



Original Research Article

BACTERIOLOGICAL PROFILE OF BLOOD STREAM INFECTIONS AND ITS ANTIBIOGRAM PATTERN AT A TERTIARY CARE HOSPITAL- A RETROSPECTIVE STUDY

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ABSTRACT

Background: Bloodstream infections (BSIs), also known as septicemia or bacteremia can occur when microorganisms enter the bloodstream and cause an infection. Emergence of resistance among the bacterial pathogens causing these infections is another issue of the public health concern. Therefore, this study aims to determine the bacteriological profile of blood stream infections along with its antibiogram pattern, to know the trends of resistance among these agents.

Materials and Methods: A retrospective study was conducted from July 2023 to June 2024 in the Department of Microbiology, Hind institute of medical sciences, Sitapur. A total of 984 blood samples with different age groups and gender were included in the study. Blood samples were collected under aseptic precaution and identified using standard protocol, which includes Gram's staining, Motility Testing, Catalase test, Oxidase test, Biochemical reactions, Sugar fermentation tests and OF tests. The antibiotic susceptibility testing was done by Kirby bauer disc diffusion method as per CLSI guidelines.

Results: From a total of 175 isolates, the predominant pathogen isolated was *S.aureus* (51.4%) followed by *P.aeruginosa* (18.2%), *SOSA* (9.7%), *Klebsiella sp* (6.2%), *Acinetobacter sp* (5.1%), *Enterococcus sp* (5.1%), *Citrobacter sp* (2.2%) and *E.coli* (1.7%). *S.aureus*, *SOSA* and *Enterococcus sp* showed good sensitivity to Vancomycin, Linezolid and Teicoplanin. *Pseudomonas aeruginosa* and *Acinetobacter sp* showed maximum sensitivity to Meropenem, Imipenem, Polymyxin B, Colistin, Ceftazidime, Piperacillin tazobactam, and Amikacin. *Klebsiella sp* showed good sensitivity to Meropenem and Imipenem, Gentamycin, Piperacillin tazobactam and Cefipime.

Conclusion: Regular epidemiological studies of BSIs, regarding the pathogens and their antibiotic susceptibility patterns are necessary to guide clinicians in choosing appropriate empirical therapy and to update the hospital antibiotic policy from time to time. This promotes rational antibiotic use and reduces resistance among bacteria.

Keywords: Blood stream infections, Antibiotic susceptibility, Antibiotic resistance, *S.aureus*, Polymyxin B, Meropenem.

INTRODUCTION

Bloodstream infections (BSIs), also known as septicemia or bacteremia can occur when bacteria or other microorganisms enter the bloodstream and cause an infection.^[1,2] Blood stream infections are among the most significant infections that cause morbidity and mortality among hospitalised patients worldwide.^[3] Approximately 200,000 cases of bacteraemia occur annually with a mortality rate ranging from 20% to 50% worldwide.^[4]

The most common bacterial genera responsible for BSIs are members of *Enterobacteriaceae*, *Staphylococcus*, *SOSA*^[5,6] and other non-fermentative gram-negative bacilli such as *Pseudomonas sp* and *Acinetobacter sp*^[7,8] Severe sepsis is a life threatening condition characterized by shock, disseminated intravascular coagulation and acute renal failure.^[9,10]

Blood culture is a vital tool for the detection of BSI and remains the gold standard for bacteremia detection.^[11] Appropriate initiation of antimicrobial therapy is important to reduce morbidity and mortality rates.^[12,13] Empiric antimicrobial therapy is based on knowledge of the microbial profile and their antimicrobial sensitivity patterns, clinical and epidemiological data.^[11,12,14] Therefore, the present study was undertaken to determine the bacteriological profile of blood stream infections and its antibiogram pattern at a tertiary care hospital in Uttar Pradesh, India.

MATERIALS AND METHODS

Study design: This is a retrospective laboratory record based study, carried out at Department of Microbiology, HIMS, Sitapur from July 2023 to June 2024. A total of 984 blood samples were included in this study. Approval from the Institutional Ethical Committee (IEC) was obtained on 11.9.24 with document no SOP/11.12-N/A.

Inclusion Criteria: All blood cultures received in the Department of Microbiology, HIMS, Sitapur irrespective of age group and gender, attending both outpatient and inpatient departments were included in the study.

