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Original Research Article

Trends of carbapenem resistance in *Acinetobacter baumannii, Pseudomonas aeruginosa* and Enterobacterales in a tertiary care hospital

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Abstract

Introduction: Gram-negative pathogens resistant to carbapenems, including *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and Carbapenem-resistant Enterobacterales (CRE) represent a significant public health threat. Enterobacterales are implicated in a wide range of medical infections, which pose significant concerns due to limited treatment options. These bacteria are frequently responsible for infections in healthcare environments.

Aim: The objective of this study is to evaluate the epidemiological burden of carbapenem-resistant Gram-negative bacteria obtained from clinical specimens over a two-year period within a tertiary healthcare facility.

Materials and Methods: This research was performed across a two-year interval, from June 2022 to May 2024, during which several clinical samples were collected and evaluated in the Microbiology Laboratory. Gram-negative bacteria from various clinical samples were isolated and subjected to identification and antimicrobial susceptibility testing via the VITEK 2 automated system (BioMérieux, Durham, North Carolina) with results interpreted conferring to CLSI 2022 rules**.**

Result: The analysis of 1,750 *Pseudomonas aeruginosa, Acinetobacter baumannii* and Enterobacterales isolates was performed. Among these, Carbapenem resistance was detected in 639 isolates, representing 36.5% of the total. The distribution of resistant isolates was as follows: 398 (62.2%) were Enterobacterales, 132(20.6%) were *Acinetobacter baumannii* and 109 (17%) were *Pseudomonas aeruginosa*.

Conclusion: Current research recognized a total prevalence of 36.5% for Carbapenem-resistant A.baumannii, *P.aeruginosa* and Enterobacterales constant with earlier records from India. Isolates resistant to carbapenem are at the maximum frequency in these locations underscoring the critical need for stringent infection prevention measures in addition to well-employed antimicrobial stewardship programs to reduce infection rates.

Keywords: Carbapenem resistance, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, Gram-negative, Enterobacterales, *Escherichia coli* and *Klebsiella pneumonia.*

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1. Introduction

Gram-negative bacilli (GNB) are associated with diverse infections.[1] Carbapenems, a type of β-lactam antibiotic with broad-spectrum effectiveness, have been vital in the therapeutic modalities for severe infections resulting in Gram-negative bacterial pathogens with drug-resistant.[2]

Carbapenems are observed as the preceding line of resistance for medicating multidrug-resistant infections however, their effectiveness has been compromised by the emergence of various resistance mechanisms due to their overuse and misuse.[3.] Infections caused by carbapenem-resistant organisms (CRO) are linked to extended lengths of stay, elevated medical expenses, and a significant rise in death rates.[4] Due to plasmid-mediated nature of carbapenem resistance allows for the efficient horizontal transfer of carbapenemase genes, leading to the rapid dissemination of resistance amongst susceptible Enterobacteriaceae, *Pseudomonas species*., and *Acinetobacter species* isolates.[5]

The rising incidence of CRE has emerged as a major concern due to its association with heightened fatality rates with widespread dissemination.[6] CRO are those that demonstrate resistance to one or more carbapenem antibiotics—such as ertapenem, imipenem, meropenem or doripenem.[7] Maximum frequently encountered CROs include carbapenem-resistant Acinetobacter baumannii

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(CRAB), carbapenem-resistant Pseudomonas aeruginosa (CRPA) and carbapenem-resistant Enterobacterales (CRE).[8]

Clinically significant carbapenem resistance is primarily driven by carbapenemase invention, the enzymatic breakdown of carbapenems and β-lactam drugs.[9] In CRE, carbapenemase-coding genes remain frequently transferred within and between species through horizontal plasmid-mediated transfer.[8] Additional carbapenem resistance mechanisms comprise the excessive expression of microbial a decreased binding affinity of PBPs, carbapenems to penicillin-binding proteins (PBPs), loss of porins in the cell membrane of bacteria and efflux pumps and efflux pumps.[10] Carbapenems exhibit broad-spectrum activity against a diverse range of bacteria, owing to their unique structure that includes a β-lactam ring, defends most β-lactamases, extended-spectrum β-lactamases (ESBLs) and together with Metallo-β-lactamases.[11]

