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Bacterial Predominance and Antimicrobial Susceptibility Patterns of Blood Culture at a Tertiary Care Hospital, Bangladesh

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Abstract

Background: The problem of widespread resistant bacteria has become a major threat to reduce the effectiveness of antibiotics worldwide. **Objective:** The aim of this study was to identify the bacterial pathogens from bold culture and determine their antibiotic susceptibility pattern. **Methodology:** This cross-sectional study was conducted in the department of Microbiology at Monno Medical College, Manikganj, Bangladesh during the period from January 2019 to December 2019 for duration of one year. Blood was collected according to blood collection guidelines and inoculated into BacT/ALERT FA plus and BacT/ALERT PF plus aerobic blood culture bottles respectively. After collection these bottles were immediately incubated in BacT/ALERT 3D (manufactured by bioMerieoux, France) a fully automated blood culture system. Antimicrobial susceptibility test was done for all isolated bacteria by disc diffusion method. **Results:** A total number of 180 patients were recruited, among them 12 (7%) yielded growth of different bacteria. The most frequently isolated bacteria were Streptococcus Spp 5(42%) followed by Staphylococcus aureus and Escherichia coli which were 4(33%) and 3(25%) respectively. Escherichia Coli which showed highly sensitive to Imipenum, Amikacin, Linezolid, Gentamicin (N=3, 100%). **Conclusion:** There were a high percentage of bacteria isolated from blood culture and resistant to several antibiotics. For the selection of appropriate antibiotic therapy in bacterial infections the antibiotic susceptibility testing is mandatory. *[Journal of National Institute of Neurosciences Bangladesh, January 2023;9(1):48-53]*

Keywords: Blood culture, BSI, Antimicrobial susceptibility; Multidrug resistance

Introduction

The blood culture is a laboratory test in which blood, taken from the patient, is inoculated into bottles containing culture media to determine whether infection-causing microorganisms are present in the patient's bloodstream. It is the most important way to diagnose the etiology of bloodstream infections and sepsis and has major implications for the treatment of those patients. A positive blood culture either establishes or confirms that there is an infectious etiology for the patient's illness[1]. Bloodstream infections (BSIs), which include bacteremia when the infections are bacterial and fungemia when the infections are fungal, are infections present in the blood[2]. Sepsis is life-threatening organ dysfunction caused by a deregulated host response to infection. Septicemia is a clinical syndrome characterized by fever, chills, malaise, tachycardia, etc. when circulating bacteria multiply at a rate that exceeds removal by phagocytosis[3]. Neonatal sepsis refers to systemic and generalized bacterial infection of the newborn documented by a positive blood culture in the first 4 weeks of life, and is one of the four leading causes of neonatal mortality in Bangladesh and other developing countries[4]. Efforts to decrease mortality rates in children under the age of 5 years, as targeted in the United Nations Millennium Development Goal 4, have been

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more successful in reducing the death rate in older children than in neonates. In 2010, deaths in the neonatal period contributed 40% and 52% of mortality rates in children under the age of 5 years worldwide and in Southeast Asia, respectively. The rate of blood stream infections in children is about 20 to 50% in developing countries[5].

The diagnosis of BSI can be confirmed by blood culture. A wide variety of bacteria has been isolated from the blood samples collected from critically ill patients. The most common include gram-negative bacteria such as Escherichia coli, Pseudomonas aeroginosa, Klebsiella species, Enterobacter, Serratia, Citrobacter, Salmonella and Acinetobacter[6]. However, a rapid rise in the trend of gram-positive isolates such as Streptococci/Enterococci or Staphylococci as a cause of sepsis have been observed over the period, possibly due to frequent use of invasive procedures and the increasing trends of hospital-acquired infection[7]. Recently reported that the frequency of gram-negative bacilli among clinical isolates were over twice higher than that of gram-positive cocci[8]. The geographic variety is likely associated with regional differences in socioeconomic development[9].

The timely diagnosis and rationale use of appropriate antibiotics remains the cornerstone in treating and managing patients with sepsis. However, frequent and irrational use of broad-spectrum antibiotics in critically ill patients who stay in the ICU for more prolonged periods has increased bacterial resistance over time[10]. Antibiotics resistance is becoming an alarming problem in developing countries. To accurately estimate the challenge, the surveillance of AMR profiling at local, regional, and national levels has been employed to understand the global trends on the type of predominant pathogens and their respective resistance profiling[11].