Exclusion Criteria: Blood culture samples yielding fungal growth and contaminants were excluded from the study.

Methodology:

Specimen collection:^[15]

Blood samples were collected under aseptic precautions before the administration of antibiotics. A sterile needle and syringe were used to draw 5 mL to 10 mL of blood for adults, 2 mL to 5 mL for children and 1 mL to 2 mL for neonates which were then directly inoculated into aerobic and anerobic blood culture bottles of BACTEC at a ratio of 1:5 to 1:10 and incubated aerobically at 37°C for 18-24hrs.

Antibiotic susceptibility testing of the isolates:^[15,16]

Antibiotic susceptibility was determined using the Kirby Bauer disk diffusion method based on the Clinical and Laboratory Standards Institute (CLSI) guidelines. Mueller Hinton agar and commercially available 6 mm antimicrobial disks (HiMedia Laboratories Pvt., Ltd., Mumbai, India) were used.

Gram positive isolates: Penicillin (P-10 units), Erythromycin (E-15 µg), Trimethoprim-sulfamethoxazole (COT-1.25/23.75 µg), Ciprofloxacin (CIP-5 µg), Linezolid (LZ-30 µg), Tetracycline (TE-30 µg), Ampicillin (AMP-10 µg), Vancomycin (VA-30 µg), Teicoplanin (TEI-30 µg), High-Level Gentamicin (HLG-120 µg), and High-Level Streptomycin (HLS-300 µg).

Gram negative isolates: Amoxicillin-clavulanate (AMC-20/10 µg), Cefotaxime (CTX-30 µg), Ceftriaxone (CTR-30 µg), Levofloxacin (LE-5 µg), Imipenem (IMP-10 µg), Gentamicin (GEN-10 µg), Amikacin (AK-30 µg), Piperacillin-tazobactam (PIT-100/10 µg), and Cefepime (CPM-30 µg).

Vancomycin susceptibility testing for staphylococcal isolates was performed using the Epsilometer test (E-test) with Vancomycin Ezy MIC™ Strip (VAN) having concentrations ranging from 0.016 to 256 µg/mL (HiMedia Laboratories Pvt., Ltd., Mumbai, India), following the CLSI guidelines.

Methicillin resistance among staphylococcal isolates was detected using the cefoxitin disk (CX-30 µg) diffusion method as per CLSI guidelines.

Extended spectrum β-lactamase production in *K. pneumoniae* and *E. coli* isolates was determined by the double disk diffusion method using Cefotaxime (CTX-30 µg), and Cefotaxime-clavulanate (CEC-30/10 µg) disks.

Quality control: The reference ATCC strains used were *S. aureus* (ATCC 25923), *E. coli* (ATCC 25922), *K. pneumoniae* (ATCC 27736), *E. faecalis* (ATCC 29212) and *P. aeruginosa* (ATCC 27853).

Statistical Analysis: Analysing the distribution of different bacterial isolates, sensitivity pattern and the interpretation of results was done using SPSS software version 23.

RESULTS

A total of 984 blood culture samples were collected from various patients during the study period. Among these 597 samples (61%) were from males and 387 samples (39%) were from females. [Figure1]

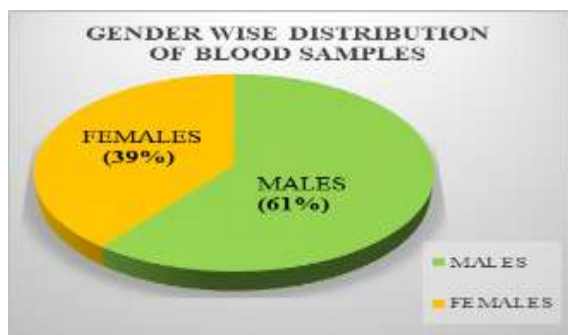


Figure 1: Gender wise distribution of blood samples (n=984)

