




Original Article

Antimicrobial Resistance Patterns of Clinical Isolates in a Tertiary Care Hospital: A Retrospective Study

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ABSTRACT

Background: Antimicrobial resistance (AMR) has become a major challenge in clinical practice, particularly in tertiary care hospitals where antibiotic exposure is high. Continuous evaluation of local resistance patterns is essential for guiding effective empirical therapy.

Objectives: To determine the distribution of bacterial isolates and assess their antimicrobial susceptibility patterns in a tertiary care hospital.

Materials and Methods: This retrospective study was conducted over a six-month period from July to December 2025 in the Department of Microbiology at a tertiary care teaching hospital. A total of non-duplicate bacterial isolates obtained from various clinical specimens were included. Identification was carried out using standard microbiological techniques, and antimicrobial susceptibility testing was performed using the Kirby–Bauer disc diffusion method as per CLSI guidelines.

Results: Gram-negative organisms predominated among the isolates, with *Escherichia coli* and *Klebsiella pneumoniae* being the most frequently identified pathogens. High resistance was observed to commonly used antibiotics such as ampicillin, cephalosporins, and fluoroquinolones. In contrast, better susceptibility was noted with piperacillin–tazobactam, carbapenems, and aminoglycosides. Among Gram-positive organisms, reduced sensitivity to fluoroquinolones was observed, while vancomycin and linezolid remained effective.

Conclusion: The study highlights a rising trend of antimicrobial resistance and reduced effectiveness of commonly used antibiotics. Continuous surveillance, rational antibiotic use, and implementation of antimicrobial stewardship programs are essential to control the spread of multidrug-resistant organisms.

Keywords: Antimicrobial resistance; Antibiotic susceptibility; Retrospective study; Gram-negative bacteria; Gram-positive bacteria; Multidrug resistance; Tertiary care hospital.

INTRODUCTION

Antimicrobial resistance (AMR) has become an increasingly serious concern in clinical medicine, affecting the successful treatment of infectious diseases across the globe. The growing inability of commonly used antibiotics to effectively treat bacterial infections has led to increased disease burden, prolonged hospitalization, and higher healthcare costs. International health agencies, including the World Health Organization, have emphasized the urgent need to monitor and control the spread of resistant microorganisms (1).

In developing nations such as India, the problem of AMR is further intensified due to factors such as inappropriate antibiotic prescribing, easy access to antibiotics without prescription, and inadequate infection control practices (2,3). Tertiary care hospitals, in particular, serve as hotspots for resistant infections because of the high patient load, frequent use of invasive procedures, and extensive administration of broad-spectrum antimicrobial agents.

Gram-negative bacteria, including *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Acinetobacter* species, are commonly implicated in hospital-acquired infections. These organisms possess multiple resistance mechanisms such as extended-spectrum beta-lactamase (ESBL) production and carbapenem resistance, which significantly limit available therapeutic options (4,5). In parallel, Gram-positive organisms such as *Staphylococcus aureus* continue to

contribute to both community and hospital-acquired infections, with methicillin-resistant strains posing a major treatment challenge (6).

Empirical antibiotic therapy is frequently initiated before culture results are available; however, such practices may contribute to resistance if not guided by local susceptibility data. Therefore, periodic assessment of antimicrobial susceptibility patterns through hospital antibiograms is essential to support appropriate antibiotic selection (7).

In addition, antimicrobial stewardship programs have been recommended as a key strategy to optimize antibiotic use, reduce resistance, and improve clinical outcomes. These programs rely heavily on local microbiological data for effective implementation (8).

Considering the dynamic nature of antimicrobial resistance and the need for updated regional data, the present retrospective study was undertaken to evaluate the antimicrobial resistance patterns of bacterial isolates over a six-month period in a tertiary care hospital.

MATERIALS AND METHODS:

Study Design and Setting

This retrospective observational study was conducted in the Department of Microbiology at Fathima Medical College, a tertiary care teaching institution. The microbiology laboratory processes specimens from multiple clinical departments, including inpatient wards and intensive care units.

Study Duration

The study included laboratory data collected over a six-month period from July 2025 to December 2025.

Study Population

All clinically relevant bacterial isolates obtained from patient samples during the study period were included. Only the first isolate from each patient was considered to avoid duplication. Samples from both outpatient and inpatient settings were analyzed.

