ORIGINAL RESEARCH

Bacteriological profile of bloodstream infections in critical care units of a tertiary care hospital of central India: Reserve drugs still a threat to our distant future

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ABSTRACT
Introduction: Bloodstream infections (BSI) are leading causes of morbidity and mortality especially in patients admitted to the intensive care unit (ICU). This study aimed to analyze the prevalence and antimicrobial drug resistance patterns of microorganism isolated from blood samples in patients with severe invasive infections admitted in the ICU.
Materials and methods: This prospective study conducted in the department of microbiology from January 2023 to December 2023 at a tertiary care teaching hospital incentral India. Identification of the microorganism done using standard methods and antimicrobialsusceptibility testing conducted as per Clinical Laboratory Standards Institute (CLSI) guidelines.
Results: A total of 600 blood culture sample were received in the microbiology laboratory, of which 300 were culture positive and 220 were sterile and 80 were skin contaminants. *Staphylococcus aureus* was most common organism isolated among Gram positive cocci accounting for 43(14.3%) and among the Gram negatives, the most common organism isolated was *Acinetobacter baumannii 45(15%). Staphylococcus aureus and Enterococcus Faecalis* showed 100% susceptibility to reserve drug Vancomycin where as *Enterococcus faecium* showed 90.9% susceptibility to Vancomycin. 12% of *Klebsiella pneumoniae,* 6.7% of *Pseudomonas aeruginosa* and 4.5% of *Acinetobacter baumannii* were resistant to reserve drug Colistin where as *Escherichia coli* and *Stenotrophomonas maltophilia* showed 100% colistin susceptibility.
Conclusion: Gram negative bacteraemia (60.7%) were more frequently isolated than Gram positive bacteraemia (39.3%). Gram-negative bacteriahave the highest drug resistance rateamong the bloodstream infection strains in ICU.In order to prevent or reduce the establishment of antibiotic resistance, it is important to rationalize the use of antibiotics and improve infection control strategies, as 42% of the blood stream organisms isolated were multidrug resistant.
Keywords: bloodstream infections, multidrug resistant, antibiotic, intensive care unit.

INTRODUCTION

Bloodstream infections (BSIs) are one of the most common acquired illnessesamong the patients admitted in intensive care unit (ICU)(1).Formally, a bacterial invasion of the normally sterile bloodstream is referred to as a BSI. Clinically, a combination of appropriate clinical symptoms and positive blood culture results in BSI(2). Hospital acquired blood stream infections (BSI's) accounts for about 35-50% among patients admitted in ICU (3).
Globally, antimicrobial resistance (AMR) is a serious danger to public health(4).

The 2019 Centers for Disease Control and Prevention (CDC) Antibiotic Resistance Threats in the United States Report states that from 2012 through 2017, antimicrobial-resistant bacteria caused over 2.8 million illnesses and over 35,000 deaths yearly(5)..
According to The World Health Organization (WHO) in its ; most recent model list of essential medicines,Reserve drug includes fourth and fifth-generation cephalosporins, polymyxins, aztreonam, and linezolid to be used when all other alternatives have failed(6).
The polypeptide antibiotic colistin targets Gram-negative bacteria's outer membrane. Because of the scarcity of novel antimicrobial agents and the rise in

the number of bacteria that are highly resistant to drugs, it is regarded as a last-resort antibiotic. It is specifically used to treat infections caused on by *Pseudomonas aeruginosa, Acinetobacter baumannii*, and *Klebsiella pneumoniae* (7). Linezolid is a synthetic oxazolidinone antimicrobial agent. It is indicated for Gram-positive infections and permitted for the treatment of bacterial pneumonia, vancomycin-resistant enterococcal (VRE) infections, and skin structure infections (8).

Clinician should be updated about the most prevalent resistant species found in various geographic locations, as well as their mechanisms of resistance, also be informed of the primary risk factors for bloodstream infections (BSIs) caused by resistant bacteria in the ICU. The purpose of this study is to give clinicians a current understanding of BSI in ICU patients that is brought on by resistant bacteria.

MATERIALS & METHOD

This prospective, study was conducted in the department of microbiology of a tertiary care teaching centre of central India for a duration of 12 months after institutional approval for the study was taken.

Blood samples for blood culture were collected taking all aseptic precautions and transferred to conventionally prepared blood culture bottles containing BHI broth and transported to the aerobic section of the bacteriology. After overnight incubation at 37 °C samples were sub-cultured on to 5% sheep blood agar, MacConkey agar and chocolate agar to look for any growth. Identification of the *organism* was done by Gram staining, observing colony morphology, pigment production and standard biochemical tests.

Blood culture bottles showing no signs of any growth after 5 days of aerobic incubation, either growth on 5% sheep blood agar, MacConkey agar or haemolysis/turbidity were reported as sterile after a final confirmatory subculture.

The antimicrobial susceptibility testing of bacterial isolates was done by the Kirby-Bauer disk diffusion method, and minimal inhibitory concentration (MIC) of vancomycin and colistin was determined by the broth microdilution method (gold standard)(9). Interpretation of antibiotic susceptibility testing was done as per the Clinical and Laboratory Standards Institute (CLSI) interpretative criteria (M100; 33rd Edition)(10). *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 were used as the quality controls for disc diffusion testing.

