

Original Article**The Bacterial Profile And Antibiotic Susceptibility Pattern Among Patients With Suspected Blood Stream Infection**

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Abstract

Background: Blood stream infection cause by bacterial pathogens are common in Bangladesh and are often treated empirically. Knowledge of local antimicrobial activity profiles of the most common bacteria causing such infections are of great importance and may play a positive role in healthcare management. The aim of the study was to identify the bacterial pathogens causing bacteraemia among the patients of Sylhet Womens Medical College, Bangladesh and to determine their antibiotic susceptibility pattern.

Method: This retrospective cross-sectional study was conducted from April 2019 to January 2020. A total of 2085 blood samples were studied for the etiological and antimicrobial susceptibility pattern. The blood cultures positive isolates were identified by Versa TREK automated blood culture machine. Further identification of bacterial pathogens and their antimicrobial susceptibility test were performed using standard microbiological procedures.

Result: Overall, 19.4% of the culture blood samples were positive and Gram positive bacteria were predominant throughout the study period. Staphylococci aureus was the most frequently isolated organism (55%) followed by Escherichia Coli (21%) and a high percentage of these isolates were multidrug resistant (MDR).

Conclusion: This study reveals a significant prevalence of bacterial isolates in blood with multi drug resistance and it highlights the need for periodic surveillance of etiologic agent and antibiotic susceptibility to prevent further emergence and spread of resistant bacterial pathogens.

Keywords: Antibiotic susceptibility, bloodstream infection, multidrug resistance.

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Introduction

Blood stream infection due to bacterial pathogen leads to a significant amount of morbidity and mortality worldwide and in Bangladesh.

Recent studies have shown a rapid increase in the number of both community acquired and nosocomial blood stream infection^{1,2}. Although blood culture results do not always come positive for bacteremia or septicemia patients, it remains the gold standard to diagnose infection in blood³. Bacteraemia is often life threatening depending on severity, age, sex and other risk factors. It has a mortality rate ranging from 4%-41.5%⁴. So prompt empirical treatment choices based on clinical symptoms has to be made before laboratory test results are available. Early detection of causative organisms and immediate treatment with appropriate antibiotic can reduce the length of hospital stay, a significant amount of healthcare related cost and most significantly a high rate of morbidity and mortality. In recent years there has been both a surge in antibiotic

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resistance (ABR)⁵⁻⁷ and a decline in the rate of single, multi and extensively drug resistance bacteria is alarming and a matter of huge concern worldwide¹⁰. Bangladesh together with other developing countries are facing higher degree of threat poses by ABR in term of mortality and economic burden. Because of the widespread misuse of antibiotics, non-human antibiotic use, poor quality of drugs inadequate surveillance and factors associated with individual and national poverty poor healthcare standards, malnutrition, chronic and repeated infections, unaffordability of more effective and costly drug^{11,12}. Unavailability of newer antibiotic means resistance must be contained before we run out of options to battle it. Our healthcare systems operate on poor hygiene system and lack proper facilities to contain infections. Early treatment is usually based on the patients clinical symptoms rather than diagnostic results. So the knowledge of most frequently isolated pathogenic organisms and their antibiotic sensitivity and resistance pattern can help physicians in making proper empirical treatment choices before laboratory test result are available. The present study deals with the isolation of blood culture isolate from patients of a hospital in Sylhet, Bangladesh and their susceptibility pattern.

Aims and Objectives

The study was conducted to identify the causative agents of bacteraemia and to assess the antibiotic susceptibility pattern of the isolates among bacteraemia suspected patients.

Materials and method

This retrospective cross-sectional study was conducted from 20.04.19-20.01.21 at SWMCH. A total of 2085 samples from adult and children were processed for culture using Versa TREK automated blood culture machine to identify the presence of bacterial pathogens.

new antibiotic development^{8,9}. The emerging of Collected blood samples were incubated in the machine for 5 days.

The preliminary signal of bacterial growth in Versa TREK PUDOX 1 bottle was detected and displayed on the 3D monitor mentioning the detection time. Specific identification of all cultures positive samples was accomplished by sub culture on Blood agar, Chocolate agar and MacConKey's agar media. The culture positive samples were identified by colony morphology, microscopy and conventional biochemical test as per the standard protocol followed in microbiology laboratory. Blood culture bottles that do not show any significant growth till the 5th day of incubation were reported as culture negative.

Antimicrobial susceptibility test was carried out by the Kirby-Bauer disc diffusion method using Muller Hintor agar media and antibiotic disc.

The results of susceptibility test were classified into sensitive (S) and resistant (R). The isolates with intermediate susceptibility were included in resistant categories.

