IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 24, Issue 4 Ser. 2 (April. 2025), PP 10-16
www.iosrjournals.org

Bacterial Etiology Of Bloodstream Infections And Antimicrobial Resistance Patterns From A Tertiary Care Hospital Of Jaipur, Rajasthan

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Abstract
Background: *Bloodstream infections significantly raise morbidity and death, especially in hospitalized patients. Antimicrobial resistance complicates treatment. The identification of pathogenic organisms and the resistance patterns they display are essential for effective management and prevention in tertiary care settings.*
Methods: *Blood culture samples were collected from hospitalized patients at tertiary care facilities and processed in the Bacteriology Laboratory, Department of Microbiology, NIMS&R, Jaipur.*
Results: *The majority of the 125 blood culture isolates came from patients who were aged 51-60 years (22.4%) followed by 61-70 years (16%), 41-50 years (14.4%), 31-40 years (14.4%), 71-80 years (12.0%), 21-30 years (12.0%), 17-20 years (7.2%), and 81-90 years (1.6%). In this study gram-negative organism, Escherichia coli (34.40%) was the predominant isolate from patients with bloodstream infection followed by Klebsiella pneumoniae (29.50%), Acinetobacter baumannii (19.70%), Pseudomonas aeruginosa (11.50%), Enterobacter cloacae (3.30%), and Serratia marcescens (1.60%), it is observed that highest resistance against Ampicillin (52%) followed by Cephalosporins (45-51%), and Aminoglycosides (33-36%), while highest sensitive antimicrobials were colistin (100%), followed by Minocycline (90.2%), and Tigecycline (87.70%) against gram-negative bacterial isolates.*
Conclusions: *This study found that bacterial pathogen resistance to commonly used antibiotics for treating bloodstream infections. With Gram-positive bacteria being more common (51.2%) than Gram-negative (48.8%). These findings pose a significant risk to septicemia. Further research and evaluations are needed to reduce the number of blood infection patients burden. This study emphasizes the significance of regular surveillance, antiseptic adherence, infection control and rational antibiotic policy for early diagnosis and better management of bloodstream infections.*
Keywords: *BSI, Drug resistance, S. aureus, Antimicrobial resistance pattern, Gram-positive bacteria, Gram-negative bacteria*

Date of Submission: 26-03-2025 Date of Acceptance: 06-04-2025

I. Introduction

Bloodstream infections (BSIs) are broadly defined as the presence of live bacteria in the bloodstream, which can induce host inflammation, alter clinical and hemodynamic parameters, and result in severe health consequences [1]. BSIs represent a significant global health burden, affecting approximately 30 million individuals annually and causing 6 million deaths worldwide [2]. Notably, 3 million newborns and 1.2 million children suffer from sepsis every year [3]. Even transient or occasional microbial presence in the bloodstream poses substantial risks to vital organs [4]. Left untreated, BSIs can progress to life-threatening complications, including shock, Disseminated Intravascular Coagulation (DIC), multiple organ failure, and ultimately death. These infections are associated with high morbidity and mortality rates, underscoring their critical impact on global public health [5].

The presence of bacteria in the bloodstream is one of the most critical and severe circumstances in infectious diseases, as microorganisms, whether present persistently or transiently, threaten all major organs [6]. The prevalence and antimicrobial susceptibilities of microorganisms vary depending on environmental factors and antibiotic usage. BSIs can lead to complications such as shock, diffuse intravascular coagulation, and organ failure, as well as extended hospital stays and increased healthcare costs [7]. Bloodstream infections are classified as community-acquired bacteremia (CAB) if the initial positive blood culture is obtained before or within 48 hours of admission [8]. Conversely, they are considered nosocomial when symptoms appear after 48 hours of hospitalization or if the patient has been recently hospitalized [9].

Common bacterial isolates from ICU patients include *Staphylococcus aureus*, *Enterococcus*, and gram-negative aerobic bacteria such as *Enterobacteriaceae* and *Pseudomonas aeruginosa*. Additionally, *Candida albicans* is a frequently isolated fungal pathogen in both immunocompetent and immunocompromised individuals. Coagulase-negative *Staphylococcus* species (CoNS), once dismissed as contaminants, are now recognized as clinically significant pathogens [10]. Patients with catheters or vascular prostheses are particularly susceptible to catheter-associated bacteremia, complicating therapeutic management [11].