RESULTS

A total of 600 blood culture sample were received in the microbiology laboratory, of which 300 were culture positive and 220 were sterile and 80 were skin contaminants. Most of the blood culture positive isolates were from medical ICU followed by neonatal ICU and then surgical ICU. Majority of the isolates were from age group 45-65 years (29.6) and least were from age group 1-18 years (14%).

Table 1: Demographic and clinical details of patient admitted.

Parameter	% distribution (number of cases) Total number of cases=
Age	
0-1 years	50(16.6%)
1-18 years	42(14%)
18-45 years	56(18.6%)
46-65 years	89(29.6%)
>65 years	63(21%)
Gender	
Male	173(57.6%)
Female	127(42.3%)
Primary diagnosis	
Diabetes mellitus	59(19.6%)
Hypertension	56(18.6%)
Myocardial infarction	28(9.3%)
Congestive cardiac failure	30(10%)
Chronic obstructive lung disease	24(18%)
Chronic kidney disease	12(4%)
End stage renal disease	9(3%)
Infective endocarditis	15(5%)
Sepsis	30(10%)
Preterm	15(5%)
Failure to thrive	10(3.3%)
Hypoxic Ischemic encephalopathy	12(4%)
Location of patient	
Medical ICU	126(42%)
Surgical ICU	76(25.3%)

Neonatal ICU	98(32.6%)
Mean duration of stay in the hospital (among admitted patients)	5.3 days

Table 2: Various organism isolated from blood stream infections

Organism isolated	Cases(% isolated)
Gram positive	
Staphylococcus aureus	43(14.3%)
Coagulase negative Staphylococcus(CONS)	28(9.3%)
Enterococcus faecium	22(7.3%)
Enterococcus faecalis	25(8.3%)
Total Gram positive	**118(39.3%)**
Gram Negative	
Escherichia coli	18(6%)
Klebsiella pneumoniae	25(8.3%)
Klebsiella oxytoca	12(4%)
Pseudomonas aeruginosa	30(10%)
Acinetobacter baumannii	45(15%)
Salmonella	10(3.3%)
Proteus mirabilis	17(5.6%)
Proteus vulgaris	14(4.6%)
Stenotrophomonas maltophilia	8(2.6%)
Burkholderia cepacia	3(1%)
Total Gram negative	**182(60.7%)**

Staphylococcus aureus was most common organism isolated among Gram positive cocci accounting for 43(14.3%) and among the Gram negatives, the most common organism isolated was Acinetobacter baumannii 45(15%).

Table 3:-Antibiotic susceptibility profile of Gram positive organism isolated from blood stream infections as per as CLSI

Antimicrobials	*Staphylococcus aureus*	*Enterococcus faecium*	*Enterococcus faecalis*
Erythromycin(15)	48.8%	36.3%	36%
Clindamycin (2)	46.5%	-	-
Cefoxitin(30)	34.8%	-	-
Ciprofloxacin(5)	32.5%	50%	60%
Gentamicin(10)	81.3%	-	-
Tetracycline(30)	65%	72.7%	68%
Doxycycline(30)	58.1%	72.7%	68%
Linezolid(30)	100%	100%	100%
Penicillin(10)	34%	54.5%	60%
Cotrimoxazole (25)	55.8%	-	-
Vancomycin (MIC)	100%	90.9%	100%

Staphylococcus aureus and *Enterococcus Faecalis* showed 100% susceptibility to reserve drug Vancomycin where as *Enterococcus faecium* showed 90.9% susceptibility to Vancomycin.

Table 4:- Antibiotic susceptibility profile of Antibiotic susceptibility profile of Gram negative organism isolated from blood stream infections as per as CLSI

Antimicrobials (mcg)	*Escherichia coli*	*Klebsiella pneumonaie*	*Klebsiella oxytoca*	*Pseudomonas aeruginosa*	*Acinetobacter baumannii*	*Salmonella*	*Proteus vulgaris*	*Proteus mirabilis*	*Stenotrophomonas maltophilia*	*Burkholderia cepacia*
Amikacin (30)	72.2%	44%	33.3%	50%	62.2%	-	58.8%	64.7%	-	-
Ciprofloxacin (5)	44.4%	20%	25%	26.6%	40%	50%	21.4%	17.6%	-	-
Gentamicin (10)	66.6%	44%	33.3%	56.6%	66.6%	-	64.2%	52.9%	-	-