The drugs for disc diffusion testing were in following concentration.

Data collection and analysis

Raw data were collected manually. Data were collected and extracted manually from the microbiology registration books of the hospital laboratory using checklists and were analyzed and expressed descriptively.

Ethical Consideration:

An ethical approval letter was obtained from the Sylhet Women's Medical College Hospital prior to the commencement of data collection. The data were retrospective and, therefore, there was no need for consent.

Result

Socio-demographic Characteristics

During the period of study, 2085 blood samples were analyzed and bacteremia was confirmed in 406 cases (19.47%). From the cases 1041 (49.92%) were male and 1044 (50%) were female. Culture positivity rate was highest in the age group of <28 days (32% 97/246)

followed by those in age group of 29 Days to 5 years (20.65% 82/397). The age group >60 years had the maximum request for blood culture 412 (19.76%) followed by age group between 29 days to 5 years 397 (19%) whereas age group 51-59 years found to have least request with 111 (5.3%).

Characteristics	Number of tested sample	Positive results, n(%)
Gender		
Male	1041	234(22)
Female	1044	172(16)
Age < 28 days	246	79(32.1)
29 days-5 years	397	82(20.6)
6-14 years	138	28(20.2)
15-23 years	297	55(18.5)
24-32 years	221	41(18.5)
33-41 years	131	16(12.2)
42-50 years	131	16(12.2)
51-59 years	111	16(14.4)
>60 years	412	71(17.2)
Total	2085	406

Table 1: Prevalence of bacteremia among sex and age groups of suspected patients in the Sylhet Womens Medical College & Hospital from April 2019- January 2020.

Bacterial Isolates

From a total 2085 bacterimia-suspected cases 406 were positive. From the isolated bacteria gram-positive bacteria were more prevalent than gram negative bacteria. The most commonly isolated bacteria were staphylococci

aureus accounting for (55%) of the total isolates. Other frequently isolated pathogens included Escherichia Coli (21%), coagulase negative staphylococcus (12.8%), Streptococcus pneumonia (5.17%) and Klebsiella pneumonia (2.46%).

Table Two

Organisms	Frequency (%)
Staphylococcus aureus	223 (55)
Coagulase(-) staphylococci (CONS)spp	52 (12.80)
Streptococcus pneumoniae	21 (5.17)
Escherichia Coli	88 (21)
Klebsiella pneumoniae	10 (2.46)
Pseudomonas	3 (0.7)
Salmonella spp	3(0.7)
Haemophilus influenzae	2 (0.49)
Neisseria meningitidis	1 (0.24)
Candida	2 (0.49)
Enterococcus Faecalis	1(0.24)
Total	406

Table 2: The frequency of bacterial isolates from patients with bacteremia at Sylhet Womens Medical College & Hospital from April 2019-January 2020.

Abbreviation: CONS, coagulase-negative staphylococci spp.

Antibiotic Susceptibility Pattern

The observed drug susceptibility pattern of gram positive isolates shows an intermediate level of resistance (60%-90%). The most commonly isolated blood culture pathogen staphylococcus aureus shows 90% resistance toward cefixime with 85% resistance toward Amoxicillin and 83% to Co-trimoxazole, 82% to Cefazidime and 80% toward azithromycin with only a low level of resistance to the rest of the tested drug. Coagulase negative staphylococci (CONS) were also highly resistant to Co-trimoxazole (100%), Cefixime (96%), Amoxicillin (84%) and Cefazidime (80%). Another gram positive bacteria Streptococcus pneumoniae shows more

than 90% resistance to Co-trimoxazole, Cefixime and more than 85% resistance to Amoxicillin and Cefuroxime.

Among the gram negative bacilli Escherichia coli revealed high level of resistance (97% for Linezolid) and Vancomycin (94%) followed by 86% resistance toward amoxicillin and 85% and 82% resistance to Cefuroxime and Co-trimoxazole respectively. All the Gram Negative isolate showed mild to moderate level of sensitivity towards the rest of the antibiotics. As indicated in the table, the most sensitive antibiotics were Amikacin, Meropenem and Ceftriaxone. Most resistance ones were Amoxicillin, Cefixime and Co-trimoxazole.