Under normal conditions, blood is a sterile environment, and the detection of microorganisms through blood cultures is uncommon unless there is an active infection. While sepsis is the result of a host's systemic inflammatory response to infection, BSIs themselves are relatively rare [12]. Targeted therapy cannot commence until the causative organisms are identified, and their antibiotic susceptibility patterns are analyzed. This process, traditionally reliant on culture methods, can be time-consuming. However, advances such as the BACTEC 9050 automated blood culture system have improved detection rates, ranging from 15% to 50% [13].

The management of BSIs often requires prolonged antibiotic treatment. For infections related to central vascular catheters, more than 90% of clinicians recommend a treatment duration of at least 10 days [14]. BSIs can be categorized based on their occurrence in specific patient populations: those with compromised physiological defenses (e.g., newborns and the elderly), those with pathological or pharmacological predispositions, and immunocompetent hosts with normal defenses [15].

Early symptoms of sepsis, which can complicate the differential diagnosis between viral and bacterial infections, include slurred speech, confusion, shivering, muscle pain, oliguria, respiratory distress, pallor, and hypotension. BSIs disproportionately affect individuals with underdeveloped or weakened immune systems, with common pathogens including *Escherichia coli, Klebsiella spp., Candida*, group B streptococci, and pneumococci [16].

The present study aims to enhance the understanding of BSIs by exploring the pathogen spectrum, antimicrobial susceptibility patterns, and factors influencing the clinical outcomes of patients with BSIs. This research further seeks to evaluate current treatment protocols, particularly for catheter-associated infections, to optimize management strategies and reduce morbidity and mortality rates. By addressing these objectives, the study aims to provide critical insights that can improve diagnostic precision, treatment efficacy, and overall patient care in the management of BSIs.

II. Materials And Methods

Study Design, Data Collection and Patient Declaration:

The present study was conducted at a tertiary care hospital in Jaipur, Rajasthan. Blood samples were aseptically collected from clinically suspected cases of bloodstream infections and processed department of Microbiology Laboratory of National Institute of Medical Science and Research Rajasthan, Jaipur between July 2024 and October 2024. The A total of 125 blood samples from adults of both genders with clinically diagnosed bloodstream infections were included. Samples other than blood and polymicrobial flora grown in cultures were excluded from the study.

The study was cross-sectional observational study. Sample ware taken using simple randomised sampling techniques. Whereas the inclusion criteria of the patient was wide all the OPD and IPD patients of age group 18 or above of all the genders who were ready to provide written consent with bloodstream infections were considered. In the present study both consent and ascent was taken. Written ascent was taken Clinical IPD patients. In the PIS (Patient Information Sheet) all the required patient were informed about use of the data of research and publication purpose, with non-discloser of patient identify.

Samples other than blood and polymicrobial flora grown in cultures were excluded from the study.

Bacterial Isolation:

All collected blood samples were cultured using the BD BACTEC FX-40 fluorescent series device to detect bacterial pathogens. Positive culture bottles were sub cultured on MacConkey agar and Blood agar, followed by incubation at 37°C for 18–24 hours [17]. After incubation, bacterial colonies were analysed for culture characteristics, gram staining, and biochemical tests.

Blood Culture and Biochemical Testing:

Blood samples were collected from respective hospital wards and transported to the Microbiology Laboratory. Blood culture samples were inoculated into the BD BACTEC FX-40 system and incubated at 37°C for up to 7 days. Positive culture samples were sub cultured onto blood agar and MacConkey agar and incubated at 37°C for 24 hours. Biochemical tests were performed for bacterial identification, including catalase and coagulase tests for Gram-positive bacteria, and indole, methyl red, citrate, urease, TSI, and MMM tests for Gram-negative bacteria

Antimicrobial Susceptibility Testing (AST):

Antimicrobial susceptibility testing was performed using the Kirby-Bauer disc diffusion method on Mueller-Hinton agar following CLSI 2022 guidelines. The tested antibiotics included Amikacin, Ampicillin, Aztreonam, Amoxicillin, Colistin, Cefuroxime, Cefotaxime, Ceftazidime, Ciprofloxacin, Cefepime, Ceftriaxone, Cotrimoxazole, Cefoxitin, Chloramphenicol, Clindamycin, Doxycycline, Erythromycin, Gentamicin, Imipenem, Linezolid, Levofloxacin, Minocycline, Meropenem, Penicillin, Piperacillin, Tigecycline, Teicoplanin, Tetracycline, Tobramycin, Ticarcillin, and Vancomycin (Hi-Media Laboratories, India).

