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Original Research Article

PREVALENCE AND ANTIMICROBIAL SENSITIVITY PATTERN OF PATHOGENS ISOLATED FROM BLOOD IN A TERTIARY CARE INSTITUTE IN NORTH INDIA

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ABSTRACT

Background: Bloodstream infections are a significant cause of morbidity and mortality worldwide. Timely detection of the causative pathogens and their antimicrobial susceptibility patterns is essential for guiding empirical therapy and combating antimicrobial resistance. The aim is to determine the prevalence and antimicrobial sensitivity pattern of pathogens isolated from blood cultures.

Materials and Methods: In this hospital based observational study, 164 blood samples of suspected cases of bloodstream infections were enrolled. Samples were processed using conventional blood culture methods for detection of growth in blood culture for 5 to 7 days, with periodic subculturing if turbidity or signs of growth were observed. Bottles signaling positive were subjected to Gram staining and subculture onto blood agar and MacConkey agar plates, followed by incubation at 35–37 °C for 18–24 hours. Bacterial isolates were identified based on colony morphology, Gram staining, and a battery of standard biochemical tests. AST was performed on all clinically significant isolates by the Kirby–Bauer disk diffusion method on Mueller–Hinton agar, following Clinical and Laboratory Standards Institute (CLSI) guidelines.

Results: Among 164 blood culture samples, 70 (42.7%) samples were positive for growth. Among the positive cultures, Gram-positive bacteria were isolated in 50 cases (71.4%) and Gram-negative bacteria in 20 cases (28.6%). The most frequently isolated organism was MR-CoNS, accounting for 38 isolates (54.3%). Gram-positive organisms retained high susceptibility to Vancomycin, Linezolid, Gentamicin, and Levofloxacin. Gram-negative isolates showed high sensitivity to Amikacin, Gentamicin and Levofloxacin.

Conclusion: The observed susceptibility patterns emphasize the need for rational antibiotic use guided by local antibiograms and the implementation of stringent infection control measures.

Keywords: Bloodstream Infections, Antimicrobial Sensitivity, Gram-negative bacteria, Blood Culture.

INTRODUCTION

Bloodstream infections (BSIs) are among the most serious infectious diseases encountered in clinical practice. They can progress rapidly to sepsis, septic shock and death, and they place a major burden on healthcare systems through prolonged hospital stays, increased resource use, and higher costs.[1,2] Timely detection of the causative pathogen by blood culture and prompt institution of appropriate antimicrobial therapy are central to improving outcomes.[3] However, the emergence of antimicrobial resistance (AMR) has made treatment increasingly challenging. Many bacterial pathogens have developed resistance to multiple classes of antibiotics, creating a serious global health crisis with significant economic and social consequences.[4]

Globally, antimicrobial resistance (AMR) among common BSI pathogens has been rising for decades, reducing the effectiveness of first-line agents and increasing reliance on broader-spectrum or last-resort antibiotics.[5] This problem is particularly acute in low- and middle-income countries where antibiotic use is high, infection-control measures may be

variable, and laboratory surveillance networks are still expanding.[6,7] The spectrum of organisms causing BSIs and their antimicrobial susceptibility patterns vary by geography, patient population and healthcare setting, so local surveillance data are essential to guide empiric therapy and infection-control strategies.[8]

India is one of the Countries most severely affected by AMR, with increasing resistance to both essential and last-resort antibiotics.[9] Although some national surveillance data exist, there remains a paucity of detailed studies on the prevalence, etiology, and resistance profiles of bacterial BSIs in different regions of the country. Hospital-based studies across India have consistently shown a predominance of Gram-negative bacilli, particularly Escherichia coli, Klebsiella pneumoniae and Pseudomonas spp. among blood culture isolates, with coagulase-negative staphylococci and Staphylococcus aureus the common Gram-positive pathogens. Importantly, these studies consistently document high rates of resistance to third-generation cephalosporins, fluoroquinolones and, increasingly, carbapenems among Enterobacterales and non-fermenters, frequently driven by ESBLs and carbapenemases while methicillin resistance among staphylococci and reduced susceptibility to first-line agents among enterococci are also reported. These evolving patterns undermine standard empiric regimens and make up-to-date local antibiograms indispensable for clinicians.[10] The shifting epidemiology and rising resistance threaten the effectiveness of antibiotics frequently used to prevent and treat BSIs.[11]