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Antibiotic										
Ceftazidime (30)	22.2%	16%	16.6%	10%	20%	-	14.2%	11.7%	-	33.3%
Cefotaxime (30)	38.8%	36%	25%	-	37,7%	-	35.7%	23.5%	-	-
Ceftriaxone (30)	44.4%	36%	25%	-	37.7%	-	42.8%	35.2%	-	-
Cefepime (30)	55.5%	40%	33.3%	30%	51.1%	-	57%	47%	-	-
Doxycycline (30)	44.4%	24%	25%	-	46.6%	-	-	-	-	-
Piperacillin-tazobactam (100/10)	61.1%	48%	41.6%	43.3%	46.6%	-	70.5%	78.5%	-	-
Cotrimoxazole (25)	38.8%	24%	25%	-	20%	70%	35.7%	29.4%	62.5%	33.3%
Minocycline(30)	-	-	-	-	84.4%	-	-	-	75%	100%
Meropenem (10)	88.8%	60%	58.3%	83.3%	75.5%	90%	85.7%	82.3%	-	66.6%
Levofloxacin (5)	50%	32%	33.3%	30%	48.8%	60%	42.8%	41%	37.5%	-
Colistin MIC ≤ 2 µg/mL (intermediate)	100%	88%	91.6%	93.3%	95.5%	-	-	-	100%	-

Gram negative organism isolated from blood stream infections showed good susceptibility to the reserve drug Colistin where as least susceptibility to Ceftazidime.

DISCUSSION
Nosocomial BSIs are linked with a significant morbidity and mortality[11]. Due to their weakened state from underlying disease and the frequent invasive diagnostic and therapeutic procedures, patients in intensive care units are at a heightened risk of nosocomial bloodstream infections[12]. Our research sheds light on the patterns of antibiotic resistance in frequently isolated microbes seen in central Indian intensive care unit patients. The scenario is distressing and clearly illustrates that drug resistance is on the rise and clinicians are left with very few alternatives for treating patients with severe illnesses in the ICU.Depending upon the locationof patients within the institution,different organisms can cause nosocomial bloodstream infections[13].

A total of 600 blood culture sample were received in the microbiology laboratory, of which 300 were culture positive and 220 were sterile and 80 were skin contaminants. Most of the blood culture positive isolates were from medical ICU followed by neonatal ICU and then surgical ICU which is in similar to study conducted by M.A. Ababneh et al. where most of the blood stream infections were from medical ICU's [14].

The demographic data from this study indicated that majority of the isolates were from age group 45-65 years (29.6) and least were from age group 1-18 years (14%) which is incordance with study conducted by Kaur & Sharma where highest positive blood culture results were from patients between theages of 46–60 years, followed by the ages 0–15years[15].

In the present study,Gram negative bacteraemia(60.7%) were more frequently isolated than Gram positive bacteraemia (39.3%), which is similar to study conducted by Agrawal et al.'s in whichbacteraemia by Gram negative and Gram positive was found to be 68.35% and 31.65%, respectively[16].

Staphylococcus aureus was most common organism isolated among Gram positive cocci accounting for 43(14.3%) followed by *Coagulase negative Staphylococcus(CONS)*28(9.3%), *Enterococcus faecalis*25(8.3%) and the *Enterococcus faecium*22(7.3%). A study conducted by Golli et al demonstrated similar finding that Staphylococcus aureus was most frequently isolated among Gram positive bacteraemia [17]. *Methicillin resistant Staphylococcus aureus* was found to be 65.2% .The high frequency of MRSA could be attributed to numerous risk factors, including the transmission of MRSA by patients and healthcare professionals, improper use and abuse of antibiotics, and extended hospital stays.

Staphylococcus aureus and Enterococcus Faecalis showed 100% susceptibility to reserve drug Vancomycin where as *Enterococcus faecium* showed 90.9% susceptibility to Vancomycin. A study conducted by Trivedi MS et al. also found 100% susceptibility of *Staphylococcus aureus* to Vancomycin(3). All the Gram positive cocci showed 100% susceptibility to Linezolid.

Among the Gram negative organism *Acinetobacter baumannii* was most frequently isolated accounting 15% bacteraemia followed by *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* 10% and 8.6% respectively. Gram negative organism showed good susceptibility to colistin where as least susceptibility to Ceftazidime.

12% of *Klebsiella pneumoniae,*6.7% of *Pseudomonas aeruginosa and 4.5% of Acinetobacter baumannii*were resistant to reserve drug Colistin where as *Escherichia coli* and *Stenotrophomonas maltophilia* showed 100% colistin susceptibility .As a last resort, colistin is used to treat serious infections brought on by drug resistant bugs. The usage of colistin has grown by almost ten times over time . Resistance has increased as a result of greater colistin usage(18).

In order to prevent or reduce the establishment of antibiotic resistance, it is important to rationalize the use of antibiotics and improve infection control strategies, as 42% of the blood stream organisms isolated were multidrug resistant.As a last resort, polymyxins are being employed more and more. Colistin can be utilized in combination with other antimicrobials to treat multidrug resistant circumstances(6).

CONCLUSION

The majority of the bacteria recovered from BSI patients were Gram-negative. Clinicians in every location must stay up to speed on the most recent prevalence and antibiotic susceptibility pattern of the circulating pathogens due to the dramatic increase of MDR Gram-negative bacilli, particularly causing blood stream infection in Intensive care unit.

Frequent monitoring of isolate sensitivity trends, the development of hospital antibiotic policies based on available information, and adherence to treatment protocols can encourage sensible antibiotic usage and lessen the emergence of bacterial resistance. To make sure that colistin is still a viable therapeutic option, it is critical to evaluate and createrecommendations for its usage.

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