Multidrug resistance (MDR) was defined as resistance to at least three classes of antibiotics. Antibiotic susceptibility results for Gram-positive cocci (GPC) and Gram-negative bacilli (GNB) were analyzed and recorded as per the CLSI recommendations [18].

Antimicrobial Resistance Analysis:

Antimicrobial resistance patterns were assessed based on the CLSI guidelines [2013], and the Kirby-Bauer disc diffusion results were tabulated for analysis. The antibiotic discs used were sourced from Hi-Media, Mumbai, as described in Tables 1–3 [18].

III. Statically Analysis:

We analysed the data with IBM SPSS (version 29; IBM, Armonk, NY, USA). We expressed all categorical variables as number (percentage) and continuous variable as mean and standard deviation. A *p* value of <0.05 was considered statistically significant.

IV. Results And Discussion

On analyzing the data of the studied samples.

Age-Wise Distribution

In this study, a total of 125 clinical isolates were obtained from blood cultures. The highest proportion of isolates was found in patients aged 51–60 years (22.4%), followed by those aged 61–70 years (16%), 41–50 years (14.4%), and 31–40 years (14.4%). Other age groups included patients aged 71–80 years (12.0%), 21–30 years (12.0%), 17–20 years (7.2%), and 81–90 years (1.6%), as shown in Table 1.

Table 1: Age wise distribution of patients attended at NIMS Hospital

Age groups (In years)	17-20 years	21-30 years	31-40 years	41-50 years	51-60 years	61-70 years	71-80 years	81-90 years
Total **(125)**	9	15	18	18	28	20	15	2
Percentage (100)	7.2	12.0	14.4	14.4	22.4	16.0	12.0	1.6

Table 1- 125 clinical isolates were obtained from blood cultures for this investigation. Patients between the ages of 51 and 60 had the highest percentage of isolates (22.4%), followed by those between the ages of 61 and 70 (16%), 41 and 50 (14.4%), and 31 and 40 (14.4%).

Gender-Wise Distribution

The study revealed that 60.7% of Gram-negative isolates and 71.9% of Gram-positive isolates were predominantly isolated from male patients. In contrast, 39.3% of Gram-negative isolates and 28.1% of Gram-positive isolates were recovered from female patients, as shown in Table 2.

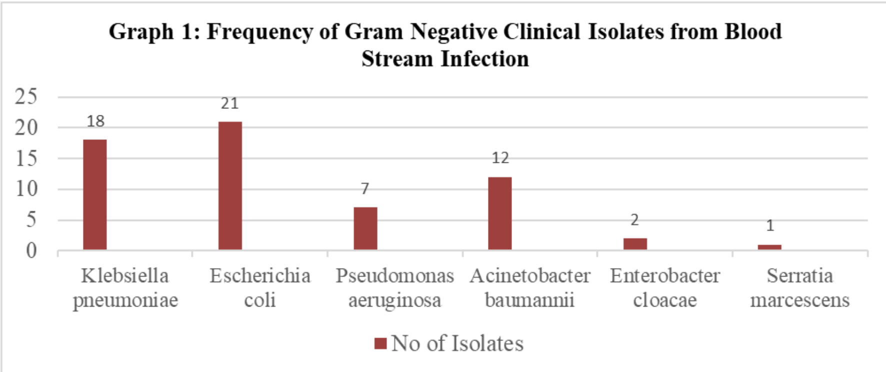
Table 2: Gender wise distribution of Clinical Isolates

Gender	Gram-Negative Isolates	Gram-Positive Isolates	Total
Male	37 (60.7%)	46 (71.90%)	83 (66.4%)
Female	24 (39.3%)	18 (28.10%)	42 (36.6%)
Total	61	64	125

Table 2 - Gram-positive isolates (71.9%) and Gram-negative isolates (60.7%) were recovered from female patients in greater proportion than from male patients (39.3% and 28.1%, respectively), according to the study.