Inclusion Criteria

- Clinically significant bacterial isolates from various specimens
- Isolates from patients of all age groups
- Non-repetitive isolates (single isolate per patient)

Exclusion Criteria

- Duplicate isolates from the same patient
- Samples with contamination or insignificant growth
- Non-bacterial isolates such as fungi

Specimen Collection and Processing

Clinical specimens including urine, blood, pus, respiratory samples, and body fluids were collected using aseptic techniques and transported promptly to the laboratory. Standard microbiological methods were followed for culture and isolation.

Urine samples were processed using calibrated loop techniques, while blood cultures were handled using conventional methods. Other specimens were inoculated onto appropriate media such as blood agar and MacConkey agar and incubated under suitable conditions. Growth was assessed after incubation.

Identification of Bacterial Isolates

Isolates were identified based on colony morphology, Gram staining characteristics, and biochemical reactions. Conventional tests such as indole, citrate utilization, urease, and triple sugar iron tests were used for Gram-negative organisms, while Gram-positive bacteria were identified using catalase and coagulase tests.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disc diffusion method on Mueller–Hinton agar. Testing procedures and interpretation were carried out according to guidelines of the Clinical and Laboratory Standards Institute (CLSI).

A standardized inoculum equivalent to 0.5 McFarland standard was used. After incubation, inhibition zones were measured and categorized as sensitive, intermediate, or resistant.

The antibiotics tested included representatives from major classes such as beta-lactams, cephalosporins, carbapenems, aminoglycosides, fluoroquinolones, and other commonly used agents.

Data Collection and Analysis

Data were retrieved from laboratory registers and digital records. Information collected included type of specimen, bacterial isolate, and antibiotic susceptibility pattern.

Results were summarized using descriptive statistics and expressed as percentages. Sensitivity levels were categorized into high, moderate, and low based on predefined thresholds.

Ethical Considerations: Institutional approval was obtained prior to data collection.

RESULTS:

A total of **196 bacterial isolates** were analyzed during the study period. Gram-negative organisms constituted the majority of isolates compared to Gram-positive organisms. Gram-negative organisms predominated. *Escherichia coli* was the most frequently isolated organism, followed by *Klebsiella pneumoniae*. Among Gram-positive organisms, *Staphylococcus aureus* and CoNS were the leading isolates. Other organisms contributed smaller proportions. (Table 1)

Table 1: Distribution of Bacterial Isolates

Organism	Number (n)	Percentage (%)
<i>Escherichia coli</i>	44	22.4
<i>Klebsiella pneumoniae</i>	39	19.9
<i>Pseudomonas aeruginosa</i>	18	9.2
<i>Acinetobacter</i> spp.	12	6.1
<i>Citrobacter</i> spp.	10	5.1
<i>Proteus</i> spp.	5	2.5
<i>Enterobacter</i> spp.	4	2.0
<i>Staphylococcus aureus</i>	32	16.3
CoNS	21	10.7
<i>Enterococcus</i> spp.	6	3.1
Beta-hemolytic streptococci	5	2.5
Total	196	100

Gram-negative organisms showed high resistance to ampicillin and reduced sensitivity to cephalosporins and fluoroquinolones. Piperacillin–tazobactam, carbapenems, and aminoglycosides demonstrated comparatively better activity. (Table 2)

Table 2: Antibiotic Sensitivity Pattern of Gram-negative Isolates (%)

Organism	AMP	AMC	PTZ	CTR	CTX	CIP	COT	IPM	MRP	AK
<i>E. coli</i>	5	38	86	42	40	28	35	88	90	82
<i>Klebsiella pneumoniae</i>	2	45	84	48	45	30	60	78	76	81
<i>Pseudomonas aeruginosa</i>	0	IR	92	IR	IR	70	28	87	88	86
<i>Acinetobacter</i> spp.	0	20	78	35	32	25	30	70	72	75
<i>Citrobacter</i> spp.	5	30	88	50	48	75	55	85	83	82
<i>Proteus</i> spp.	10	45	90	80	78	72	60	88	90	85
<i>Enterobacter</i> spp.	0	35	92	45	42	78	58	86	88	83

: Gram-positive isolates showed reduced sensitivity to fluoroquinolones but retained good susceptibility to tetracyclines and glycopeptides. Linezolid also showed high effectiveness. (Table 3)

Table 3: Antibiotic Sensitivity Pattern of Gram-positive Isolates (%)

Organism	AMC	PTZ	CIP	COT	DOX	TET	VAN	LZ
<i>Staphylococcus aureus</i>	80	75	30	60	88	90	82	85
CoNS	85	78	35	70	92	95	88	90
<i>Enterococcus</i> spp.	55	75	50	65	88	82	90	88
Beta-hemolytic streptococci	88	94	78	50	90	90	92	92