India, being a developing economy and a recognized hotspot for emerging infectious diseases, faces alarming rates of AMR, an important cause of treatment failure and mortality. While national surveillance efforts have been valuable, substantial heterogeneity exists between hospitals due to variations in case mix (medical, surgical, neonatal/paediatric, ICU), laboratory methodologies, and patient referral patterns. This makes single-centre, institution-specific studies highly valuable. Tertiary care institutes in North India cater to a diverse and often critically ill population, making local data on pathogen prevalence and antimicrobial susceptibility crucial for effective empiric therapy, antimicrobial stewardship, and infection prevention programs. Given the clinical urgency in selecting appropriate empiric therapy for suspected BSIs and the dynamic nature of AMR, the present study aims to determine the prevalence and species distribution of pathogens isolated from blood cultures in a tertiary care institute in North India, and to describe their antimicrobial susceptibility patterns.

MATERIALS AND METHODS

This hospital based observational study was conducted in the Department of Microbiology, Government Medical College, Baramulla, over a period of one year from January 2023 to December 2024. The study was approved by the Ethics Committee of Government Medical College, Baramulla. The department receives blood culture samples from various inpatient units, including medical, surgical, paediatric, and intensive care units (ICUs), as well as from outpatient departments. In this study 164 blood samples of suspected cases of bloodstream infection, for whom blood culture was requested by the treating physician during the study period, were included. Patients of all ages and both sexes were considered.

Inclusion criteria

- Patients with clinical suspicion of BSI
- Blood samples processed in the microbiology laboratory during the study period

Exclusion criteria

- Repeat cultures from the same patient yielding the same organism within the same episode of illness
- Blood cultures yielding contaminants (e.g., skin commensals) without supportive clinical evidence

Blood samples were collected before the administration of any antibiotic and under strict aseptic precautions by trained healthcare personnel. For adults, 5-10mL of blood while 1-5mL blood was collected from pediatric patients, and 1-2mL from neonates for blood culture was collected. Samples were processed using conventional blood culture methods for detection of growth in blood culture for 5 to 7 days, with periodic subculturing if turbidity or signs of growth were observed. Bottles signaling positive were subjected to Gram staining and subculture onto blood agar and MacConkey agar plates, followed by incubation at 35–37 °C for 18–24 hours. Bacterial isolates were identified based on colony morphology, Gram staining, and a battery of standard biochemical tests (including catalase, coagulase, oxidase, indole, citrate utilization, urease, triple sugar iron reaction, motility, and others as applicable). In certain cases, identification was confirmed using automated identification systems such as VITEK-2 (bioMérieux). AST was performed on all clinically significant isolates by the Kirby–Bauer disk diffusion method on Mueller–Hinton agar, following Clinical and Laboratory Standards Institute (CLSI) guidelines.12 The antibiotic panels tested were selected according to the organism group and included:

- Gram-negative isolates: ampicillin, amoxicillin–clavulanic acid, piperacillin–tazobactam, ceftriaxone, ceftazidime, cefepime, gentamicin, amikacin, ciprofloxacin, meropenem, imipenem, colistin, tigecycline.
- Gram-positive isolates: penicillin, oxacillin/cefoxitin (for MRSA detection), erythromycin, clindamycin, gentamicin (high-level), ciprofloxacin, linezolid, vancomycin, teicoplanin.

Minimum inhibitory concentrations (MICs) for certain drugs (e.g., vancomycin, colistin) were

determined by broth microdilution or E-test strips where indicated.

