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Original Research Article

The Causative Organism of Neonatal Sepsis by Blood Culture

Dr. Rawshan Ara[1*], Dr. Fauzia Nahid[2], Dr. Jebunnesa[3], Dr. Jesmine Akter Mitu[4], Dr. Nadia Haque[5]

[1]Specialist, Department of Paediatrics & Neonatology, Bangladesh Specialised Hospital, Dhaka, Bangladesh
[2]Assistant Professor, Department of Pediatrics, Dhaka Central International Medical College and Hospital, Dhaka, Bangladesh
[3]Associate Professor, Department of Paediatrics, Ashulia Women and Children Hospital, Savar, Dhaka, Bangladesh
[4]Specialist, Department of Paediatrics & Neonatology, Bangladesh Specialised Hospital, Dhaka, Bangladesh
[5]Specialist, Department of Paediatrics & Neonatology, Bangladesh Specialised Hospital, Dhaka, Bangladesh

***Corresponding Author**
Dr. Rawshan Ara
Specialist, Department of Paediatrics & Neonatology, Bangladesh Specialised Hospital, Dhaka, Bangladesh

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Abstract: ***Background:*** Neonatal septicemia is a clinical illness that occurs within the first four weeks of life and is defined by systemic signs and symptoms brought on by a widespread bacterial infection. The most frequent cause of illness and mortality in the neonatal era is bacterial infections. Early detection of this potentially fatal illness is essential for prompt treatment and a successful outcome since complications like shock, disseminated intravascular coagulation, and multi-system organ failure can lead to the fulminant and lethal course of infection. ***Objective:*** The present study was undertaken to detect the causative organism of neonatal sepsis by blood culture. ***Method of the Study:*** This is a cross- sectional study carried out in the Department of Neonatology, BSMMU, Dhaka over a period of 6 months between January 2013 to June 2013. Newborns with suspected sepsis admitted to BSMMU were the study population. Clinically diagnosed cases of neonatal sepsis aged < 28 days of both sexes whose parents or guardians provided informed consent were eligible for enrollment in the study. The subjects were selected consecutively from the study population. Data were collected using a structured questionnaire containing all the variables of interest. Data were processed and analyzed using the computer software SPSS (Statistical Package for Social Sciences). The test statistics used to analyze the data were Chi-square ($\chi 2$) or Fisher's Exact Probability Test & Student's t-Test. ***Result:*** The study included a total of 138 neonates (both preterm and term) of clinically diagnosed cases of sepsis. All the neonates were subjected to test for WBC, IT ratio CRP, and blood culture. In terms of gestational age over 80% were preterm and the rest 19.6% were term neonates. Among the neonates, 40 (29%) had early onset sepsis (EOS) and 98(71%) had late-onset sepsis (LOS). Of the culture- positive cases, Klebsiella was predominant (33.33%) followed by E. coli (22.2%), Acinetobacter (16.66%) and Pseudomonas (11.1%). Less commonly found organisms are Staphylococcus aureus, Enterobacter, Enterococcous Cytrobacter, and others. ***Conclusion:*** Sepsis screening is more useful for ruling out the diagnosis of newborn septicemia, which can be made rationally if two screenings conducted 12 to 24 hours apart are negative. It is preferable to wait for the findings of the sepsis screen before starting antibiotics in a neonate who is otherwise stable or suspected of having sepsis due to maternal risk factors.

Keywords: Neonatal Sepsis, Blood Culture, Newborns, Causative Organism.

INTRODUCTION

Neonatal septicemia is a clinical illness that occurs within the first four weeks of life and is defined by systemic signs and symptoms brought on by a widespread bacterial infection [1]. The most frequent cause of illness and mortality in the

neonatal era is bacterial infections. Early detection of this potentially fatal illness is essential for prompt treatment and a successful outcome since complications like shock, disseminated intravascular coagulation, and multi-system organ failure can lead to the fulminant and lethal course of infection [1-3].

Sepsis is the leading cause of death in developing countries, accounting for 30 to 50 percent of the 5 million newborn fatalities worldwide each year. In Asia, the prevalence of neonatal sepsis has been found to range from 7.1 to 38 per 1000 live births [4]. According to India's National Neonatal Perinatal Database (NNPD, 2002–2003), the incidence ranged from 0.1% to 4.5%. Sepsis was one of the most common causes of neonatal mortality, accounting for 19% of all neonatal deaths, according to a database compiled from 18 tertiary care neonatal centers located throughout India. The most prevalent clinical category, septicemia, had an incidence of 23 per 1000 live births [5]. Numerous Indian research has shown that Gram-negative microbes are more likely to blame for septicemia than Gram-positive species [6]. The clinical presentation is frequently vague or obscure and is frequently imitated by a number of different conditions. The early diagnosis and treatment of newborn septicemia necessitate a high index of suspicion. Without treatment, there is a high case fatality rate. In order to facilitate the management, some prenatal risk variables have been assessed as markers to anticipate newborn septicemia and scored consistently. One such helpful tool that doctors frequently use to diagnose and treat neonatal septicemia is the scoring system developed by Takkar VP and Bhakoo ON [7] consisting of six prenatal risk variables. For epidemiological and therapeutic reasons, several writers divide newborn septicemia into early-onset (occurring within the first 72 hours of life) and late-onset (typically occurring after 72 hours of life) [8].