Carbapenemases are a diverse class of enzymes categorized into β-lactamase molecular classes A, B, and D.[12] In *Acinetobacter baumannii*, the most common mechanism of carbapenem resistance involves class D oxacillinases, whereas in *Pseudomonas aeruginosa*, resistance is mainly driven by class B metallo-β-lactamases.[13,14]

The frequent occurrence of cross-resistance towards aminoglycosides and fluoroquinolones in carbapenem-resistant organisms (CRO) presents a significant clinical challenge by limiting therapeutic alternatives.[15] Pharmacokinetic limitations, toxicity issues, and restricted availability limit the few treatment options currently available.[16] The prevalence of carbapenem-resistant organisms (CRO) differs among countries and healthcare settings, with an estimated rate of around 13% in India.[17]

Early identification of carbapenemase production and other resistance mechanisms is essential for guiding appropriate antibiotic therapy and controlling the spread of CRO infections.

This study assesses the clinical impact of carbapenem resistance, and analyzed an incidence of carbapenem-resistant Gram-negative bacteria (CRGNB) from over two years within a tertiary healthcare facility.

2. Materials and Methods

2.1. Study setting

Samples were processed and analyzed in the Microbiology Department, Government Institute of Medical Sciences (GIMS), Greater Noida, India.

2.2. Ethical committee approval

Under reference number GIMS/IEC/HR/2022/03, the Institutional Ethics Committee (IEC) approved this study.

2.3. Inclusion criteria

The study included Gram-negative bacilli isolated over a two-years duration from June 2022 to May 2024 from both inpatient departments (wards and ICU) and outpatient departments (OPD). Healthcare-associated infections

2.4. Exclusion criteria

Incomplete data were excluded during collection: Isolates as of the similar case and bugs not belonging to *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and Enterobacterales.

2.5. Sample processing

Clinical specimens, including urine, blood, respiratory samples, sterile body fluids and pus were cultivated on MacConkey and Blood agar.

Using the VITEK 2 automated system (BioMérieux, Durham, NC, USA), bacterial isolates were identified and subjected to antimicrobial susceptibility testing. This system utilizes biochemical reactions and optical readings to rapidly and accurately identify bacteria and assess their resistance or susceptibility to various antibiotics. In conferment with Clinical and Laboratory Standards Institute (CLSI) instructions,[7] minimum inhibitory concentrations (MICs) were concluded and elucidated.

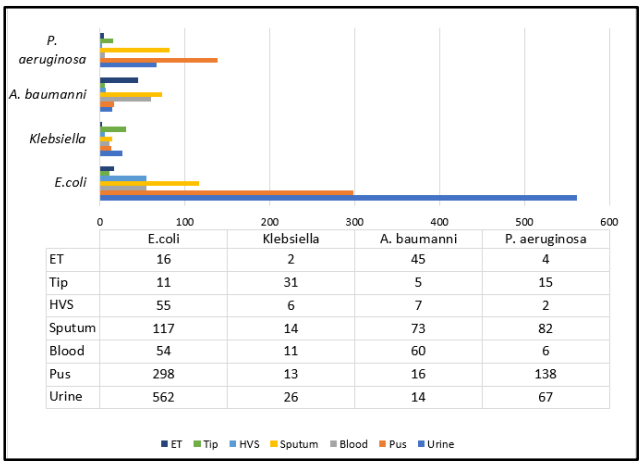
Carbapenem resistance was defined as a MIC of ≥16 microgram/mL pointed at either meropenem or imipenem, as determined by VITEK 2. Resistance was confirmed if the microorganism showed reduced susceptibility toward any of the carbapenem tested antibiotics together with meropenem, imipenem or ertapenem.[18]

3. Results

Among 7,224 clinical samples processed, 1,750 (24.2%) were identified as culture-positive for bacterial growth. The distribution of Paediatric isolates across different sample types revealed that 40% of the pathogens were recovered from urine samples, followed by pus samples (28.6%), sputum samples (14.1%), high vaginal swabs (HVS) (6.5%), and blood (5%). Incidence of Gram-negative isolates was less frequently isolated from catheter tips (3.7%) and endotracheal (ET) samples (1.3%). Among 1750 (24.2%) culture-positive isolates, *Escherichia coli* was identified in 1113 cases, *Klebsiella pneumoniae* in 103 cases, *Acinetobacter baumannii* in 220 cases, and *Pseudomonas aeruginosa* in 314 cases.