Detection of resistance mechanisms

- Extended-spectrum β-lactamase (ESBL) production: confirmed by combination disk method using ceftazidime and cefotaxime with and without clavulanic acid.
- Carbapenemase production: screened using meropenem disk; confirmed by modified carbapenem inactivation method (mCIM).
- Methicillin resistance in S. aureus: detected using cefoxitin disk diffusion method.

Statistical Analysis: The recorded data was compiled and entered in a spreadsheet (Microsoft Excel) and then exported to data editor of SPSS Version 20.0 (SPSS Inc., Chicago, Illinois, USA). Descriptive statistics (frequencies, percentages) were used to summarize categorical variables. Resistance percentages were calculated for each organism–antibiotic combination to prepare the institutional antibiogram.

RESULTS

Out of 164 blood culture samples, 94 samples (57.3%) were sterile (no growth), while 70 samples (42.7%) yielded significant bacterial growth. Among the positive cultures, Gram-positive bacteria were isolated in 50 cases (71.4%) and Gram-negative bacteria in 20 cases (28.6%). The most frequently isolated organism was methicillin-resistant coagulase-negative staphylococci (MR-CoNS), accounting for 38 isolates (54.3% of positive cultures), followed by methicillin-sensitive Staphylococcus aureus (MSSA) in 4 cases (5.7%) and methicillin-resistant S. aureus (MRSA) in 8 cases (11.4%).

Among Gram-negative isolates, Klebsiella pneumoniae was the most common (10 isolates; 14.3%), followed by Acinetobacter spp. (6 isolates; 8.6%), Pseudomonas spp. (2 isolates; 2.9%), and S. berea (2 isolates; 2.9%) [Table 1].

Table 1: Distribution of bacterial isolates from blood cultures

Organism isolated	Number of isolates (n)	Percentage of positive cultures (%)	Percentage of total samples (%)
Gram-positive isolates			
MR-CoNS	38	54.3	23.2
MSSA	4	5.7	2.4
MRSA	8	11.4	4.9
Gram-negative isolates			
Klebsiella pneumoniae	10	14.3	6.1
Acinetobacter spp.	6	8.6	3.7
Pseudomonas spp.	2	2.9	1.2
S. Berea	2	2.9	1.2
Sterile cultures	94	–	57.3
Total samples processed	164	–	100

In the present study, methicillin-resistant coagulase-negative staphylococci (MRCONS) showed very low susceptibility to penicillin (5%) and erythromycin (5%), while moderate susceptibility was noted for clindamycin (25%) and levofloxacin (37%). High susceptibility was observed for vancomycin (100%), linezolid (80%), gentamicin (80%), and tetracycline (95%), whereas resistance to ceftazidime (20%) and piperacillin–tazobactam (5%) was marked. Methicillin-sensitive Staphylococcus aureus (MSSA) isolates demonstrated complete susceptibility to penicillin, piperacillin–tazobactam, linezolid, and levofloxacin (100%), with moderate susceptibility to clindamycin (50%) and gentamicin (50%), but lower susceptibility to vancomycin (50%). Methicillin-resistant Staphylococcus aureus (MRSA) displayed high susceptibility to vancomycin (100%) and linezolid (100%), moderate to levofloxacin (50%), clindamycin (25%), and gentamicin (50%), with poor response to other agents tested. Among Gram-negative bacilli, Klebsiella pneumoniae exhibited high susceptibility to amikacin, piperacillin–tazobactam, and polymyxin B (100%), followed by meropenem (80%) and levofloxacin (60%), but lower rates for ceftriaxone (40%) and cotrimoxazole (60%). Acinetobacter spp. showed complete susceptibility to polymyxin B (100%) and relatively better susceptibility to levofloxacin (67%), but reduced susceptibility to ciprofloxacin (33%) and cotrimoxazole (33%). S. berea isolates were uniformly susceptible to meropenem, colistin, and gentamicin (100%), while Pseudomonas aeruginosa demonstrated full susceptibility to ciprofloxacin, gentamicin, amikacin, piperacillin–tazobactam, and levofloxacin (100%). Overall, vancomycin, linezolid, amikacin, polymyxin B, and levofloxacin showed the highest activity against the major blood culture isolates, whereas penicillin, erythromycin, and ceftazidime exhibited the lowest activity [Table 2].