A positive blood culture is a gold standard for the diagnosis of newborn septicemia [1, 8] Treatment delays occurs because definitive culture findings take at least 48 to 72 hours. Therefore, in addition to the Sepsis Score [7], some rapid diagnostic tests are used to diagnose septicemia early and begin presumptive treatment while awaiting a culture report.

OBJECTIVES

General Objective

- To detect the causative organism of neonatal sepsis by blood culture.

Specific Objective

- To correlate clinical and laboratory findings.

METHODOLOGY AND MATERIALS

This is a cross-sectional study carried out in the Department of Neonatology, BSMMU, Dhaka over a period of 6 months between January 2013 to June 2013. Newborns with suspected sepsis admitted to BSMMU were the study population. Clinically diagnosed cases of neonatal sepsis aged < 28 days of both sexes whose parents or guardians provided informed consent were eligible for enrollment in the study. The subjects were selected consecutively from the study population. After admission informed written consent from parents or guardians was taken and emergency management was given and septic screening was sent. Blood samples were collected by using an aseptic technique by applying povidone iodine and 70% alcohol at the site of the vein puncture. Four ml of venous blood was drawn from the peripheral vein. Two ml of blood were sent for CBC and CRP to the clinical pathology laboratory. Two ml of blood for culture were inoculated into a blood culture bottle containing tryptone soya broth (TSB). The specimen was transported immediately to the microbiological laboratory and incubated for 12-24 hours in 37 degree celcius and checked for evidence of bacterial growth. For positive broth cultures, subcultures were made on solid media (Blood Agar & Mac Conkey Agar) and incubated at 370c for 24 to 48 hours. The growth of bacteria were identified by colony morphology, Gram staining & biochemical test. Antimicrobial sensitivity testing was performed for all blood culture isolates according to the criteria of the National Committee for Clinical Laboratory Standards by the disk diffusion method. Data were collected using a structured questionnaire containing all the variables of interest. Data were processed and analyzed using the computer software SPSS (Statistical Package for Social Sciences). The test statistics used to analyze the data were Chi-square ($\chi 2$) or Fisher's Exact Probability Test & Student's t-Test. The accuracy of WBC, IT ratio, and CRP in the diagnosis of neonatal sepsis was evaluated using components of validity tests like sensitivity, specificity, positive and negative predictive values (PPVs and NPVs), etc. The level of significance was set at 0.05 and $p < 0.05$ was considered significant.

- **Inclusion Criteria**
  - Newborns with suspected sepsis.
  - Neonatal sepsis aged < 28 days.
  - Neonatal sepsis of both sexes.

- **Exclusion Criteria**
  - Prior treatment with antibiotics.
  - Developed the signs of sepsis within 72 hours of discontinuation of the antibiotics.
  - Newborns with perinatal asphyxia or meconeum aspiration syndromes.

- Inborn errors of metabolism and
- Baby with congenital anomalies.

RESULT

The present study intended to detect the causative organism of neonatal sepsis by blood culture. The study included a total of 138 neonates (both preterm and term) of clinically diagnosed cases of sepsis. All the neonates were subjected to test for WBC, IT ratio CRP and blood culture. In terms of gestational age over 80% were preterm and the rest 19.6% were term neonates (Table I). Male neonates were higher (57%) than females (Figure I). The result shows that 81% of the neonates were of low birth weight (<2.5 kg) (Figure II). The predominant clinical finding at presentation was lethargy (89.1%) followed by tachypnoea (47.1%), poor feeding (43.5%), abdominal distension (34.1%), grunting (27.5%), hypothermia (25.4%), vomiting (23.2%), apnoea (20.3%), hyperthermia (14.5%). Other seldom observed signs were cyanosis (9.4%), convulsion (7.2%), bulged fontanel (2.9%) and tachycardia (2.9%) (Table II). Approximately 43% of the neonates were born by normal delivery and 57.2% by caesarean section (Table III). Among the neonates, 40 (29%) had early onset sepsis (EOS) and 98(71%) had late-onset sepsis (LOS) (Table IV). Of the 138 cases subjected to blood culture, 36(26.08%) yielded growth of pathogenic microorganisms (Table V). Based on WBC count, IT ratio and serum CRP 21.7%, 31.1% and 88.4% cases were screened respectively as having sepsis (Table VI). About one-quarter (23.2%) of the neonates were released in less than 14 days, 29.7% in 14 – 21 days and 47.1% in 3 or >21days time. The mean duration of hospitalization was 21.2 days (Table VII). Of the culture-positive cases, *Klebsiella* was predominant (33.33%) followed by *E. coli* (22.2%), *Acinetobacter* (16.66%) and *Pseudomonas* (11.1%). Less commonly found organisms are *Staphylococcus aureus, Enterobacter*, *Enterococcous Cytrobacter,* and others (Table VIII).