The maximum frequently isolated Gram-negative bacteria from clinical samples (**Figure 1**) were *Escherichia coli* (63.6%), followed by *Pseudomonas aeruginosa* (18%), *Acinetobacter baumannii* (12.5%) and *Klebsiella pneumoniae* (5.8%). The carbapenem susceptibility profile revealed varying degrees of resistance, with *Acinetobacter*

baumannii showing the highest resistance to imipenem (36%) and *Pseudomonas aeruginosa* demonstrating over 50% sensitivity to imipenem (**Table 1**).



	E.coli	Klebsiella	A. baumanni	P. aeruginosa
ET	16	2	45	4
Tip	11	31	5	15
HVS	55	6	7	2
Sputum	117	14	73	82
Blood	54	11	60	6
Pus	298	13	16	138
Urine	562	26	14	67

Figure 1: Frequency of organism isolates across different sample sources

Comprehensive evaluation of carbapenem susceptibility was conducted for 1,750 Gram-negative isolates as summarized in **Table 1**. Among these, 639 (36.5%) were identified as carbapenem-resistant *Acinetobacter baumannii* (CRAB), carbapenem-resistant *Pseudomonas aeruginosa* (CRPA) and carbapenem-resistant Enterobacterales (CRE) The maximum carbapenem-resistant species was *Escherichia coli* (55.3%) followed by *Acinetobacter baumannii* 20.6%, and *Pseudomonas aeruginosa* 17%. The least isolated species that showed resistance to carbapenem was *Klebsiella pneumoniae*, found at a rate of 6.8%.

The majority of the CRO isolates were from ICUs (48.8%). The predominant isolate source of CRO was intensive care units (ICUs) accounting for 48.8% of the total. (**Figure 2**)

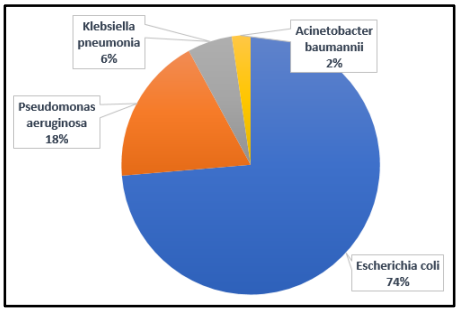


Figure 2: Organism wise distribution

The carbapenem susceptibility data (**Table 1**) reveals significant resistance among gram-negative isolates, with *Acinetobacter baumannii* showing the highest resistance to imipenem (36%) and meropenem (24%). In contrast, *Escherichia coli* demonstrated the lowest resistance rates across all three carbapenems, with 9.4% resistance to ertapenem and 9.7% to imipenem.

4. Discussion

The order Enterobacterales is commonly linked to severe infections, including nosocomial pneumonitis, community-acquired disease and complex peritoneal disease. Despite CRAB, CRPA and CRE represent a major concern are primarily contributing to higher death rates in addition to burden of illness. Consequently, research efforts focused on innovative drugs should prioritize CR-NLFs, notably *A.bumannii* and *P.aeruginosa*, alongside CRE.

The growing clinical challenge posed by Enterobacterales exhibiting carbapenemase production is largely attributable to the increased reliance on carbapenems for alleged ESBL disease.[19] Consistent with the world wide tendency of growing carbapenem resistance,[20] this study documented a 36.5% prevalence of CRE, CRPA and CRAB among clinical isolates at our healthcare facility. While carbapenems remain crucial for therapeutic management of Gram-negative infections with multidrug-resistant, the rising resistance necessitates careful antimicrobial stewardship.