Table 2: Antimicrobial susceptibility profile of major isolates (% susceptible)

Antibiotic	MRCONS (n=38)	MSSA (n=4)	MRSA (n=8)	Klebsiella pneumoniae (n=10)	Acinetobacter spp. (n=6)	S. berea (n=2)	Pseudomonas aeruginosa (n=2)
Penicillin	5	100	0	0	0	0	0
Erythromycin	5	0	0	0	0	0	0

Clindamycin	25	50	25	0	0	0	0
Vancomycin	100	50	100	0	0	0	0
Linezolid	80	100	100	0	0	0	0
Ciprofloxacin	0	0	0	25	33	50	100
Gentamicin	80	50	50	80	0	50	100
Amikacin	0	0	0	100	0	50	100
Ceftriaxone	0	0	0	40	0	50	0
Ceftazidime	20	0	0	0	0	0	0
Piperacillin–Tazobactam	5	100	0	100	0	0	100
Meropenem	0	0	0	80	0	100	0
Colistin	0	0	0	0	0	100	0
Imipenem	0	0	0	40	0	0	0
Morepenim	0	0	0	40	0	0	0
Levofloxacin	37	100	50	60	67	0	100
Polymyxin B	0	0	0	100	100	0	0
Tigecycline	0	0	0	25	0	0	0
Cotrimoxazole	0	0	0	60	33	0	0
Ertapenum	0	0	0	40	0	0	0
Tetracycline	95	50	25	0	0	0	0

DISCUSSION

Bloodstream infection (BSI) remains a major clinical challenge due to its potential to rapidly progress to severe sepsis, septic shock, and death if not promptly diagnosed and managed. Timely detection, accurate identification, and antimicrobial susceptibility testing of blood-borne pathogens are therefore among the most critical responsibilities of the diagnostic microbiology laboratory. In the present study, a total of 164 blood culture samples were processed, of which 42.7% yielded significant bacterial growth. Gram-positive cocci were the predominant isolates, with methicillin-resistant coagulase-negative staphylococci (MR-CoNS) accounting for the majority (54.3% of positive cultures), followed by methicillin-resistant Staphylococcus aureus (MRSA) and methicillin-sensitive S. aureus (MSSA). Gram-negative bacilli were less frequently encountered and included Klebsiella pneumoniae, Acinetobacter spp., Pseudomonas aeruginosa, and S. berea.

Our findings are consistent with several previous studies, highlighting the continuing predominance of Gram-positive organisms in BSIs. Khanal et al. reported a comparable blood culture positivity rate of 44%,[13] while Sharma et al. documented a positivity rate of 33.9%.[14] Similarly, a study by Ali et al. in Gonder, Ethiopia (2008) observed a prevalence of 24.2%.[15] Such variations in positivity rates across studies may be attributable to differences in patient populations, sampling techniques, prior antibiotic exposure, and laboratory protocols. The predominance of MR-CoNS in our study aligns with the increasing recognition of its role as a significant pathogen in healthcare-associated BSIs, particularly in patients with indwelling medical devices or compromised immunity.

MR-CoNS exhibited high resistance to penicillin and erythromycin but remained highly susceptible to vancomycin (100%), linezolid (80%), gentamicin (80%), and tetracycline (95%). This pattern resonates with findings from Mumbai tertiary hospitals where CoNS demonstrated reduced susceptibility to multiple antibiotics, with therapeutic agents like vancomycin and linezolid maintaining effectiveness.[16] MSSA isolates were fully sensitive to linezolid and levofloxacin, with moderate sensitivity to clindamycin and gentamicin, although vancomycin sensitivity was surprisingly only 50%, which could reflect local prescribing pressures or methodological factors. MRSA isolates showed universal susceptibility to vancomycin and linezolid, aligning with broader Indian data where these remain reliable anti-MRSA agents.[16] However, reduced susceptibility to ciprofloxacin, clindamycin, and gentamicin underscores limited alternative options.