Frequency of Gram-Negative Clinical Isolates in Bloodstream Infections

Among the Gram-negative clinical isolates, *Escherichia coli* (34.4%) was the most frequently isolated pathogen, followed by *Klebsiella pneumoniae* (29.5%), *Acinetobacter baumannii* (19.7%), *Pseudomonas aeruginosa* (11.5%), *Enterobacter cloacae* (3.3%), and *Serratia marcescens* (1.6%). These findings are illustrated in Graph 1.

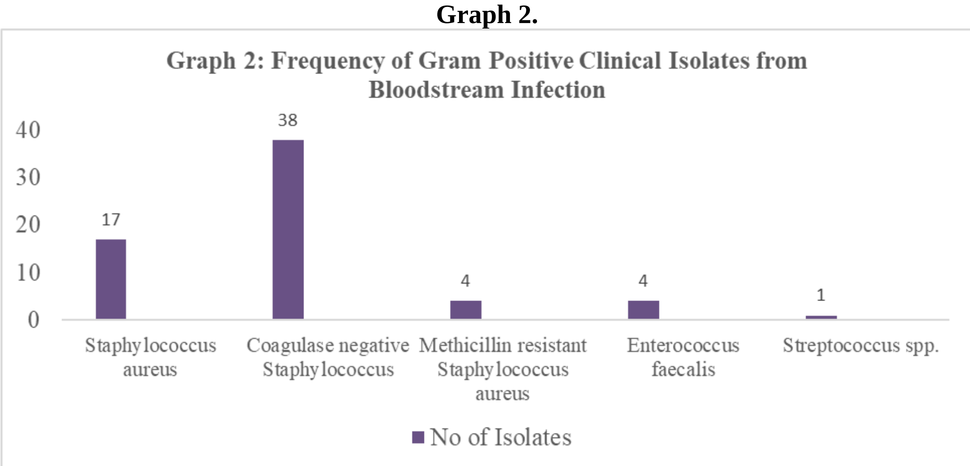


Graph 1- Escherichia coli (34.4%) was the most common Gram-negative isolate, followed by Klebsiella pneumoniae (29.5%), Acinetobacter baumannii (19.7%), Pseudomonas aeruginosa (11.5%), Enterobacter cloacae (3.3%), and Serratia marcescens (1.6%).

Frequency of Gram-Positive Clinical Isolates in Bloodstream Infections

For Gram-positive clinical isolates, coagulase-negative *Staphylococcus aureus* (59.38%) was the most prevalent organism, followed by *Staphylococcus aureus* (32.81%), *Enterococcus faecalis* (6.25%), and *Streptococcus* spp. (1.56%). Among the 32.81% of *Staphylococcus aureus* isolates, 6.25% were identified as methicillin-resistant *Staphylococcus aureus* (MRSA), as shown in

Graph 2.



Graph 2- Coagulase-negative Staphylococcus aureus (59.38%) was the most prevalent Gram-positive isolate, followed by Staphylococcus aureus (32.81%), Enterococcus faecalis (6.25%), and Streptococcus spp. (1.56%). Of the Staphylococcus aureus isolates, 6.25% were identified as MRSA.

Antimicrobial Susceptibility Pattern of Gram-Negative Clinical Isolates

Colistin (100%), Minocycline (90.2%), and Tigecycline (87.70%) were the most sensitive antimicrobials against gram-negative bacterial isolates in this investigation, whereas the largest resistance was to Ampicillin (52%), Cephalosporins (45-51%), and Aminoglycosides (33-36%) as presented in Table 3.