Table I: Distribution of neonates by gestational age (n = 138)

Gestational age (weeks)	Frequency (N)	Percentage (%)
28-<37	111	80.43
37-42	27	19.6

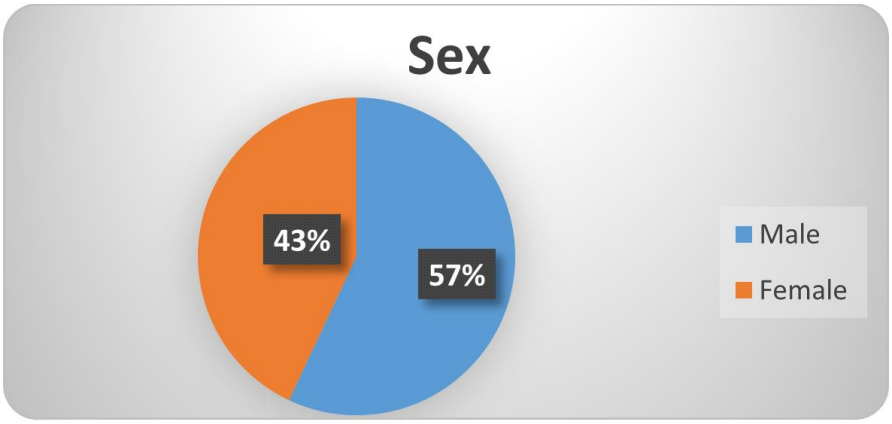

Figure I: Distribution of patients by their sex (n=138)

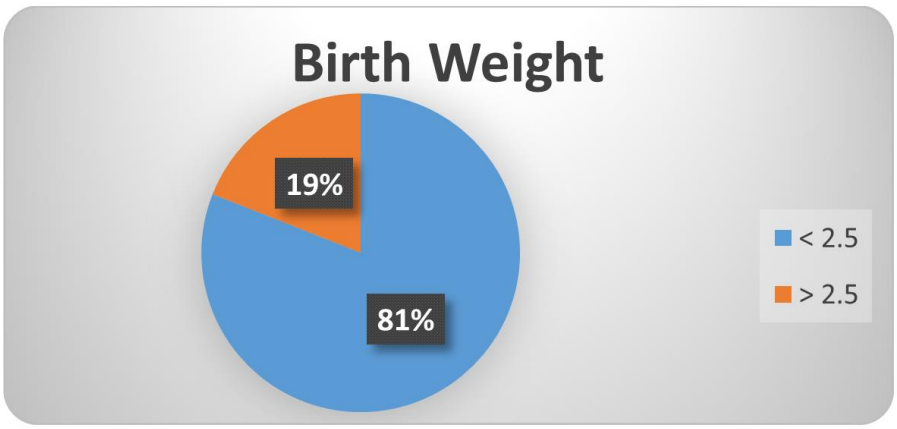

Figure II: Distribution of patients by birth weight (n=138)

Table II: Distribution of patients by clinical findings (n = 138)

Clinical Findings	Frequency	Percentage
Lethargic	123	89.1
Tachypnoea	65	47.1
Poor Feeding	60	43.5
Abdominal distension	47	34.1
Grunting	38	27.5
Hypothermia	35	25.4
Vomiting	32	23.2
Apnoea	28	20.3
Hyperthermia	20	14.5
Cyanosis	13	9.4
Convulsion	10	7.2
Bulged fontanel	04	2.9
Tachycardia	04	2.9

Table III: Distribution of neonates by mode of delivery (n = 138)

Mode of Delivery	Frequency	Percentage
Normal	59	42.8
LUCS	79	57.2

Table IV: Distribution of neonates by mode of onset of sepsis (n = 138)

Mode of onset of sepsis	Frequency	Percentage
Early onset sepsis (EOS)	40	29
Late-onset sepsis (LOS)	98	71

Table V: Diagnosis of sepsis established by blood culture (n = 138)

Diagnosis	Frequency	Percentage
Culture proven sepsis	36	26.08
Suspected or Clinical sepsis	102	73.91

Table VI: Sepsis cases screened by total WBC count, IT ratio and serum CRP (n = 138)

Screening tests	Frequency	Percentage
WBC count		
<5000 cu-mm	5	3.6
5000 – 25,000 /cu-mm	108	78.3
> 25,000 /cu-mm	25	18.1
IT ratio		
> 0.2	43	31.1
< 0.2	95	68.9
Serum CRP		
> 6	122	88.4
< 6	16	11.6

Table VII: Distribution of the neonates by duration of hospitalization (n =138)

Duration of hospitalization* (days)	Frequency	Percentage
< 14	32	23.2
14 – 21	41	29.7
≥21	65	47.1
* Mean = (21.2 ± 7.5) days; range = (9 – 35) days.		

