



## “Bacteriological Profile and Antimicrobial Sensitivity Pattern of Neonatal Sepsis”

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**Abstract: Background:** Neonatal sepsis is one among the leading cause for neonatal mortality rate and morbidity in neonatal ward and neonatal medical care unit especially in developing countries. Frequent monitoring on pathogens with recent updates and their antimicrobial sensitivity pattern is mandatory for the higher management.

**Objective:** This study was designed to evaluate the Bacteriological Profile and Antimicrobial Sensitivity Pattern of Neonatal Sepsis. **Methods:** This prospective observational study was under taken in the neonatal unit, department of pediatrics, Faridpur Medical College Hospital, Faridpur, Bangladesh from March 2020 to March 2021. Blood samples from 90 clinically suspected neonatal sepsis were collected and processed in the microbiological protocol and their antimicrobial sensitivity pattern was determined. **Results:** Of 90 cases, 61(67.78%) showed positive blood culture. Gram positive isolates were 48(78.79%) and Gram negative isolates were 23(37.71%). Staphylococcus aureus was the most common organism 75.51% followed by E.Coli 27.87%, Klebsiella pneumoniae 9.84% and Streptococcus pneumoniae 3.28% among the isolates. All the three common isolates showed 100% resistance to Ampicillin. Gram positive isolates were sensitive to Amikacin and Amoxicillin & Clavulenic acid whereas Gram negative isolates were sensitive to Gentamicin and Meropenem. **Conclusion:** Neonatal septicemia was found to be 67.78% during this study supported blood culture because the gold standard investigation for diagnosis. This result also showed that there was an increasing rate of antibiotic resistance to the commonly used antibiotics. These two changes in microbial spectrum and antimicrobial susceptibility pattern as noticed during this study will definitely help in treating such cases with appropriate antibiotic and thereby help to decrease neonatal morbidity and mortality.

**Keywords:** Neonatal sepsis, Coagulase negative bacteria, Antimicrobial susceptibility.

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### INTRODUCTION

Neonatal sepsis may be a clinical syndrome characterized by systemic signs of circulatory compromise caused by invasion of the blood stream by bacteria within the first four weeks of life and is more common in compared with developed countries [1]. In developing countries, unsafe birthing practices have critical role to cause neonatal infections. It is estimated that neonatal septicemia is liable for approximately 25% of the neonatal

deaths within the world and mostly in developing countries. Neonatal mortality rate (NMR) is 27/1000 live birth and neonatal sepsis contributes 36% of total death in Bangladesh [2, 3]. Some maternal risk factors may be associated with neonatal sepsis like premature rupture of membrane, maternal fever within 2 weeks prior to delivery, foul smelling liquor or instrumental delivery and poor maternal nutrition. The foetal risk factors are low birth weight, birth asphyxia and congenital anomalies.