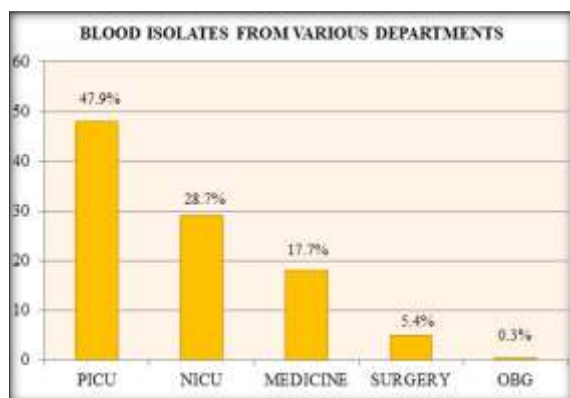


Figure 2: Department wise distribution of blood samples (n=984)

In our study, maximum number of blood samples were received from PICU (47.9%) followed by NICU (28.6%), Medicine (17.6%), Surgery (5.4%) and OBG (0.3%). [Figure 2]

Among the age group distribution, maximum number of samples were received from 0-10 years age group (46.3%) followed by 11-20years (11.7%). Patients more than 60 years of age showed minimum incidence (3.2%). [Table 1]

Out of total 175 isolates, *S.aureus* 90 (51.4%) was the predominant isolate followed by *Pseudomonas aeruginosa* 32 (18.2%), *SOSA* 17(9.7%), *Klebsiella sp* 11 (6.2%), *Acinetobacter sp* 9 (5.1%), *Enterococcus sp* 9 (5.1%), *Citrobacter sp* 4 (2.2%), and *E.coli* 3 (1.7%). [Table 2]

S.aureus showed good sensitivity to Vancomycin (88%), Linezolid (92%) and Teicoplanin (97%)

followed by Gentamycin (70%) and Clindamycin (65%). *SOSA* showed 90% sensitivity to Vancomycin, 100% sensitivity to Linezolid and Teicoplanin followed by Gentamycin (59%). *Enterococcus sp* showed maximum sensitivity to Vancomycin (90%), Linezolid (100%) and Teicoplanin (100%) followed by High level streptomycin (86%) and High level gentamycin (72%).[Table3]

Among all the **Gram positive isolates**, maximum resistance was recorded for Penicillin (20%) and Erythromycin (40%). [Table3]

Methicillin resistant *S.aureus*:

Out of 90 *S.aureus* isolates, 49 (55%) were *MRSA* and among 17 *SOSA* isolates, 9 (53%) were MR-*SOSA*. [Table 3]

Pseudomonas aeruginosa showed good sensitivity to Polymyxin B (100%), Colistin (100%), Meropenem and Imipenem (94%) followed by Ceftazidime (81%) Amikacin (76%) and Piperacillin tazobactam (76%). *Klebsiella sp* showed 82% sensitivity to Meropenem and Imipenem followed by 73% sensitivity to Gentamycin, Piperacillin tazobactam and Cefipime. *Acinetobacter sp* showed 100% sensitivity to Polymyxin B and Colistin followed by 88% sensitivity to Meropenem and Imipenem (Table: 6). *Citrobacter sp* showed 100% sensitivity to Ceftazidime and Cefipime followed by 75% sensitivity to Cefotaxime, Ceftriaxone, Gentamycin, Tobramycin and Fluroquinolones. *E.coli* showed 100% sensitivity to Piperacillin tazobactam, Meropenem, Imipenem and Cefipime followed by 66% sensitivity to Cefotaxime, Ceftriaxone, Gentamycin, and Amikacin.

ESBL producers

Out of 59 Gramnegative isolates, 26 (44%) were ESBL producers. Highest ESBL production in our study was observed in *Klebsiella pneumoniae* (55%) followed by *Escherichia coli* (45%). [Table 4]

MBL producers:

Among 59 Gramnegative isolates, 13 (22%) were MBL producers. Highest MBL production was seen with *Klebsiella pneumoniae* (40%) followed by *Pseudomonas sp* (30%) and *Acinetobacter sp* (30%). [Table 4]

Table 1: Age wise distribution of blood samples (n=984)

Age	No. of Samples	Percentage (%)
0-10	456	46.3%
11-20	110	11.1%
21-30	116	11.7%
31-40	107	10.8%
41-50	97	9.8%
51-60	66	6.7%
>60 years	32	3.2%

Table 2: Isolates from Blood Samples (n=175)

