.5%) neonates. Regarding microbial spectrum, Acinetobacter baumannii was the most prevalent microorganism, found in 6 cases, accounting for 33.3% of the isolates. Following this, Klebsiella species were identified in 3 cases, representing 16.7%. Both Staphylococcus aureus (including MRSA) and Pseudomonas aeruginosa were each found in 2 cases, contributing to 11.1% of the total isolates. Escherichia coli, methicillin-resistant Staphylococcus epidermidis (MRSE), and Streptococcus pneumoniae were each isolated in 1 case, with each microorganism representing 5.5% of the frequency distribution.

Conclusion:

The frequency of culture-positive sepsis in the present study was 14.5%. Acinetobacter baumannii and Klebsiella species are common microbes causing neonatal sepsis.

INTRODUCTION

Neonatal sepsis is a serious medical condition that primarily occurs within the first 28 days of life, a period typically known as the neonatal stage. It is considered one of the major causes of illness and death among newborns globally (Nyenga et al., 2021). This critical condition emerges when the immune system responds aggressively to infections that may be caused by bacteria, viruses, or fungi. The symptoms of neonatal sepsis can vary significantly, ranging from nonspecific signs like fever, lethargy, poor appetite, respiratory difficulties, and low body temperature to more severe conditions such as septicemia, pneumonia, bone infections, and meningitis (Guo et al., 2019; Pandit and Vyas, 2020). Neonatal sepsis can

be classified into two main types: early-onset sepsis, which arises within the first 72 hours of birth and is often linked to maternal health issues during pregnancy or difficulties during labor, and late-onset sepsis, which manifests after the initial 72 hours and is often associated with exposure to pathogens in hospital or community settings. Early onset sepsis (EOS) primarily arises from bacterial infections that the newborn acquires either during pregnancy or at the time of birth through the maternal reproductive system. In contrast, late onset sepsis is linked to infections obtained in the environment, either at home or in healthcare settings, after the baby has been born (Flannery and Puopolo, 2022). In Pakistan, sepsis plays a critical role in the country's alarmingly high neonatal mortality rate, accounting for 17.2% of all deaths among newborns (Strunk et al., 2024).

Identifying sepsis in newborns can be quite difficult, primarily due to the often vague and nonspecific nature of clinical signs. Sepsis is diagnosed either through clinical assessment or by laboratory tests, with a definitive diagnosis relying on positive cultures from blood or other bodily fluids. Even with improvements in diagnostic methods, blood culture remains the cornerstone of the investigative protocols (Popescu et al., 2020; Wen et al., 2021).

The range of organisms responsible for sepsis is not only different across various geographic locations but also shifts over time, even within a single area. This variability is largely due to evolving antibiotic usage patterns and alterations in lifestyle factors. Additionally, the rates of sepsis can differ significantly between neonatal intensive care units (NICUs) in different hospitals, influenced by the specific conditions that may make infants more susceptible to infections (Aku et al., 2018). This research aims to determine the frequency of culture proven sepsis (CPS) and to investigate the occurrence of various bacterial pathogens that lead to neonatal sepsis. By identifying the most common organisms involved in these cases, neonatologists will be better equipped to start appropriate antibiotic therapies while waiting for the confirmation of blood culture results.

METHODS:

This study is a descriptive cross-sectional analysis that focused on 124 neonates, ranging from birth to 28 days old, who were suspected to have neonatal sepsis. Infants with dysmorphic features, chromosomal abnormalities, or congenital malformations were not included in the study population. A neonate was classified as having suspected sepsis if they exhibited any of the following clinical signs: a body temperature exceeding 38.5 °C or falling below 36 °C, a heart rate above 160 beats per minute or below 100 beats per minute, a respiratory rate greater than 60 breaths per minute, white blood cell counts exceeding 20000 cells/µL or dropping below 5000 cells/µL, or having more than 10% of immature neutrophils. Additional symptoms indicative of possible infection included lethargy, refusal to feed, difficulty breathing, altered consciousness, or seizures. Guardians were offered a detailed study objective explanation before obtaining written consent.

A sterile, single-use syringe with a capacity of 1cc was utilized to collect blood samples, ranging from 0.5 to 1 ml, from neonates while adhering to strict aseptic protocols. After collection, the blood was carefully transferred to appropriate culture bottles using aseptic techniques. These samples were then analyzed using automated blood culture methods, specifically BACTEC and BACTALERT systems.

Urine samples were obtained through catheterization, also under aseptic conditions, just prior to collection. The samples were then transferred to sterile containers, facilitating safe transport to the laboratory for testing.