Table VIII: Causative organisms among culture-positive cases (n = 36)

Causative organisms	Frequency	Percentage
Klebsiella	12	33.33
E. coli	8	22.22
Acinetobacter	6	16.66

Causative organisms	Frequency	Percentage
Pseudomonas	4	11.11
Staphylococcus aureus	2	5.55
Enterobacter	1	2.77
Enterococcous	1	2.77
Cytrobacter	1	2.77
Others (Providentia)	1	2.77

DISCUSSION

Neonatal septicemia is a clinical illness that occurs within the first four weeks of life and is defined by systemic signs and symptoms brought on by a widespread bacterial infection [9]. The most frequent cause of illness and mortality in the neonatal era is bacterial infections. Neonatal bacterial sepsis occurs between 0.3% and 3/1000 live births (LB) in Europe [10], between 1/1000 and 4/1000 LB in North America [11], between 1.4/1000 LB and a hospital study in Jamaica [12], between 8.9% and 10/1000 LB in Guadeloupe [13], and between 10/1000 LB at the San Fernando general hospital in South Trinidad [14]. In the present study, there were 40 (29%) cases of early and 98 (71%) of late-onset sepsis. We found that early-onset sepsis was less common than late-onset disease which is in contrast with the reports from other developing countries [11, 15], but compatible with a report from another study in Bangladesh that late-onset disease was more common [16]. In our study, the causative organisms were *Klebsiella* (33.33%), *E. coli* (22.22%), *Acinetobacter* (16.66%), *Pseudomonas* (11.11%). Almost similar findings from a study done in India showed Pseudomonas aeruginosa was the most common organism, followed by Klebsiella spp. and Escherichia coli [17]. Similar patterns have been reported in Trinidad [14] and Nigeria [18], with Pseudomonas aeruginosa contributing at 26%, Klebsiella pneumonia at 14%, Escherichia coli 7% and Enterobacter aerogenes 5%. Total leucocyte count is of little clinical in the diagnosis of neonatal sepsis because of the wide variation in values. About 72% of neonates had normal WBC count in culture-proven cases in this study. The reason for this high false negative result might be due to the time interval between the onset of bacteremia and sampling. The WBC count was found to be the most specific of all tests but the least sensitive. The specificity of WBC is consistent with other studies [19-21] recommend an elevated IT ratio is statistically significant in the diagnosis of neonatal sepsis. Each test used in this study had different specificity, sensitivity, and predictive accuracy. The tests were selected on the basis of ease, speed of performance, cost, and availability. These tests have the practical advantage of being applicable to all infants including those who have received antibiotic therapy prior to evaluation. It has been demonstrated that a combination of tests increases the sensitivity, specificity and positive predictive accuracy compared with a single test for the diagnosis of neonatal sepsis. Besides, it is also known that these tests can be positive in a variety of non-infective disorders, conversely, septicemia can occur with normal findings.

CONCLUSION AND RECOMMENDATION

Sepsis screening is more useful for ruling out the diagnosis of newborn septicemia, which can be made rationally if two screenings conducted 12 to 24 hours apart are negative. It is preferable to wait for the findings of the sepsis screen before starting antibiotics in a neonate who is otherwise stable or suspected of having sepsis due to maternal risk factors. The confirmation of sepsis by the sepsis screen tests may help in avoiding unneeded antibiotic therapy because symptoms indicative of sepsis may be brought on by a variety of different conditions. Blood culture is still the "Gold standard" for septicemia diagnosis in newborns and ought to be performed in every instance of suspected septicemia. Positive blood culture and the testing of the isolates for antibiotic susceptibility are the best tools for determining the best antimicrobial therapy for treating neonatal septicemia due to the fluctuating spectrum of the causative agents of neonatal septicemia and their antibiotic susceptibility patterns from time to time and from hospital to hospital. A scoring system should be created using tests that are simple to administer, affordable, and readily available. The tests should also, ideally, identify all infants who are infected, so that disease can be confidently excluded with negative test results (high negative predictive value), and the antibiotic can be stopped early, lowering the cost, length of hospital stay, and parents' anxiety. The results of a large-scale multicenter investigation should be extrapolated in order to develop policy guidelines for the identification and treatment of newborn sepsis in our environment.

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