Klebsiella pneumoniae demonstrated complete susceptibility to amikacin, piperacillin–tazobactam, and polymyxin B; meropenem susceptibility was high (80%), with moderate sensitivity to levofloxacin and cotrimoxazole, but only 40% to ceftriaxone. This contrasts with a multicentric Indian study showing lower susceptibilities: β-lactam and cephalosporin susceptibility ranged from ~45–52% (49% average); carbapenem susceptibility ranged from 55–72% and aminoglycoside (amikacin/gentamicin) susceptibility around 55–65%.[17,18] The higher susceptibilities observed in our study may reflect local antimicrobial usage patterns, lower prevalence of carbapenemase-producing strains, or differences in infection control practices; however, the reduced activity of ceftriaxone underscores the growing challenge of extended-spectrum β-lactamase (ESBL) production among Klebsiella pneumoniae.

In our cohort, Acinetobacter spp. isolates exhibited complete susceptibility to polymyxin B and a comparatively higher susceptibility to levofloxacin (67%), while demonstrating markedly lower sensitivity to ciprofloxacin (33%) and cotrimoxazole (33%). This resistance profile aligns with global surveillance reports that consistently identify Acinetobacter as a multidrug-resistant opportunistic pathogen and a major cause of difficult-to-treat nosocomial infections, particularly in critically ill patients.[18] The high susceptibility to polymyxin B underscores its continued role as a last-resort therapeutic option, although its toxicity profile and the risk of emerging resistance remain concerns.

Although Pseudomonas aeruginosa were isolated infrequently in our study, demonstrated complete susceptibility to multiple antimicrobial classes, including ciprofloxacin, gentamicin, amikacin, Piperacillin + Tazobactam and levofloxacin. While this is encouraging from a therapeutic standpoint, such findings must be interpreted cautiously, as low isolate numbers and potential patient selection bias could overestimate true susceptibility rates. Sustained vigilance through ongoing local antimicrobial resistance surveillance remains essential to detect early shifts in these patterns and guide empirical therapy effectively.

The high susceptibility of many Gram-negative isolates to last-resort agents like polymyxins B and Colistin must be seen in the context of rising resistance in India. Alarmingly, some outbreaks of colistin-resistant Klebsiella pneumoniae have been documented.[19] National surveillance by GLASS shows ESBL prevalence of 50–90%, colistin resistance in CRE rising to 37%.[18]

Implications and recommendations

The antimicrobial susceptibility patterns observed in our study highlight a mixed picture: Gram-positive pathogens remain largely treatable with vancomycin and linezolid, while multidrug-resistant Gram-negatives pose persistent threats. The high susceptibility to aminoglycosides and polymyxins is encouraging but calls for advocacy to prevent overuse and preserve their utility. Institutional antibiograms informed by ongoing surveillance are critical for guiding empiric therapy decisions and updating stewardship protocols. In addition, enhanced infection control and antimicrobial stewardship are urgently needed to curb the emergence and spread of resistance, especially carbapenemase-producing and colistin-resistant strains.

Limitations: This study had certain limitations. First, it was conducted at a single tertiary care hospital, which may limit the generalizability of the findings to other healthcare settings with different patient populations and infection control practices. Second, the sample size, though adequate for preliminary observations, was relatively small for certain bacterial species, restricting the statistical power to draw robust conclusions for those organisms.

CONCLUSION

The observed susceptibility patterns emphasize the need for rational antibiotic use guided by local antibiograms and the implementation of stringent infection control measures. Strengthening antimicrobial stewardship programs, coupled with regular updates of empirical therapy guidelines based on local data, remains essential to mitigate the growing threat of antimicrobial resistance.

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