In a similar manner, cerebrospinal fluid (CSF) was collected using sterile methods and placed in specially designated sterile containers to ensure proper transportation to the laboratory for further analysis. Patients who showed bacterial growth in these samples were classified as having sepsis confirmed by culture.

In microbiology laboratories, the identification of microorganisms was carried out with great precision following established protocols. Initially, Gram staining was performed to examine their microscopic characteristics. The isolates were then categorized based on their unique appearances on specific growth media, further confirmed through biochemical reactions employing standardized methods. This systematic approach guarantees both the accuracy and reliability of the identification process. When identifying members of the Enterobacteriaceae family, a wide array of tests was conducted. This includes

assessing indole production, hydrogen sulfide (H2S) production, citrate utilization, motility, urease activity, oxidase activity, and various carbohydrate utilization tests, among others. Such a comprehensive testing strategy ensures that identifications are thorough and precise. For gram-positive bacteria, a diverse set of tests was used to discern their specific traits. Tests such as coagulase, catalase, bacitracin susceptibility, and optochin susceptibility are meticulously chosen for their effectiveness in identifying these microorganisms. Additionally, blood culture broths that do not exhibit microbial growth within a week were deemed culture negative, leading to the exclusion of those patients from further analysis. Only patients with confirmed positive cultures were retained for study.

The data analysis was carried out using SPSS version 23. Neonatal age was expressed by calculating the mean and standard deviation. Other factors such as gender, pre-term delivery, geographic location, socio-economic background, educational attainment, culture-confirmed sepsis, and the bacteriological spectrum were summarized through the use of frequencies and percentages.

RESULTS:

The baseline characteristics of the neonates in the study, which included 124 subjects are presented in Table 1. The average age of the neonates was 6.5±3.6 days. Among the participants, there were 69 males (55.6%) and 55 females (44.4%). In terms of birth status, 67 neonates (54.0%) were pre-term, while 57 (46.0%) were not. The majority of the neonates resided in rural areas, accounting for 83 participants (66.9%), compared to 41 (33.1%) from urban areas. Educational status of the mothers varied, with 21 mothers (16.9%) being illiterate, 57 (45.9%) having a middle-class education, 28 (22.5%) reaching secondary education, and 18 (14.5%) achieving higher secondary education. Socioeconomic status of the parents also indicated a predominance of middle-class families, with 86 (69.4%) in this category, while 17 (13.7%) were classified as poor and 21 (16.9%) as upper class. Regarding gestational age at birth, 1 neonate (0.8%) was born before 28 weeks, 3 (2.4%) between 28-32 weeks, 63 (50.8%) between 32-37 weeks, and 57 (46.0%) were born after 37 weeks. The source of culture tested predominantly came from blood samples, with 113 cases (91.1%), followed by urine samples from 10 (8.1%) and cerebrospinal fluid from 1 (0.8%). Lastly, CPS was identified in 18 neonates (14.5%), while 106 (85.5%) did not have CPS (Table 1).

Table 2 presents the frequency of common isolates identified. Acinetobacter baumannii was the most prevalent microorganism, found in 6 cases, accounting for 33.3% of the isolates. Following this, Klebsiella species were identified in 3 cases, representing 16.7%. Both Staphylococcus aureus (including MRSA) and Pseudomonas aeruginosa were each found in 2 cases, contributing to 11.1% of the total isolates. Candida species, which also includes Candida albicans, were present in 2 cases as well, also making up 11.1%. Escherichia coli, methicillin-resistant Staphylococcus epidermidis (MRSE), and Streptococcus pneumoniae were each isolated in 1 case, with each microorganism representing 5.5% of the frequency distribution (Table 2).

Table 1. Baseline Characteristics of Neonates (N=124).

Variable	Variables
Age (Days)	6.5±3.6
Gender (%)	
Male	69 (55.6%)
Female	55 (44.4%)
Pre-term Birth (%)	
Yes	67 (54.0%)
No	57 (46.0%)
Area of Residence (%)	
Rural	83 (66.9%)
Urban	41 (33.1%)
Mother's Education Status (%)	
Illiterate	21 (16.9%)
Middle Class	57 (45.9%)
Secondary	28 (22.5%)
Higher Secondary	18 (14.5%)
Parents Socioeconomic status (%)	
Poor	17 (13.7%)
Middle Class	86 (69.4%)
Upper Class	21 (16.9%)
Gestational Age at birth (%)	
<28 weeks	01 (0.8%)
28-32 weeks	03 (2.4%)
32-37 weeks	63 (50.8%)
>37 weeks	57 (46.0%)
Source of Culture (%)	
Blood	113 (91.1%)
Urine	10 (8.1%)
Cerebrospinal Fluid	01 (0.8%)
Culture Positive Sepsis (%)	
Yes	18 (14.5%)
No	106 (85.5%)

Table 2. Frequency of Common Isolates in Neonates (N=18).