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Neonatal sepsis has been classified as either early onset sepsis (EOS) meaning sepsis occur within the first 72 hours of life is defined as early onset sepsis and late onset sepsis (LOS) meaning sepsis that occurring beyond 72 hours defined as late onset neonatal sepsis. Early onset sepsis is thanks to vertical transmission during labour or birth .It includes bacteremia and / or sepsis, meningitis and pneumonia. Late onset sepsis is due to vertical, horizontal or nosocomial infection. It's most clinical manifestations are: meningitis (30- 40%), bacteremia (40%), septic arthritis (5 -10%), and rarely omphalitis. The reported incidence of neonatal sepsis varies from 7 to 38 per 1000 live birth in ASIA [4]. Microorganisms responsible for neonatal sepsis developed resistance to antibiotics over last few decades and thus making treatment extremely difficult. Hence this study was under taken to determine the bacteriological profile of neonatal septicemia and their antibiotic susceptibility pattern. This will help in formulating the use of antibiotics locally in order to decide about the best possible use antibiotics for the treatment of neonatal sepsis. The microorganism most commonly associated with early onset sepsis include Group -B streptococcus (GBS), Escherichia coli, Coagulase negative staphylococcus species (CONS), Hemophilus influenza, and Listeria monocytogen and late onset septicemia is caused by CONS, S. aureus, E.coli, Klebsiella, Pseudomonas, Enterobactor, Candida, GBS. The recent trends show an increase infections due to CONS. Increased rate of extended spectrum beta lactamase (ESBL) and methicillin resistance staphylococcus aureus and multi drugs resistant (MDSR) straining may be an explanation for concern in neonatal medical care unit and neonatal ward worldwide [5]. An early and accurate Aetiological diagnosis isn't always easy. Because the disease may start with minimal or nonspecific symptoms, it can delay start of treatment until clinical signs and symptoms of sepsis have established which can increase neonatal mortality rate. On the opposite hand empirical antibiotic therapy may help within the treatment of sepsis. The first and appropriate initiation of antimicrobial agents in high risk neonates before the results of blood culture susceptibility is defined as "empirical antibiotic therapy". Although isolation of a pathogen in culture may be a prerequisite, proven bacterial sepsis, culture results take a minimum of 48 to 72 hours to be reported. Numerous more babies are treated in sepsis with antibiotic than the amount that even has the condition. Though various studies in bacteriological profile of neonatal sepsis have already been wiped out different hospital like Dhaka Shishu Hospital and BIRDEM hospital, it's important to recollect that bacterial flora is dynamic, different from one place to a

different and it changes within the same place over a period of your time [2]. So, it is important to conduct periodic surveillance to access the changing pattern of organisms causing neonatal sepsis. Therefore knowledge of the pattern of bacterial isolates and their antimicrobial susceptibility pattern is useful for prompt treatment of patient. As there was no such type of study in FMCH within the past, this study was designed in the neonatal intensive care unit of FMCH to evaluate the common organisms associated with neonatal sepsis. This prospective observational study was performed to work out the bacteriological profile of neonatal sepsis and to research antimicrobial susceptibility pattern of the isolates which can help the physicians to settle on the foremost appropriate antibiotic therapy.

## MATERIAL AND METHODS

This prospective observational study was under taken in the neonatal unit, department of pediatrics, Faridpur Medical College Hospital, Faridpur, Bangladesh from March 2020 to March 2021. Blood samples from 90 clinically suspected neonatal sepsis were collected and processed in the microbiological protocol and their antimicrobial sensitivity pattern was determined.

### Inclusion and Exclusion Criteria

Babies born in the hospital and admission from outside were included. For purpose of all this study, neonatal sepsis was defined a neonates presenting with one or more features like reluctant to feed, lethargy, presence of fever ( $>38^{\circ}\text{C}$ ), or hypothermia ( $<36^{\circ}\text{C}$ ), abdominal distension, vomiting, diarrhea, jaundice, convulsion, bulging fontanel, respiratory distress, hypoglycemia, umbilical discharge were considered as the main clinical features of neonatal sepsis. Mother of those neonates who delivered by either cesarean section or vaginal delivery with history of high risk delivery like premature rupture of membrane  $>12$  hours, prolonged labour, foul smelling liquor, meconium stained amniotic fluid were also included in this study. Suspected neonates who died upon arrival before blood culture report could be obtained or neonates had clinical picture of sepsis without positive blood culture were excluded. Neonates with congenital anomalies, extreme low birth weight babies were also excluded. A thorough physical examination was carried out after taking detailed and careful history of each case. Patient's care givers were interviewed and only those babies were enrolled where the guardians gave permission for collection of samples.

### Sample Size Calculation

Out of 324 neonates 90 were selected who were admitted in the Neonatal intensive care

unit (NICU) and neonatal ward during the time of study.

### DATA COLLECTION

Data on the pathogens isolate and their antibiotic susceptibility pattern were collected after laboratory test was done. Data was collected for the subsequent variables: demographic profile, blood culture result and antibiotic sensitivity pattern.