Isolates	S.aureus	CONS	S.Pneumoniae	E.Coli	Klebsiella	Pseudomonas	Salmonella	H.Influenzae	N.Meningitidis	Enterococcus
	Sp.(223)	Sp.(52)	Sp.(21)	Sp.(88)	Sp.(10)	Sp.(3)	Sp.(3)	Sp.(2)	Sp.(1)	Sp.(1)
Antibiotics	SR	SR	SR	SR	SR	SR	SR	SR	SR	
Amoxicillin	S55(24.6) R 168(75.3)	S8(15.3) R44(84.6)	S3 (14.2) R 18(85.7)	S23(26) R65(73.8)	S2(20) R8(80)	S0(0) R3(100)	S2(66.6) R1(33.3)	S0(0) R2(100)	S0(0) R1(100)	S1(100%) R0(0%)
Amikacin	S202(90.5) R 21(9.4)	S47(90.3) R5(9.6)	S 14(66.6) R7(33.3)	S71(80.6) R17(19.3)	S10(100) R0(0)	S3(100) R0(0)	S3(100) R0(0)	S2(100) R0(0)	S1(100) R0(0)	S1(100%) R0(0%)
Azithromycin	S47(21) R 176 (78.9)	S15(28.8) R37(71)	S8(38) R16(61.9)	S40(45.4) R48(54.5)	S6(60) R4(40)	S1(33.3) R2(66.6)	S3(100) R0(0)	S2(100) R0(0)	S0(0) R1(100)	S1(100%) R0(0%)
Cefaclor	S122(54.7) R 101(45.2)	S23(44.2) R29(55.7)	S6(28.5) R15(71.4)	S43(48.8) R45(51)	S5(50) R5(50)	S2(66.6) R1(33.3)	S2(66.6) R1(33.3)	S0(0) R2(100)	S0(0) R1(100)	S1(100%) R0(0%)
Cefixime	S22(9.8) R201 (90)	S2(3.8) R50(96)	S2(9.5) R19(90.4)	S24(27.2) R64(72.7)	S3(30) R7(70)	S0(0) R3(100)	S2(66.6) R1(33.3)	S0(0) R2(100)	S1(100) R0(0)	S1(100%) R0(0%)
Ceftriaxone	S 143(64) R80(35.8)	S32(61.5) R20(38.4)	S7(33.3) R14(66.6)	S58(65.9) R30(34)	S6(60) R4(40)	S2(66.6) R1 (33.3)	S2(66.6) R1(33.3)	S1(33.3) R2(66.6)	S1(100) R0(0)	S1(100%) R0(0%)
Cefuroxime	S96(43) R127(56.9)	S19(36.5) R33(63.4)	S3(14.2) R18(85.7)	S17(19.3) R71(80.6)	S1(10) R9(90)	S1(33.3) R2(66.6)	S1(33.3) R2(66.6)	S0(0) R2(100)	S0(0) R1(100)	S1(100%) R0(0%)
Ceftazidime	S 36(16.1) R187(84)	S10(19.2) R42(80.7)	S9(42.8) R12(57)	S34(31.6) R54(61.3)	S2(20) R8(80)	S1(33.3) R2(66.6)	S1(33.3) R2(66.6)	S1(50) R1(50)	S1(100) R0(0)	S1(100%) R0(0%)
Ciprofloxacin	S171(76.6) R52(23.3)	S35(67.3) R17(32.6)	S21 (100) R0(0)	S67(76) R21(23.8)	S9(90) R1(10)	S3(100) R0(0)	S(66.6) R1(33.3)	S2(100) R0(0)	S1(100) R0(0)	S1(100%) R0(0%)
Colistin	S130(58.2) R93(41.7)	S28(53.8) R24(46)	S9(42.8) R12(57)	S64(72.7) R24(27.2)	S7(70) R3(30)	S2(66.6) R1(33.3)	S3(100) R0(0)	S1(50) R1(50)	S0(0) R1(100)	S1(100%) R0(0%)
Cotrimoxazole	S30(13.4) R 193(86.5)	S0(100) R52(100)	S2(9.5) R19(90.4)	S12(13.6) R76(86.3)	S1(10) R9(90)	S0(0) R3(100)	S0(0) R3(100)	S0(0) R2(100)	S1(100) R0(0)	S1(100%) R0(0%)
Doxycycline	S197(88.3) R 26(11.6)	S44(84.6) R8(15.3)	S13(61.9) R8(38)	S64(72.7) R24(27.2)	S9(90) R1(10)	S1(33.3) R2(66.6)	S2(66.6) R1(33.3)	S2(100) R0(0)	S0(0) R1(100)	S1(100%) R0(0%)
Gentamicin	S194(86.9) R29(13)	S35(67.3) R17(32.6)	S12(57) R9(42.8)	S71(80.6) R17(19.3)	S9(90) R1(10)	S3(100) R0(0)	S3(100) R0(0)	S2(100) R0(0)	S0(0) R1(100)	S1(100%) R0(0%)
Imipenem	S148(66.3) R75(33.6)	S32(61.5) R20(38.4)	S14(66.6) R7(33.3)	S66(75) R22(25)	S9(90) R1(10)	S3(100) R0(0)	S3(100) R0(0)	S0(0) R2(100)	S0(0) R1(100)	S1(100%) R0(0%)
Levofloxacin	S 182(81.6) R41(18.3)	S36(69.2) R16(30.7)	S15(71.4) R6(28.5)	S65(75.8) R23(26)	S8(80) R2(20)	S2(66.6) R1(33.3)	S2(66.6) R1(33.3)	S2(100) R0(0)	S1(100) R0(0)	S1(100%) R0(0%)
Linezolid	S184(82.5) R39(17.4)	S40(76.9) R12(23)	S9(42.8) R12(57)	S5(5.6) R83(94.5)	S6(60) R4(40)	S1(33.3) R2(66.6)	S1(33.3) R2(66.6)	S2(100) R0(0)	S0(0) R1(100)	S1(100%) R0(0%)
Meropenem	S164(73.5) R 59(26.4)	S44(84.6) R8(15.3)	S16(76) R5(23.8)	S78(88.6) R10(11.3)	S10(100) R0(0)	S2(66.6) R1(33.3)	S3(100) R0(0)	S2(100) R0(0)	S1(100) R0(0)	S1(100%) R0(0%)
Piparacillin Tazobactam	S177(79.3) R 46(20.6)	S36(69.2) R16(30.7)	S13(61.9) R8(38)	S73(82.9) R15(17)	S10(100) R0(0)	S2(66.6) R1(33.3)	S3(100) R0(0)	S2(100) R0(0)	S1(100) R0(0)	S1(100%) R0(0%)
Vancomycin	S192(86) R31(13.9)	S38(73) R12(23)	S9(42.8) R12(57)	S8(9) R80(90.9)	S4(40) R6(60)	S0(0) R3(100)	S1(33.3) R2(66.6)	S2(100) R0(0)	S0(0) R1(100)	S1(100%) R0(0%)