Table 3: Antimicrobial susceptibility pattern of Gram-Negative Clinical Isolates				
Antibiotic	**Sensitive**	**Percentage**	**Resistant**	**Percentage**
AK	25	41.00%	36	59.00%
AMP	9	14.80%	52	85.20%
AT	15	32.80%	36	67.20 %
AMC	18	44.25%	25	55.75%
CL	59	100%	00	-
CXM	12	19.70%	49	80.30%
CTX	9	20.60%	45	79.40%
CAZ	32	52.50%	29	47.50%
CIP	25	41.00%	36	59.00%
CPM	13	21.30%	48	78.70%
CTR	3	10.65%	51	89.35%
COT	19	36.85%	35	63.15%
GEN	28	45.90%	33	54.10%
IPM	37	60.70%	24	39.30%
MI	55	90.20%	6	9.80%
MRP	41	67.20%	20	38.80%
TGC	50	87.70%	4	12.30%
TE	28	45.90%	33	54.10%
TOB	31	50.80%	30	49.20%
TCC	12	42.80%	21	57.20%
PIT	38	62.30%	23	37.70%
DO	4	46.75%	8	53.25%
C	25	56.55%	17	43.45%

****AK***-Amikacin, ***AMP***-Ampicillin, ***AT***-Aztreonam, ***AMC***-Amoxicillin, ***CL***-Colistin, ***CXM***-Cefuroxime, ***CTX***-Cefotaxime, ***CAZ***-Ceftazidime, ***CIP***-Ciprofloxacin, ***CPM***-Cefepime, ***CTR***-Ceftriaxone, ***COT***-Cotrimoxazole, ***GEN***-Gentamicin, ***IPM***-Imipenem, ***MI***-Minocycline, ***MRP***-Meropenem, ***TGC***-Tigecycline, ***TE***-Tetracycline, ***TOB***-Tobramycin, ***TCC***-Ticarcillin, ***PIT***-Piperacillin, ***DO***-Doxycycline, ***C***-Chloramphenicol.*

Antimicrobial Susceptibility Pattern of Gram-Positive Clinical Isolates

The antimicrobial resistance trends for Gram-positive clinical isolates revealed the highest resistance to commonly used antibiotics, including Erythromycin (76.6%), Ampicillin (68.8%), Penicillin (64.1%), Ciprofloxacin (62.5%), Levofloxacin (56.3%), and Clindamycin (53.1%). Methicillin resistance among *Staphylococcus* species was detected using the Cefoxitin disc, with zone diameters measured as per CLSI guidelines. However, Vancomycin, Tigecycline, Teicoplanin, Tetracycline, and Minocycline were the most effective antimicrobial agents, showing 98.4% sensitivity against Gram-positive cocci infections, as presented in Table 4.

Table 4: Antimicrobial susceptibility pattern of Gram-Positive Clinical Isolates				
Antibiotic	**Sensitive**	**Percentage**	**Resistant**	**Percentage**
AMP	20	31.30%	44	68.80%
CIP	24	37.50%	40	62.50%
C	59	92.20%	5	7.80%
COT	39	60.90%	25	39.10%
CD	30	46.90%	34	53.10%
DO	46	71.90%	18	28.10%
E	15	23.40%	49	76.60%
GEN	50	78.10%	14	21.90%
LZ	62	96.90%	2	3.10%
LEVO	28	43.80%	36	56.30%
MI	63	98.40%	1	1.60%
P	23	35.90%	41	64.10%
TOB	48	75.00%	16	25.00%
TEI	63	98.40%	1	1.60%
TGC	63	98.40%	1	1.60%
TE	47	73.40%	17	26.60%
VA	63	98.40%	1	1.60%
AK-Amikacin, AMP-Ampicillin, AT-Aztreonam, AMC-Amoxicillin, CL-Colistin, CXM-Cefuroxime,CTX-Cefotaxime,CAZ-Ceftazidime,CIP-Ciprofloxacin, CPM-Cefepime, CTR-Ceftriaxone, COT-Cotrimoxazole, GEN-Gentamicin, IPM-Imipenem, MI-Minocycline, MRP-Meropenem,TGC-Trigecycline,TE-Tetracycline, TOB-Tobramycin, TCC-Ticarcillin,PIT-Piperacillin, DO-Doxycycline, C-Chloramphenicol.				