#### Laboratory Investigations

As a sample 1- 2 ml of blood was aseptically obtained from the peripheral vein of every neonate and dispensed into a sterilized universal bottle containing 20 ml of Tryptone Soy Broth (TSB) and Brain Heart infusion Broth to form a 1:10 dilution. The location was cleaned with povidine and chlorhexidane before blood was collected. Blood culture samples were then transported to the laboratory and incubated at 37 c for 24 hours using thermo scientific detection system. After detection of presence of organism, subculture was done on Solid media (blood agar, Mc Conkey agar and chocolate agar media) on appearance of turbidity. On day 1, 2, 3 and 7. Organisms were identified according to the standard microbial procedures including gram stain, colony morphology, motility, and biochemical reactions. All the records of this study were carefully revised and data including sex, birth weight, preterm or term and Clinical features consistent with sepsis results of culture antibiotic sensitivity were recorded into a data collection sheet. Cerebro spinal fluid was collected by lumbar puncture only when meningitis was a suspicion. Antibiotic susceptibility testing was performed for all blood culture isolates, according to the criteria of the National Laboratory Standard by disk diffusion method.

### DATA ANALYSES

Data collected was checked for completeness and double entry was done. Data was entered into Microsoft power point and cleaned by cross checking for missing data, duplicates and outliers pathogens isolated from the laboratory investigation and antibiotic susceptibility of the

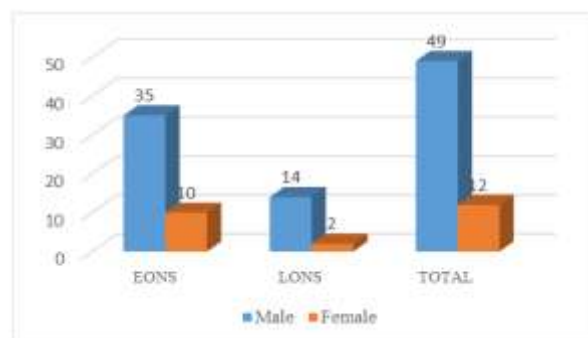
isolates was presented in frequencies and proportions.

### RESULTS

During the study period 90 samples were collected from the neonates admitted in NICU and neonatal ward. Among them blood culture positive were 61 (67.78%) and culture negative were 29 (32.22%). Among culture positive cases, early onset sepsis was 41 (80.33 %), male 49. The medium gestational week was 35 weeks (34-38) weeks and medium birth weight was 2kg (1.5-2.5) kg. The commonest risk factors were mother had premature rupture of membrane and had history of prolong labour. More common organisms were gram positive bacteria. Staphylococcus aureus were the most isolated prevalent gm positive bacteria (75.51%). Death was low in this study. In this study out of 90 cases of suspected neonates was culture positive sepsis which was 67.78%. We also found that pre term babies (80.33%) and low birth weight babies (72.13%) are more susceptible to infection. The majority of the study population was poor and delivered at home, largely in the hands of untrained birth attendants. In this study, staphylococcus aureus was the commonest isolate, accounting about 75.51% of neonatal septicemia. The second commonest organism was E.coli which was seen in 17 neonates. That was 27.87%.

**Table-1: Sex distribution of patients.**

Sex	Number			Percentage
	EONS	LONS	TOTAL	
Male	35	14	49	80.33%
Female	10	2	12	19.67%