Table 3: Antibiotic susceptibility pattern of bacterial isolates from blood culture at the Sylhet Womens Medical College & Hospital from April 2019-January 2020.

Multidrug resistance pattern of bacterial isolates

Multidrug resistance strains were common for both Gram-Positive and Gram-negative isolates. Out of 406 isolates 177 (45.5%) were found to

be resistant to more than 7 drugs. Among the isolates 20 (4.9%) were found to be resistant to atleast one antibiotic used in the susceptibility tests and 13(3.2%) were found not to be resistant to any antibiotics used for the susceptibility test.

Isolates	Frequency	Ro (%)	R1(%)	R2(%)	R3(%)	R4(%)	R5(%)	R6(%)	R7(%)
S.aureus	223	5(2.2)	9(4)	18(8)	17(7.6)	30(13.4)	25(11.2)	35(15.6)	84(37.6)
CoNS	52	0	4(7.6)	3(1.3)	4(7.6)	10(19.2)	4(7.6)	5(9.6)	22(42.3)
E.Coli	88	0	0	5(5.6)	11(12.5)	10(11.3)	11(12.5)	9(10.2)	42(47.7)
Pseudomonas	3(100)	0	0	0	0	0	0	0	3(100)
N.meningitits	1(100)	0	0	0	0	0	0	0	1(100)
s.pneumonia	21	1(4.7)	0	1(4.7)	0	0	1	0	18(85.7)
Klebsiella	10	0	0	1(10)	0	1	2(20)	0	6(60)
H.influenza	2	0	0	1(50)	0	0	1(50)	0	0
Salmonella	3	1(33.3)	0	0	1(33.3)	0	0	0	1(33.3)
Enterococcus	1(100)	0	1(100)	0	0	0	0	0	0

Table 4: Multi-drug resistance pattern of bacterial isolates from blood culture at the Sylhet Womens Medical College & Hospital from April 2019-January 2020.

Abbreviation:CoNS, coagulase-negative staphylococci.

Discussion

The aim of this retrospective study was to isolate the most prevalent organism causing blood stream infection and to determine the antimicrobial susceptibility pattern of the isolated pathogens against multiple antibiotics to achieve a clear out look on the changing trend of their antibiotic susceptibility among the patients visiting Sylhet Women's Medical College. The isolation rates of blood culture positive cases were 406(19.4%) which was similar to the study conducted by Nahid et al. in Sylhet and other

studies done in Nepal (20%) and India (20.5%) and Addis Ababa, Ethiopia (21.4%)¹³⁻¹⁵. However our finding is slightly higher than the research done in BSMMU where the isolation rate was 9.88% and two other studies from Bangladesh where recovery rate of bacterial pathogens from blood was 11.6% and 14.3%¹⁶⁻¹⁸. The difference in the blood culture positivity may be due to a variation in the blood culture system, the study design or epidemiological difference in the etiological agents.