Microorganism	Frequency
Acinetobacter baumannii	06 (33.3%)
Klebsiella species	03 (16.7%)
Staphylococcus aureus (including MRSA)	02 (11.1%)
Pseudomonas aeruginosa	02 (11.1%)
Candida spp. (including candida albicans)	02 (11.1%)
Escherichia coli	01 (5.5%)
Methicillin-resistant Staphylococcus epidermidis (MRSE)	01 (5.5%)
Streptococcus pneumoniae	01 (5.5%)

DISCUSSION:

Neonatal sepsis poses a significant threat to infants, particularly in Neonatal Intensive Care Units (NICUs), and is a leading contributor to neonatal mortality. The challenge is intensifying due to the growing issue of antibiotic resistance (Liu et al., 2019). Early detection of the specific microbial agents responsible for infection and the swift initiation of the correct antibiotic treatment are essential for improving patient outcomes (Toan et al., 2018). Although blood cultures are still considered the primary method for identifying these pathogens, they come with certain drawbacks. The blood culture process can be time-consuming, often experiences low sensitivity, and carries a risk of contamination during collection. Furthermore, the rate of positive blood cultures can vary greatly among different studies, which is influenced by factors such as the methodologies used, study designs, and the characteristics of the patient groups involved. Reports have shown that the rates of blood culture isolation can fluctuate between 6.7% and 55.4% (Jatsho et al., 2020). In our research, we observed a blood culture positivity rate of 14.5%, which is consistent with similar rates reported by Gupta and Kashyap, as well as Ansari et al. (Thakur et al., 2016; Ansari et al., 2015). The relatively low positivity rates found in numerous studies may stem from various reasons. These include the prior use of antibiotics, challenges in sample collection, and the presence of non-bacterial pathogens like fungi, viruses, or anaerobic organisms. These complexities underscore the urgent need for advancements in diagnostic techniques and the establishment of standardized practices to improve the accuracy and consistency of diagnosing neonatal sepsis (Bansal et al., 2004).

Ehsan et al. conducted a study on frequency of culture proven sepsis and on bacteriological spectrum in neonates admitted with sepsis. The authors reported culture proven sepsis in 13.3% of cases. Regarding bacteriological spectrum, Escherichia coli was reported in 1 (3.1%) patient, Burkholderia cepacia in 3 (9.3%) patients, Serratia marcescens in 3 (9.3%) patients, Klebsiella pneumoniae in 2 (6.2%) patients, Pseudomonas aeruginosa 6 (18.7%) patients, Acinetobacter baumannii 12 (37.5%) patients, Staphylococcus aureus 1 (3.1%) patients and streptococcus pneumoniae in 4 (12.5%) patients (Ehsan and Marium, 2023).

Mohammad et al. in another study, reported culture proven sepsis in 31.7% patients. Regarding bacteriological spectrum, they reported Klebsiella species in 34 (45.3%) patients, Enterobacter in 3 (4%) patients, Pseudomonas 4 (5.3%) patients, Acinetobacter in 8 (10.7%) patients, coagulase-negative staphylococci (CoNS) in 17 (22.7%) patients, methicillin-resistant Staphylococcus aureus (MRSA) in 7 (9.3%) patients and Streptococci in 2 (2.7%) patients (Almohammady et al., 2020).

Our research offers crucial insights into the frequency and microbial diversity associated with neonatal sepsis, a significant public health concern worldwide. Investigating neonatal sepsis represents a novel initiative in this region, highlighting the necessity to evaluate the severity of the issue on a national level. We anticipate that our findings will encourage additional studies in this area, ultimately assisting policymakers in developing effective prevention strategies and providing clinicians with a clearer understanding of the disease's impact in Pakistan.

CONCLUSION:

The frequency of culture-positive sepsis in the present study was 14.5%. Acinetobacter baumannii and Klebsiella species are common microbes causing neonatal sepsis.

REFERENCES

Aku, F. Y., Akweongo, P., Nyarko, K., Sackey, S., Wurapa, F., Afari, E. A., Ameme, D. K.and Kenu, E. 2018. Bacteriological profile and antibiotic susceptibility pattern of common isolates of neonatal sepsis, Ho Municipality, Ghana-2016. *Maternal health, neonatology and perinatology,* **42**.