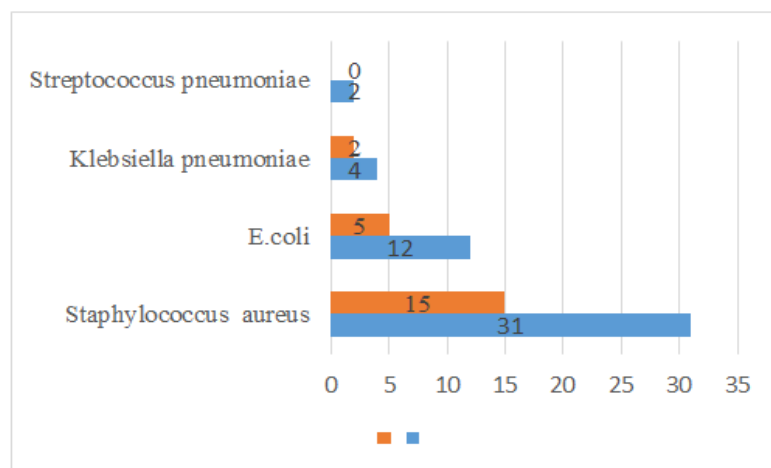
**Fig-1: Sex distribution of patients**

**Table-2: Demographic characteristics of the study population**

Blood culture findings	Number	Percentage
Culture positive	61	67.78%
Culture negative	29	32.22%
Total	90	100%
Name of organism		
Staphylococcus	46	75.51%
Echerichia Coli	17	27.87%
Klebsiella	06	9.87%
Streptococcus	02	3.28%
Parameters: Age		
0-7 days	46 (61)	80.33%
8-1month	12 (29)	19.67%
Weight		
1.5k g - 2.4 kg	44(61)	72.13%
2.5 kg - 3.9 kg	17(61)	27.87%
Gestational age		
<35 week	36(61)	59.01%
36 - 38 week	25(61)	41%
Mode of delivery		
Normal vaginal delivery	34	55.74%
Cesarean section	27	44.26%
Place of delivery		
In born	35	57.38%
Out born	26	42.62%

**Table-3: Types of organism isolates.**

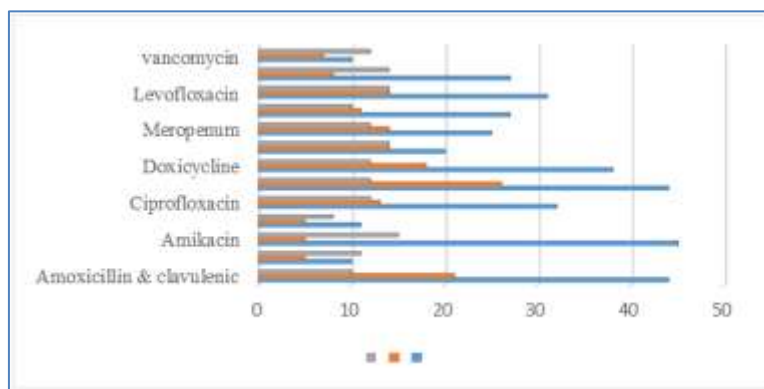
Name	Number		Total
	EONS	LONS	
Staphylococcus aureus	31	15	75.51%
E.coli	12	5	27.87%
Klebsiella pneumoniae	4	2	09.84%
Streptococcus pneumoniae	2	0	3.28%



**Fig-2: Types of organism isolates**

**Table-4: Antibiotic susceptibility Pattern**

Name of antibiotic	Isolate organism		
	Stephylococcus	E.coli	Klebsiella
Amoxicillin & clavulenic	44	21	10
Azithromycin	10	5	11
Amikacin	45	5	15
Ceftriaxone	11	5	8
Ciprofloxacin	32	13	12
Colistin	44	26	12
Doxycycline	38	18	12
gentamicin	20	14	14
Meropenem	25	14	12
Imipenem	27	11	10
Levofloxacin	31	14	14
Linezolid	27	8	14
vancomycin	10	7	12



**Fig-3: Antibiotic susceptibility Pattern**

Klebsiella was 9.84% and streptococcus also seen in about 3.28% cases. In staphylococcus, common sensitive drug were Amikacin, Amoxicillin & Clavulanic acid, Colistin, Doxycycline, Vancomycin, Ciprofloxacin, Levofloxacin, linezolid, which were 45%, 44%, 41%, 38%, 35%, 32%, 31%, 27% respectively. In E.coli most common sensitivity were Gentamicin, Meropenem, Levofloxacin, Linezolid, Ciprofloxacin, Colistin which were 16%, 14%, 14%, 14%, 12%, 12% respectively. In Klebsiella pneumoniae most common sensitive drugs were Meropenem, Piperacillin in preterm and low birth weight babies.