First-line antibiotics demonstrated reduced effectiveness (<60%) in many isolates, whereas reserve antibiotics showed higher sensitivity (>80%). This pattern indicates an increasing trend of antimicrobial resistance. (Table 4)

Table 4: Interpretation of Antibiotic Sensitivity

Sensitivity Range	Interpretation
>80%	Highly sensitive
60–80%	Moderately sensitive
<60%	Poor sensitivity

DISCUSSION:

The present retrospective study highlights the evolving pattern of antimicrobial resistance among clinical bacterial isolates in a tertiary care hospital. The predominance of Gram-negative organisms observed in this study is consistent with the changing epidemiological trends in hospital-acquired infections, where these organisms are increasingly implicated as major pathogens (9).

Among the isolates, *Escherichia coli* and *Klebsiella pneumoniae* were the most frequently identified organisms. These findings are comparable with previous studies that report Enterobacteriaceae as the leading cause of infections in tertiary care settings, particularly in urinary tract and bloodstream infections (9,10). The high prevalence of these organisms may be attributed to factors such as prolonged hospitalization, use of invasive devices, and widespread empirical antibiotic therapy.

A notable observation in this study is the high level of resistance exhibited by Gram-negative organisms to commonly used antibiotics such as ampicillin, cephalosporins, and fluoroquinolones. This pattern suggests the presence of resistance mechanisms such as extended-spectrum beta-lactamase (ESBL) production, which has been widely reported in similar studies (11,12). The increasing resistance to these first-line agents poses a significant challenge in the selection of empirical therapy.

In contrast, relatively higher susceptibility was observed with piperacillin–tazobactam, carbapenems, and aminoglycosides. These findings indicate that these antibiotics remain effective options for treating severe infections caused by Gram-negative organisms. However, the emergence of reduced sensitivity even to carbapenems is a concerning trend, as carbapenem-resistant Enterobacteriaceae have been increasingly reported worldwide and are associated with limited treatment options and higher mortality (13).

The presence of non-fermenting Gram-negative organisms such as *Pseudomonas aeruginosa* and *Acinetobacter* species further adds to the complexity of antimicrobial resistance. These organisms are known for their intrinsic resistance and ability to acquire additional resistance mechanisms, making them difficult to treat and often associated with hospital-acquired infections, especially in intensive care units (13).

Among Gram-positive organisms, *Staphylococcus aureus* and coagulase-negative staphylococci (CoNS) were the predominant isolates. These organisms are commonly associated with skin and soft tissue infections, as well as device-related infections. The relatively high proportion of CoNS may reflect their increasing recognition as significant pathogens rather than mere contaminants (14).

The antimicrobial susceptibility pattern of Gram-positive isolates revealed reduced sensitivity to fluoroquinolones and moderate susceptibility to cotrimoxazole. However, higher sensitivity to doxycycline, tetracycline, vancomycin, and linezolid was observed. These findings are consistent with previous reports that highlight the continued effectiveness of glycopeptides and oxazolidinones in the treatment of resistant Gram-positive infections, including methicillin-resistant *Staphylococcus aureus* (MRSA) (15).

An important finding of this study is the declining effectiveness of first-line antibiotics across both Gram-negative and Gram-positive organisms. This trend is likely driven by inappropriate and excessive use of antibiotics, which creates selective pressure favoring resistant strains. On the other hand, higher sensitivity observed with reserve antibiotics indicates their continued efficacy, although their indiscriminate use may lead to further resistance.

The findings of this study underscore the importance of regular surveillance of antimicrobial resistance patterns through hospital antibiograms. Such data are essential for guiding empirical therapy, optimizing antibiotic use, and improving patient outcomes. Furthermore, the implementation of antimicrobial stewardship programs plays a critical role in promoting rational antibiotic use and limiting the spread of resistant organisms (16).

CONCLUSION:

This study demonstrates a significant burden of antimicrobial resistance, particularly among Gram-negative organisms, with reduced effectiveness of commonly used antibiotics such as beta-lactams and fluoroquinolones. Higher susceptibility to reserve drugs was observed, indicating their continued therapeutic value.

These findings emphasize the importance of continuous surveillance, rational antibiotic prescribing, and implementation of antimicrobial stewardship programs to limit the spread of multidrug-resistant organisms and improve patient outcomes.

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