Methodology

This cross-sectional study was conducted in the department of Microbiology at Monno Medical College, Manikganj, Bangladesh. This study was carried out during the period from January 2019 to December 2019 for duration of one year. A total number of 180 respondents were selected both outdoor & indoor patient irrespective of age, sex, growth positivity and antibiotic susceptibility. Age variable was divided in two groups: below 15 years age group and above 15years age group. Patients who were taking antibiotics within last 14 days those were excluded in this study. About 5-10 ml blood was collected from patient using strict aseptic precautions and inoculated immediately into BacT/ALERT FA plus aerobic blood culture bottles with 0.025% of sodium polyanethol sulfonate (SPS) as anticoagulant. In cases of below 15 years age group 1-2mL of blood was inoculated in BacT/ALERT PF plus pediatric blood culture bottles.

After collection these bottles were immediately incubated in BacT/ALERT 3D (manufactured by bioMerieoux, France) a fully automated blood culture system for detection of growth in blood culture. In case of a positive growth, the BacT/ALERT automatically gives an alert. The positive bottles were then subculture on Blood Agar, Chocolate Agar and Mac Conkey Agar (HI Media Laboratories Pvt. Limited, India) and then incubated aerobically at 37°C for 24 h. Standard microbiological methods were applied for the identification of the bacterial species. All the isolated organisms were identified by their colony morphology, staining characters pigment production, motility, oxidase, catalase, TSI and MIV, citrate tests and further confirmed by relevant biochemical tests. Susceptibility to antimicrobial agents of all isolates was done by Kirby Bauer modified disc diffusion technique using Mueller Hinton agar plates and zones of inhibition were interpreted according to CLSI guidelines. Antibiotic discs such as cefotaxime (30µg), ceftriaxone (30µg), amoxicillin (20µg), ciprofloxacin (5µg), cefuroxime (30µg), amikacin (30µg), azythromyin (15µg), gentamicin (10µg), doxycycline (30µg), nalidixic acid (30µg), tetracycline (30µg), cefixim (30µg), imipenem (10µg), nitroflurantoin (300µg), vancomycin (30µg/disc), linezolid (30µg/disc), sulphamethozazole+trimethoprim (25 µg).

All data were processed and analyzed with the help of IBM SPSS (Statistical package for Social Sciences) Version 22.0. Quantitative data were expressed as mean and standard deviation and qualitative data as frequency and percentage. Association was analyzed by Pearson's Chi square (X2) test. A probability (p) value of <0.05 was considered statistically significant. Throughout this procedure informed written consent was taken from all patients or their legal guardians before specimen collection.

Results

A total number of 180 respondents were recruited after fulfilling the inclusion and exclusion criteria. Among them 12 (7%) yielded growth of different bacteria (Figure I).

Among 180 cases, participants who had above 15 years age group were predominant 93(52%) than participants had below 15 years age group which were 87(48%) (Figure II).

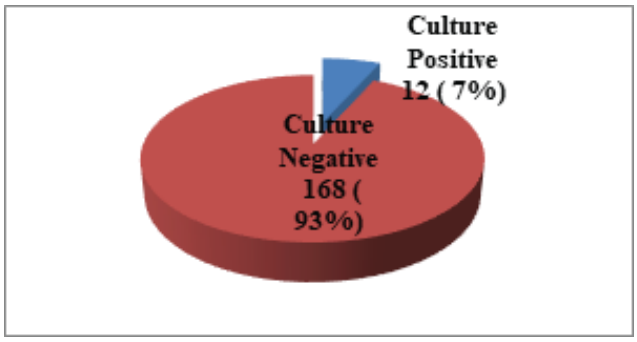

Figure I: Pie Chart Showing Distribution of Culture Positive Cases

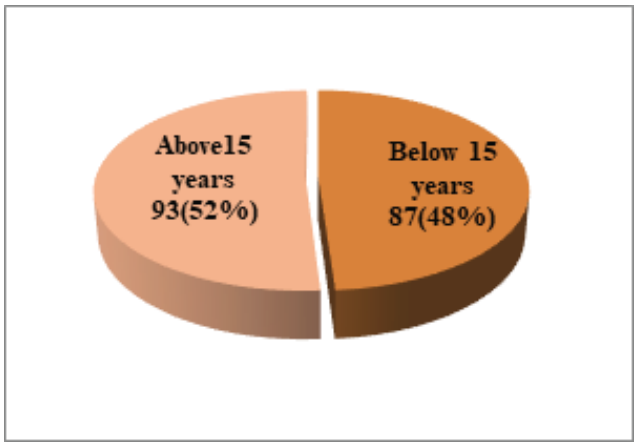

Figure II: Pie chart showing Distribution of Participants

About 87 respondents were below 15 years of age group in which male were predominant 54(62.1%) and female were 33(37.9%). About 93 respondents were above 15 years of age group in which 49(52.6%) were male and 44(47.4%) were female. The association between gender and age group did not differ significantly (P =0.161) (Table 1).