**DISCUSSION**

Sepsis is an unwanted health problem especially among low birth weight neonates and one among the leading explanation for neonatal death. Diagnosis of neonatal sepsis is one among the foremost difficult tasks for physicians because there's no single reliable test for early diagnosis of sepsis. Currently blood culture is that the most reliable method for detection of bacterial infections. A rapid laboratory test with high specificity for neonatal sepsis help in making a therapeutic decision and avoiding the

unnecessary future use of antibiotics in patients with clinical signs and symptoms of sepsis but negative blood cultures. In this study, we found that out of 90 clinical neonatal septicemic case, 67.78 % (n = 61) had growth on blood culture. In our study, among the infected newborn, male were predominant, 80.33% (n= 61). Similar observations are made by others; Bhat et al. (90.8%) and Shrestha et al. (60.64%) [6,7] The low incidence of gram positive sepsis in present study are often attributed to low infection rates with CoNS which is typically related to central lines and rarity of B Streptococcus infection in India [8]. It's thanks to the factors regulating the synthesis of globulin situated on the x- chromosome. Male has just one x- chromosome. So, he's immunologically less protected than female. We also found that pre term babies (80.33%) and low birth weight babies (72.13%) are more susceptible to infection. The majority of the study population was poor and delivered reception, largely within the hands of untrained birth attendants. Home deliveries are common in Bangladesh and these deliveries also are significantly related to birth asphyxia, which was highly prevalent in our study population and which successively, is associated with an increased

risk of great neonatal infection. In this study, staphylococcus aureus was the commonest isolate, accounting about 75.51% of neonatal septicemia. Whereas, many studies reported Klebsiella to be the most common organism [7, 9-12]. Some authors had found Staphylococcus aureus as the most common organism [13, 14]. Intrapartum antibiotic prophylaxis has also led to a substantial change in the bacterial flora responsible for early onset neonatal sepsis. The second commonest organism was E.coli which was seen in 17 neonates. That was 27.87%. Klebsiella was 9.84% and streptococcus also seen in about 3.28% cases. In staphylococcus, common sensitive drug were Amikacin, Amoxicillin & Clavulanic acid, Colistin, Doxycycline, Vancomycin, Ciprofloxacin, Levofloxacin, linezolid, which were 45%, 44%, 41%, 38%, 35%, 32%, 31%, 27% respectively. In E.coli most common sensitivity were Gentamicin, Meropenem, Levofloxacin, Linezolid, Ciprofloxacin, Colistin which were 16%, 14%, 14%, 14%, 12%, 12% respectively. The greater prevalence of resistance to commonly used antibiotics has also been reported in recent studies [15, 16]. In Klebsiella pneumoniae most common sensitive drugs were Meropenem, Piperacillin in preterm and low birth weight babies. These might be due to low level of IgG and lower defense mechanism. The alarming finding in our study were the development of multi drug resistance pathogens to commonly used antibiotics in our neonatal ward and neonatal intensive care unit. In this study most of the strains showed a low sensitivity to Azithromycin, Cefixime, and Ceftazidime. When compared with other studies, Cefotaxime was found less sensitive. Low sensitivity of Gentamicin to gram positive pathogens were found in our study whereas high degree sensitivity of gram negative pathogen to gentamicin observed in reports. Antimicrobial sensitivity pattern differs in different places, in different studies at different time in the same hospital. Indiscriminate use of antibiotics leads to emergences. Resistance observed in this study may be primarily due to excessive and inadequate doses use of antibiotics at primary health facilities from where neonates are referred to our tertiary level centers.

## CONCLUSION

In this study with 90 hospitalised patients of neonatal sepsis, (61). 67.78% were found culture positive. The main causative gram positive organism was Staphylococcus aureus and gram negative organism were klebsiella pneumoniae. Incidence of antimicrobial resistance among different organism occur from time to time. Continuous evaluation at different levels of local pattern of bacterial profile and antibiotic sensitivity of pathogen should be reviewed to develop empirical therapy.

## Abbreviation

CONS – Coagulase negative staphylococcus  
 EOS –Early onset neonatal sepsis  
 LOS – Late onset neonatal sepsis  
 NICU –Neonatal Intensive Care Unit  
 NS – Neonatal Sepsis

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