Table 1: Carbapenem susceptibility data among 1,750, Gram-Negative bacterial isolates

Species	N	Imipenem(R/S)		Meropenem (R/S)		Ertapenem (R/S)	
		Resistant N (%)	Sensitive N (%)	Resistant N (%)	Sensitive N (%)	Resistant N (%)	Sensitive N (%)
Escherichia coli	1113	108 (9.7%)	225(20.2%)	141 (12.6%)	243 (21.8%)	105 (9.4%)	291 (26.1%)
Klebsiella pneumonia	103	15 (14.5%)	20 (19.4%)	16 (15.5%)	22 (21.3%)	13 (12.6%)	17 (16.5%)
Acinetobacter baumannii	220	79 (36%)	47 (21.3)	53 (24%)	41 (18.6%)	-	-
Pseudomonas aeruginosa	314	67 (30.4%)	111 (50.4%)	42 (19%)	94 (42.7%)	-	-

* N = Number of isolates, R = Resistant, S = Sensitive

*Intrinsic ertapenem resistance is observed in *A.baumannii* and *P.aeruginosa*

Srivastava et al. stated an incidence proportion of 29.35% in a North Indian hospital-based study,[21] conversely17% to 22% of resistance to carbapenem was documented by Gupta et al.[22] in the same region. Similarly, a 26% prevalence of resistant to carbapenem isolates in Mumbai was documented by Nair and Vaz.[23]

Wattal et al. reported in Delhi a wide range of 13% to 51% in carbapenem resistance among Enterobacterales isolates within tertiary healthcare.[24]

In the present study, carbapenem resistance was most commonly observed in *Escherichia coli* (55.3%), followed by the non-fermenter *A. baumannii* (20.6%) and least was reported from *Klebsiella pneumoniae* (6.8%). Findings from Grewal et al.[25] and Bandyopadhyay et al.[26] corroborate the observed incidence of CRAB (20.6%) and CRPA (17%) in the present study. Another study from Oman reported carbapenem resistance rates of 80.4% in *A. baumannii*, 46.4% in *K. pneumoniae*, and lower rates in other species.[27] Benachinmardi et al.[28] highlighted a higher prevalence of *P. aeruginosa* infections but noted a recent upward trend in *A. baumannii* infections. Cai et al.[29] reported a higher incidence, with 45% of *A.baumannii* and 19% of *P.aeruginosa* isolates exhibiting carbapenem resistance.

Carbapenem resistance also contrasts depending type of disease. Current research found most carbapenem-resistant organisms (CROs) were isolated from urine samples (32.8%), followed by pus samples (18%), which aligns with previous reports indicating an advanced prevalence of CR isolates in urine and pus compared to blood samples.[30,31] Indian research supports our findings, which also reported the urine specimens demonstrated the maximum prevalence of CRE, with a rate of 26.36%,followed by pus (24.54%) and blood (20%).[32] Similarly, another study from northern India found that carbapenem-resistant organisms were most frequently as urine (47.1%) as the primary isolate source followed by pus (27.1%).[33] According to Mohamudha et al.[34] urine samples yielded 37% of the isolates were resistant, with blood and wound discharge accounting for 22.3% and 11.7%, respectively

Isolate distribution in our study (48.8% ICUs, 36% wards, 15% OPDs) matched prior findings.[30,31,35,36] Our findings highlight important trends in carbapenem resistance amongst clinically relevant bacterial strains.

This resistance was observed to be predominantly prevalent in isolates from intensive care units (ICUs) and in urine and pus samples. The growing incidence of carbapenem-resistant organisms presents substantial challenges for infection control. Strengthening antimicrobial stewardship programs, along with enhanced surveillance and improved hygiene practices, is essential to mitigate the spread of multidrug-resistant organisms.

To better inform clinical practice and antimicrobial stewardship, future research should focus on determining the molecular basis of carbapenem resistance, defining regional resistance patterns, and assessing the effectiveness of emerging antimicrobial therapies.

5. Conclusion

Present research highlights concerning frequency of isolates resistance to carbapenem between clinically significant Gram-negative bacilli, reinforcing the need for close monitoring of this growing threat. The highest levels of resistance were observed in *A.baumannii, P.aeruginosa, E.coli* and *K.pneumoniae* posing significant trial aimed at infection management in high-risk healthcare settings.

Infections caused by carbapenem-resistant organisms result in prolonged healthcare stays, elevated death rates, and augmented healthcare expenditures. Additionally, carbapenem-resistant non-fermenting infections (CR-NLFs) like *P.aeruginosa* and *A.baumannii* must remain comprised for progress of novel drugs, alongside carbapenem-resistant *Enterobacteriaceae* (CREs).

6. Conflict of Interest

The author(s) affirm that no potential conflicts of interest exist regarding the research, writing, or publication of this study

7. Source of funding

None.

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