Organism	No. of Isolates	Percentage (%)
<i>S.aureus</i>	90	51.4
<i>Pseudomonas aeruginosa</i>	32	18.2
<i>SOSA</i>	17	9.7
<i>Klebsiella sp</i>	11	6.2
<i>Acinetobacter sp</i>	9	5.1
<i>Enterococcus sp</i>	9	5.1
<i>Citrobacter sp</i>	4	2.2
<i>E.coli</i>	3	1.7

Table 3: Antimicrobial Susceptibility Pattern of Blood Isolates- Gram positive isolates (n=116)

ANTIBIOTICS	<i>S.aureus</i> (n=90)		<i>SOSA</i> (n=17)		<i>Enterococcus sp</i> (n=9)	
	S	%	S	%	S	%
Penicillin (P)	20	22%	3	18%	3	33%
Ampicillin (AMP)	NT	NT	NT	NT	3	32%
Erythromycin (E)	41	46%	6	35%	4	44%
Gentamycin (GEN)	63	70%	10	59%	NT	NT
Cefoxitin (CX)	41	45%	8	47%	NT	NT
Tetracycline (TE)	50	55%	9	53%	NT	NT
Clindamycin (CD)	59	65%	10	59%	NT	NT
Azithromycin (AZM)	36	40%	6	35%	NT	NT
Ciprofloxacin (CIP)	56	62%	13	76%	NT	NT
Trimethoprim sulfamethoxazole	47	52%	8	47%	NT	NT
Vancomycin (VA)	79	88%	15	90%	8	90%
Linezolid (LZ)	83	92%	17	100%	9	100%
Teicoplanin (TEC)	88	97%	17	100%	9	100%
High level Gentamycin (HLG)	NT	NT	NT	NT	6	72%
High level Streptomycin (HLS)	NT	NT	NT	NT	7	86%

❖ NT- Not tested

❖ n =Total no of isolates tested

Table 4: Antimicrobial susceptibility pattern of blood isolates- Gram negative isolates (n= 59)

ANTIBIOTICS	<i>P.aeruginosa</i> (n=32)		<i>Klebsiella sp</i> (n=11)		<i>Acinetobacter</i> <i>sp</i> (n=9)		<i>Citrobacter sp</i> (n=4)		<i>E.coli</i> (n=3)	
	S	%	S	%	S	%	S	%	S	%
Amikacin (AK)	24	76%	6	55%	7	78%	3	75%	2	66%
Gentamycin (GEN)	16	52%	8	73%	6	67%	3	75%	2	66%
Amoxycillin clavulanate (AMC)	NT	NT	0	0%	NT	NT	0	0%	1	33%
Piperacillin tazobactam (PIT)	24	76%	8	73%	6	67%	2	50%	3	100%
Tetracycline (TE)	NT	NT	5	45%	NT	NT	3	75%	1	33%
Trimethoprim sulfamethoxazole	NT	NT	5	45%	4	55%	2	50%	0	0%
Cefotaxime (CTX)	NT	NT	6	55%	5	55%	3	75%	2	66%
Ceftriaxone (CTR)	NT	NT	6	55%	5	55%	3	75%	2	66%
Ceftazidime (CAZ)	26	81%	7	64%	5	55%	4	100%	2	66%
Cefipime (CPM)	4	12%	8	73%	6	67%	4	100%	3	100%
Imipenem (IMP)	30	94%	9	82%	8	88%	2	50%	3	100%
Meropenem (MRP)	30	94%	9	82%	8	88%	2	50%	3	100%
Ciprofloxacin (CIP)	26	82%	4	36%	5	55%	3	75%	2	66%
Levofloxacin (LE)	26	82%	4	36%	5	55%	3	75%	2	66%
Aztreonam (AT)	NT	NT	6	55%	NT	NT	2	50%	2	66%
Tobramycin (TOB)	22	70%	7	64%	6	67%	3	75%	1	33%
Colistin	32	100%	NT	NT	9	100%	NT	NT	NT	NT
Polymyxin B	32	100%	NT	NT	9	100%	NT	NT	NT	NT

DISCUSSION

There is a strong association between delays in initiating effective therapy and in-hospital mortality in cases of septic shock.^[17] Each hour of delay in therapy initiation is associated with an average decrease in survival of 8%.^[18]

In this study, Out of 984 blood samples processed, 175 (17%) blood cultures had bacterial growth which is similar to the studies conducted by Qureshi et al, Mehta MP et al and Vijaya Devi et al who reported a culture-positive rate of 16.6%, 16.4%, and 16.8% respectively.^[19,20,21] Variation in culture positivity rates could be due to epidemiological

variance of the etiological causes and volume of blood culture samples.