Table 1: Distribution & percentage of gender among age group

Gender	Age Group		Total	P value
	below 15 years age group	above 15 years age group		
Female	33(37.9%)	44(47.4%)	77(43%)	
Male	54(62.1%)	49(52.6%)	103(57%)	0.161
Total	87(100.0%)	93(100.0%)	**180(100.0%)**	

Chi- Square (X^2) test was performed to see the association. P≤0.05 was determined as level of significance

The most frequently isolated bacteria were *Streptococcus Spp* 5(42%) followed by *Staphylococcus aureus* and *Escherichia coli* which were 4(33%) and 3(25%) respectively.

The most predominant isolated bacteria was *Streptococci* which showed high sensitive to Imipenum, Linezolid, Gentamicin (N=4, 80%). They

Table 3: Showing antibiotic sensitivity pattern of the isolated bacteria.

Name of Antibiotics	Name of bacteria			Total
	***Staphylococcus aureus* (4)**	***Streptococci*(5)**	***Escherichia coli* (3)**	
Vancomycin	1(25.0%)	1(20.0%)	0(0%)	2
Sulphamethoxazole	0(0.0%)	1(20.0%)	2(66.7%)	3
Imipenem	3(75.0%)	4(80.0%)	3 (100.0%)	10
Amikacin	0(0.0%)	2(40.0%)	3(100.0%)	5
Azythromyin	0(0.0%)	3(60.0%)	0(0.0%)	3
Amoxycillin	1(25.0%)	0(0.0%)	1(33.3%)	2
Ciprofloxacin	3(75.0%)	3(60.0%)	1(33.3%)	7
Linezolid	3(75.0%)	4(80.0%)	3(100.0%)	10
Tetracycline	2(50.0%)	3(60.0%)	1(33.3%)	6
Doxycycline	3(75.0%)	1(20.0%)	0(0.0%)	4
Sulphamethozazole+ Trimethoprim	1(25.0%)	0(0.0%)	0(0.0%)	1
Cefotaxime	1(25.0%)	3(60.0%)	1(33.3%)	5
Cefuroxime	2(50.0%)	0(0.0%)	0(0.0%)	2
Gentamicin	4(100.0%)	4(80.0%)	3(100.0%)	11
Ceftriaxone	3(75.0%)	3(60.0%)	1(33.3%)	7
Nalidixic Acid	1(25.0%)	0(0.0%)	0(0.0%)	1
Nitroflurantoin	2(50.0%)	2(40.0%)	0(0.0%)	4
Cefixim	2(50.0%)	0(0.0%)	0(0.0%)	2