Almohammady, M. N., Eltahlawy, E. M.and Reda, N. M. 2020. Pattern of bacterial profile and antibiotic susceptibility among neonatal sepsis cases at Cairo University Children Hospital. *J Taibah Un Med Sci,* **15**(1): 39-47.

Ansari, S., Nepal, H. P., Gautam, R., Shrestha, S., Neopane, P.and Chapagain, M. L. 2015. Neonatal Septicemia in Nepal: Early-Onset versus Late-Onset. *Int J Pediatr.,* **2015**379806.

Bansal, S., Jain, A., Agarwal, J.and Malik, G. K. 2004. Significance of coagulase negative staphylococci in neonates with late onset septicemia. *Indian J Pathol Microbiol.,* **47**(4): 586-8.

Ehsan, S.and Marium, R. J. P. J. O. H. S. 2023. Bacteriological Profile and Antibiotic Susceptibility Pattern of Isolates in Neonatal Sepsis: Antibiotic Susceptibility Pattern of Neonatal Sepsis. 44-49.

Flannery, D. D.and Puopolo, K. M. 2022. Neonatal Early-Onset Sepsis. *NeoReviews,* **23**(11): 756-770.

Guo, J., Luo, Y., Wu, Y., Lai, W.and Mu, X. 2019. Clinical Characteristic and Pathogen Spectrum of Neonatal Sepsis in Guangzhou City from June 2011 to June 2017. *Medical science monitor : international medical journal of experimental and clinical research,* **25**2296-2304.

Jatsho, J., Nishizawa, Y., Pelzom, D.and Sharma, R. 2020. Clinical and Bacteriological Profile of Neonatal Sepsis: A Prospective Hospital-Based Study. *Int J Pediatr.,* **2020:**1835945.

Liu, C., Yoon, E. J., Kim, D., Shin, J. H., Shin, J. H., Shin, K. S., Kim, Y. A., Uh, Y., Kim, H. S., Kim, Y. R.and Jeong, S. H. 2019. Antimicrobial resistance in South Korea: A report from the Korean global antimicrobial resistance surveillance system (Kor-GLASS) for 2017. *Journal of infection and chemotherapy : official journal of the Japan Society of Chemotherapy,* **25**(11): 845-859.

Nyenga, A. M., Mukuku, O., Wembonyama, S. O. J. T.and Pediatrics, C. P. I. 2021. Neonatal sepsis: A review of the literature. 394-101.

Pandit, B. R.and Vyas, A. 2020. Clinical Symptoms, Pathogen Spectrum, Risk Factors and Antibiogram of Suspected Neonatal Sepsis Cases in Tertiary Care Hospital of Southern Part of Nepal: A Descriptive Cross-sectional Study. *J Nepal Med Assoc,* **58**(232): 976-982.

Popescu, C. R., Cavanagh, M. M. M., Tembo, B., Chiume, M., Lufesi, N., Goldfarb, D. M., Kissoon, N.and Lavoie, P. M. 2020. Neonatal sepsis in low-income countries: epidemiology, diagnosis and prevention. *Expert Rev Anti-infective Therapy,* **18**(5): 443-452.

Strunk, T., Molloy, E. J., Mishra, A.and Bhutta, Z. A. 2024. Neonatal bacterial sepsis. *Lancet (London, England),* **404**(10449): 277-293.

Thakur, S., Thakur, K., Sood, A.and Chaudhary, S. 2016. Bacteriological profile and antibiotic sensitivity pattern of neonatal septicaemia in a rural tertiary care hospital in North India. *Indian J Med Microbiol.,* **34**(1): 67-71.

Toan, N. D., Darton, T. C., Boinett, C. J., Campbell, J. I., Karkey, A., Kestelyn, E., Thinh, L. Q., Mau, N. K., Thanh Tam, P. T., Nhan, L. N. T., Quang Minh, N. N., Phuong, C. N., Hung, N. T., Xuan, N. M., Thuong, T. C.and Baker, S. 2018. Clinical features, antimicrobial susceptibility patterns and genomics of bacteria causing neonatal sepsis in a children's hospital in Vietnam: protocol for a prospective observational study. *BMJ Open,* **8**(1): e019611.

Wen, S. C. H., Ezure, Y., Rolley, L., Spurling, G., Lau, C. L., Riaz, S., Paterson, D. L.and Irwin, A. D. 2021. Gram-negative neonatal sepsis in low- and lower-middle-income countries and WHO empirical antibiotic recommendations: A systematic review and meta-analysis. *PLoS Med.,* **18**(9): e1003787.