V. Discussion

Blood culture remains a cornerstone in diagnosing infectious diseases, yet the widespread misuse of antibiotics contributes significantly to the emergence of resistant bacteria, complicating treatment outcomes. Bloodstream infections (BSIs) are a leading cause of morbidity and mortality globally, necessitating immediate and appropriate therapeutic intervention. In India, the challenge of antimicrobial resistance is amplified due to factors such as antibiotic overuse, random prescribing practices, lack of awareness, prolonged ICU stays, residency in nursing homes, severe illnesses, and frequent use of invasive medical devices.

In the present study, male patients demonstrated a higher culture positivity rate (66.4%) compared to females (36.4%), consistent with previous studies conducted in the country. The findings also revealed that Gram-positive bacteria (51.2%) were slightly more prevalent than Gram-negative bacteria (48.8%), highlighting the dynamic nature of microbial prevalence influenced by regional factors. This predominance of Gram-positive organisms aligns with research by Kaur A. and Singh V., 2014 [19], who reported a similar trend. The observed difference may also be attributed to the practice of administering antibiotics before patients are referred to tertiary care centres, potentially altering the isolation rates.

Among Gram-positive bacteria, *Coagulase-negative Staphylococcus aureus* (59.38%) was the most frequently isolated pathogen, followed by *Staphylococcus aureus* (32.81%), *Enterococcus faecalis* (6.25%), and *Streptococcus* spp. However, it is important to note that *Coagulase-negative Staphylococcus aureus* can often be a skin contaminant, necessitating its repeated isolation from the same patient to establish its clinical significance. These findings corroborate the study by Kaur A. and Singh V., 2014 [19].

Regarding Gram-negative bacteria, *Escherichia coli* (34.4%) emerged as the predominant isolate, followed by *Klebsiella pneumoniae* (29.5%), *Acinetobacter baumannii* (19.7%), and *Pseudomonas aeruginosa* (11.5%). Study by Prashanth K, Badrinath S et al., 2006 reported the prevalence of Gram-negative pathogens may be attributed to nosocomial infections acquired during hospital stays, a pattern frequently observed in healthcare-associated infections by [20]. Study under the Antimicrobial Surveillance Program done by Diekema DJ, et al., 2019 similarly identified *Staphylococcus* and *Escherichia coli* as the most commonly isolated pathogens in BSIs [21]. These findings are further supported by a prospective cohort study by Clark DR, et al., 2021 from the UK, which also identified *Escherichia coli* and *Staphylococcus* species as the predominant pathogens in bloodstream infections [22].

The antimicrobial resistance profile observed in this study indicates the highest resistance rates among Gram-negative isolates to Ampicillin, followed by Cephalosporins and Aminoglycosides. However, the most effective antimicrobials were Colistin, Minocycline, and Tigecycline. This trend aligns with findings by Priyanka Pandit et al., 2021 who reported similar resistance patterns among Gram-negative bacteria [23].

For Gram-positive isolates, Vancomycin, Teicoplanin, and Linezolid demonstrated the highest sensitivity. These results are consistent with a study conducted in North India, which reported 100% efficacy of these antibiotics against Gram-positive pathogens isolated from BSIs [24].

Overall, the findings emphasize the critical need for targeted antimicrobial stewardship to address the challenges of drug resistance and ensure effective treatment of bloodstream infections.

VI. Conclusions

This study underscores the significant burden of bloodstream infections (BSIs), particularly among male patients and older age groups, with *Coagulase-negative Staphylococcus aureus* and *Escherichia coli* being the predominant pathogens. The high resistance to commonly used antibiotics, such as Ampicillin and Cephalosporins, highlights the critical challenge of antimicrobial resistance. However, Vancomycin, Teicoplanin, Linezolid, Colistin, Minocycline, and Tigecycline remain highly effective treatment options. These findings emphasize the need for robust antibiotic stewardship programs, continuous resistance monitoring, and timely, accurate diagnostic approaches to improve patient outcomes and combat antimicrobial resistance effectively.

Acknowledgments

Throughout this investigation, we thank Dr. Shama Tomar and Dr. Anupama Sharma for their helpful advice and assistance. Their knowledge, support, and wise recommendations considerably aided the successful completion of this work.

Ethical Approval

The study protocol was prepared considering the ruled and regulations of medical ethics following the declaration by Helsinki ethical principles. The study was presented in front of IRB and clearance was taken.

Source of Funding

None

Conflicts of Interest
None

Informed consent declaration

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