In our study, 61% samples were from males and 39% samples were from females which is similar to the study conducted by Gupta et al (Table:1).^[22] The maximum number of samples were received from 0-10 years of age (46.3%) followed by 11-20 years (11.1%) and 21-30 years (11.7%), which was similar to the studies reported by Devendra Kumar Tiwari et al,^[23] and Vidhya R et al,^[24] where the maximum incidence of blood stream infections were seen in ages between 0-10 years (35.71%). The high number of BSI cases among paediatric patients may be due to their susceptibility to infection owing to

their developing innate and adaptive immune systems.^[25]

The maximum number of BSI cases was observed in NICU (47.9%) followed by PICU (28.7%). These findings are compatible with the studies conducted by Arora U et al., and Bhabhor H et al.^[26,27] Out of 175 total isolates, 116 (66.2%) were Gram positive isolates followed by 59 (33.7%) Gram negative isolates which had also been reported in other studies.^[28,29,30]

Among the gram-positive isolates, the predominant isolate was *S. aureus* 90 (51.4%), followed by *SOSA* 17 (9.7%) and *Enterococcus sp* 9 (5.1%). Other studies have also reported *S. aureus* as the predominant bloodstream pathogen.^[25,31] Since *SOSA* are a part of the normal skin flora, the clinical significance can be determined by repeated positive blood cultures and the presence of long-standing indwelling devices such as central venous catheters, intravascular catheters, etc.^[32,33]

Among the Gram negative isolates in our study, *Pseudomonas aeruginosa* 32 (18.2%) was the predominant isolate followed by *Klebsiella sp* 11 (6.2%), *Acinetobacter sp* 9 (5.1%) which is similar to the findings reported by other studies.^[34,25]

In the present study, *S.aureus* showed maximum sensitivity to Vancomycin (88%), Linezolid (92%) and Teicoplanin (97%) followed by Gentamycin (70%) and Clindamycin (65%). *SOSA* showed 90% sensitivity to Vancomycin, 100% sensitivity to Linezolid and Teicoplanin followed by Gentamycin (59%), which correlates with the other studies conducted by Kumar et al, Nazir et al and Sharma et al where they showed 100% sensitivity to Linezolid and Vancomycin.^[25,35,36] In our study the prevalence of *MRSA* was 55% which is similar to the study conducted by Pal N et al.^[37]

In this study, *Enterococcus sp* showed maximum sensitivity to Linezolid (100%) and Teicoplanin (100%) followed by Vancomycin (90%), High level streptomycin (86%) and High level gentamycin (72%) which is similar to the findings of Yangzom T et al.^[38]

In this study, the antimicrobial susceptibility pattern of *P.aeruginosa* showed good sensitivity to Polymyxin B (100%), Colistin (100%), Meropenem and Imipenem (94%) followed by Ceftazidime (81%) and Piperacillin tazobactam (76%). *Acinetobacter sp* showed 100% sensitivity to Polymyxin B and Colistin followed by 88% sensitivity to Meropenem and Imipenem. Similar patterns of the non-fermenters were also observed in other studies conducted by Nautiyal et al.^[39,40]

Klebsiella sp showed 82% sensitivity to Meropenem and Imipenem followed by 73% sensitivity to Gentamycin, Piperacillin tazobactam and Cefipime. About 55% of *K. pneumoniae* isolates were ESBL producers, which is similar to the findings of Pal N et al.^[37] In our study, highest MBL production was seen with *Klebsiella pneumoniae* (40%) followed by *Pseudomonas sp* (30%) and *Acinetobacter sp* (30%)

which is comparable to the studies conducted by Oberoi et al and Rao et al.^[41,42]

CONCLUSION

The epidemiology and outcome of Blood stream infections are evolving due to the increased rate of antimicrobial resistance and changing patterns of antimicrobial usage worldwide. Implementing antibiotic consumption approaches such as restricting antibiotic use, combination therapy and antibiotic use based on antimicrobial susceptibility testing results can reduce the occurrence of BSIs and prevent resistance.

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