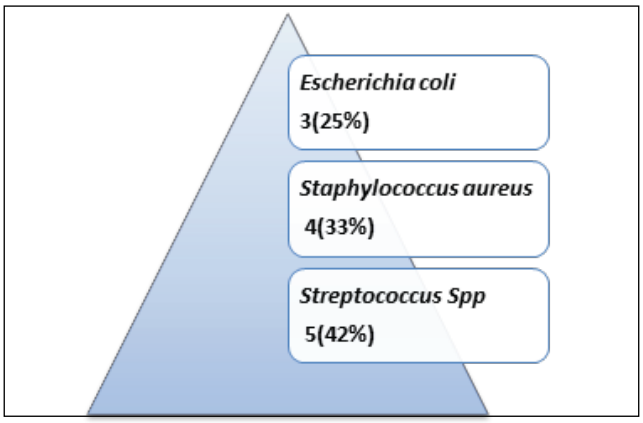

Figure III: Diagram showing distribution of different isolated bacteria

showed moderate sensitive to Ciprofloxacin, Tetracycline, Cefotaxime, Cefuroxime (N=3, 60%) and low sensitive to Amikacin, Nitroflurantoin (N=2, 40%) and considerable towards Vancomycin, Sulfamethoxzole, Doxycycline, (N=1, 20%). Another isolated bacteria was *Staphylococcus aureus* which found in equal number in both group. They showed highly sensitive to Gentamicin (N=4,100%) and considerable towards to Imipenum, Linezolid, Ciprofloxacin, Cefuroxime, Doxycycline (N=3, 75%). Moderate sensitive to Tetracycline, Cefixim, Cefuroxime, Nitroflurantoin (N=2, 50%) and low sensitive to Vancomycin, Amoxycillin, Cefotaxime, Nalidixic Acid, Sulphamethozazole +Trimethoprim(N=1,25%). *Escherichia Coli* which showed highly sensitive to Imipenum, Amikacin, Linezolid, Gentamicin (N=3, 100%). They showed moderate sensitive to Sulfamethoxzole (N=2, 66.7%) and low sensitive to Amoxycillin, Ciprofloxacin, Tetracycline, Cefotaxime,Cefuroxime(N=1,33.3%). (Table: 3)

Discussion

To evaluate the predominant bacterial in blood culture and their antimicrobial susceptibility patterns 180 patient has been selected according to inclusion and exclusion criteria. Blood culture positivity has been seen in 12 (7%) cases which is quite similar to Gohel et al (9.2%)12 and Mehta et al. (9.94%)13 and Wang et al (12.9%)[14].

The participants who were above 15 years age were predominant 91(51%) than participants were below 15 years age 87(49%). About 87 respondents were below 15 years age of which male were predominant 54(62.1%) and female were 33(37.9%). The incidence of neonatal septicemia is variable because it depends on various factors like gestational age, fetal birth weight, maternal nutrition, perinatal care and hygienic conditions, child health care facilities[15]. In this study 93 respondents were above 15 years age of which 47(51.6%) were male and 44(48.4%) were female. The association between gender and age group did not differ significantly (P =0.161).

The most frequently isolated bacteria were Streptococcus Spp 5(42%) followed by *Staphylococcus aureus* and *Escherichia coli* which were 4(33%) and 3(25%) respectively. Thus, predominance of either the Gram positive or Gram negative bacteria isolates is influenced by geographical location and changes in time16. The study of Gohel et al has been reported that the positive blood cultures were obtained in 9.2% of cases of which Gram positive bacteria accounted for 58.3% of cases with staph aureus predominance[17]. A study has been conducted among blood stream infection patients has been recorded that about 44.6% isolates are Gram-negative bacilli and frequently identified species were *Escherichia coli*, *Klebsiella pneumonia* and *Pseudomonas*[18]. Prevalence of gram negative bacterial etiology of septicemia in children has been recorded by several other authors[19,20]. On the other hand, some study has been recorded preponderance of *S. aureus* as bacterial cause of septicemia in neonates [20,21].

The most predominant isolated bacteria were *Streptococci* which showed high sensitive to imipenum, linezolid, gentamicin (n=4, 80%) and subsequent sensitive to Ciprofloxacin, Tetracycline, Cefotaxime, Cefuroxime (n=3, 60%). Badulla et al[22] and other study has been showed that the most sensitive drugs for Gram positive isolates were tetracycline, clindamycin, daptomycin, linezolid, carbapenems, colistin, and aminoglycosides. According to antimicrobial susceptibility pattern to different antibiotics, Escherichia Coli was showed highly sensitive to imipenum, amikacin, linezolid, gentamicin (100.0%) and low sensitive to amoxycillin, ciprofloxacin, tetracycline, cefotaxime, cefuroxime (33.3%). The most sensitive drugs for Gram negative bacteria were imipenum, gentamicin[23] and low sensitive to quinolones, penicillins, and cephalosporins[12]. This finding is an indication of the irrational use of new and strong antibiotics for simple infectious or even viral diseases that leads to widespread bacterial resistance to these antibiotics.

Conclusion

The study of bacteriological profiles with antibiotic susceptibility patterns plays a pivotal role in the effective management of bacteraemia cases. Early detection of causative pathogen and initiation of targeted therapy is the mainstay of treatment. This study anticipated that gram-positive bacterias are predominant isolates responsible for BSI. Among them, *Streptococcus species* and *Staphylococcus species* were the most common, respectively. The rising trends in antibiotic resistance emphasis the importance of hospital infection control policies and implementation of rational prescription of antimicrobial practices with continued surveillance to prevent the emergence and further spread of resistant bacterial pathogens.

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Data Availability
Any inquiries regarding supporting data availability of this study should be directed to the corresponding author and are available from the corresponding author on reasonable request.

Ethics Approval and Consent to Participate
Ethical approval for the study was obtained from the Institutional Review Board. As this was a prospective study the written informed consent was obtained from all study participants. All methods were performed in accordance with the relevant guidelines and regulations.

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