Overall male patients suspected with bacteremia were higher (22.47%) in comparison to female patients (16.47%). A study revealed that males are more prone to sepsis than female because of male sex hormone that suppresses cell mediated immune response^[19].

Our findings shows that higher percentage of suspected bacteremia patients were belonging to the aged group <28 Days followed by the age group 29 days to 5 days. This is comparable with the study conducted at University of Gondor, Ethiopia^[15]. This could be explained by the fact that extreme ages are susceptible to infection because of lower immunity.

The most prevalent organisms in all the age group in the current study were staphylococcus aureus followed by E.coli and coagulase negative staphylococci (CoNS). Similar observation were found in a study done by Nahid et al^[13], Swain and Otto^[20], Shahidi et al^[21] and a study conducted in Ethiopia^[15]. But this findings is contradictory to other study done by ABMA wadud^[18] and another study at BSMMU Dhaka^[16] where they found salmonella species as most common isolates. The pattern of every region is unique, However we know that enteric fever is prevalent in Dhaka city due to improper sanitation system leading to gross faecal contamination of consumable water. So higher isolation of Salmonella is generally expected in those area.

In this study Staph. aureus shows intermediate level of resistance to commonly used antibiotic such as Amoxicillin, Cotrimoxazole, Cefazidime and Azithromycin and high level of resistance (>80%) towards Cefixime whereas Amikacin, Gentamicin, Doxycycline and Vancomycin, Piperacillin and Linezolid show a high sensitivity pattern relative to other tested drugs. Similar pattern of sensitivity was shown by Nahid et al and Swain in India^{[13][20]}. Therefore both vancomycin and linezolid might be considered as a good treatment choices

against S.aureus which are resistant to commonly used antibiotics.

E.coli was the second common isolated organism (12.80%). E.coli has been reported as the leading cause of blood stream infection in developed countries. E.coli shows a high level of resistance towards Linezolid and Vancomycin and intermediate level of resistance toward Amoxicillin, Cefuroxime and Cotrimoxazole which is similar to the study conducted at Gonda, Ethiopia and Jimma.^{[15][22]}

The most resistant antibiotics in the current study were Amoxicillin, Cefixime and Cotrimoxazole. The increasing resistance of isolates towards the third generation cephalosporin and Cefixime is alarming. The reason might be due to their huge and irrational use. These types of antibiotics were very commonly used previously in empirical therapy of sepsis and in treatment of other infection.

In this study multiple drug resistance isolates were observed in both Gram-positive and Gram-Negative bacteria. More than 45.5% of the isolate were resistance to more than 7 antibiotics. And only (3.2%) were found to not be resistant to any antibiotics used for the susceptibility test.

Increasing levels of antibiotic resistance are a big concern for Bangladesh. Multiple studies demonstrated that irrational antibiotic prescribing by physicians, a habit of self medication among patients and the indiscriminate use of antibiotics in agriculture and farming in different parts of the country is the cause of high level of antibiotic resistance in our country.^{[23][24][25]}

As there is a shortage of new antibiotics, it is of utmost importance that the existing ones are used cautiously. There is evidence that controlled and lowered use of antibiotics can abate resistance. So it is not yet late if we can still alleviate the use of antibiotics through their rational use, implementing stricter regulation on antibiotic use by hospitals and government as

well as by educating healthcare professionals and the general population. At the same time regular surveillance study needs to be conducted to keep track of the resistance patterns of the pathogens.

Conclusion

Overall prevalence of bacteremia in our study was 19.4%. Among all age groups *Staphylococcus aureus* were predominant. The result of this investigation shows that Gram-positive bacteria prevailed among the isolate and *Staphylococcus aureus* was found to be the highest in number and that is approximately half of the total isolates. The frightening issue is that about 455 of the isolates were resistance to more than seven drugs. Although all the isolates were found to be sensitive with fourth generation cephalosporin, and piperacillin-tazobactam. Routine bacterial surveillance and the study for the baseline drug resistance pattern is simultaneously required to go far in combating drug resistance among pathogens.

Contribution of Authors: Concept, Design of the study and manuscript editing, Data Analysis, Data Collection, Critical review of the manuscript.

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