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To determine the prevalence of carbapenem resistance among clinical isolates of Acinetobacter baumannii

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Abstract

Introduction: The prevalence of antibiotic-resistant pathogens has become a major public health concern globally. It is known to cause a wide range of infections, including ventilator-associated pneumonia, bloodstream infections, urinary tract infections, meningitis, and wound infections, particularly in immunocompromised patients or those with prolonged hospital stays. **Objectives:** To determine the prevalence of carbapenem resistance among clinical isolates of *Acinetobacter baumannii.* **Methodology:** The prospective observational study was conducted for 24 months and jointly in the Department of Microbiology at SGT University, Gurugram, and the Department of Microbiology at Pt. B. D. Sharma PGIMS, Rohtak. **Results**: Out of the 200 *A. baumannii* isolates, 82% were resistant to carbapenems, and 4% were resistant to colistin. Most isolates (96%) remained susceptible to colistin. **Conclusion**: Colistin remains effective for the majority of CRAB infections, though resistance is emerging. Ongoing surveillance and careful antibiotic stewardship are essential to managing the spread of resistance in clinical settings.

Introduction

The prevalence of antibiotic-resistant pathogens has become a major public health concern globally. Among these, *Acinetobacter baumannii* has emerged as one of the most worrisome pathogens due to its ability to rapidly acquire resistance to multiple classes of antibiotics, making infections difficult to treat. *Acinetobacter baumannii* is an opportunistic pathogen, primarily associated with nosocomial (hospital-acquired) infections, especially in intensive care units (ICUs).[1] It is known to cause a wide range of infections, including ventilator-associated pneumonia, bloodstream infections, urinary tract infections, meningitis, and wound infections, particularly in immunocompromised patients or those with prolonged hospital stays. One of the most pressing concerns regarding *A. baumannii* is its ability to develop resistance to carbapenems, a class of β-lactam antibiotics that are often used as a last resort for treating multidrug-resistant (MDR) bacterial infections.[2] Carbapenem-resistant *Acinetobacter baumannii* (CRAB) has become increasingly prevalent worldwide, posing a severe threat to public health due to the limited treatment options available. The World Health Organization (WHO) has recognized carbapenem-resistant *A. baumannii* as a critical priority pathogen, emphasizing the need for new therapeutic strategies and enhanced surveillance to control its spread.[3] Carbapenem resistance in *A. baumannii* is often mediated through various mechanisms, including the production of carbapenemases (enzymes that degrade carbapenems), alterations in porin channels (which reduce drug entry into bacterial cells), efflux pumps (which expel antibiotics from the cell), and mutations in penicillin-binding proteins.[4] These mechanisms allow *A. baumannii* to survive in the presence of carbapenems and other antibiotics, making infections caused by this pathogen difficult to eradicate.[5]The increasing prevalence of CRAB has been linked to several factors, including the overuse and misuse of antibiotics, the ability of *A. baumannii* to persist in hospital environments, and the transfer of resistance genes through horizontal gene transfer. Additionally, the pathogen's ability to form biofilms on surfaces and medical devices further complicates infection control efforts.[6] In hospitals, *A. baumannii* can survive on surfaces for extended periods,

contributing to its transmission between patients and healthcare workers. This study aims to determine the prevalence of carbapenem resistance among clinical isolates of *A. baumannii* to better understand the scope of the problem in healthcare settings.[7] Surveillance of antibiotic resistance patterns is essential for informing treatment guidelines and infection control measures. By analyzing clinical isolates, this research will provide insight into the frequency of carbapenem resistance and its potential impact on patient outcomes. Infections caused by Multi Drug Resistant organisms (MDRO) are one of the major issues in healthcare system worldwide.[8] Factors predisposing to MDR infection are the long-term stay in health care facilities, injudicious use of broad-spectrum antibiotics, multiple comorbid condition, multiple invasive procedures and elderly age group. Gram-negative organisms are seen to be more prevalent and are more resistant as compared to gram-positive organisms, especially in developing countries. [9]Among gram-negative non-fermenting bacteria, *Acinetobacter* strains are emerging as one of the important causes of nosocomial infection in critically ill patients. It inhabits skin, mucous membranes, and soil, and can also survive for long periods on dry and moist surfaces. *Acinetobacter baumannii* are glucose non-fermentative, non-fastidious, non-motile, catalase-positive, aerobic, gram-negative coccobacilli.[10] It is one of the major hospital-associated pathogen which is responsible for the respiratory tract, bloodstream, urinary tract, surgical site, and wound infection. The Infectious Diseases Society of America has included *Acinetobacter baumannii* among the 6 antimicrobial-resistant pathogens responsible for high morbidity and mortality in patients.[11] This organism exhibits wide range of intrinsic and acquired resistance to multiple classes of antibiotics especially extended-spectrum cephalosporins, aminoglycosides and fluoroquinolones. The introduction of carbapenem was often the only drug of choice for infection caused by these multi-drug resistance organisms (MDRO). Injudicious use of this class of drug has led to the emergence of resistance to carbapenem worldwide.[12]

Objectives

To determine the prevalence of carbapenem resistance among clinical isolates of *Acinetobacter baumannii.*

Material and Methods

The prospective observational study was conducted for 24 months and jointly in the Department of Microbiology at SGT University, Gurugram, and the Department of Microbiology at Pt. B. D. Sharma PGIMS, Rohtak.

The sample size was calculated using Cochran's formula, assuming the estimated prevalence of carbapenem-resistant *A. baumannii* to be between 45% and 75%. The formula is as follows:

N=Z2×P(1−P)d2N = \frac{Z^2 \times P(1 - P)}{d^2}N=d2Z2×P(1−P)

Where:

- **N**: Minimum number of samples required
- **Z**: Standard normal distribution at a 5% significance level (1.96)
- **P**: Estimated prevalence of carbapenem-resistant *A. baumannii* (50%, or 0.5)
- **d**: Degree of precision (5%, or 0.05)

Using this formula, the calculated sample size is approximately 200.

Sample Collection:

All consecutive non-replicative isolates of *Acinetobacter baumannii* from clinical samples collected at SGT University, Gurugram, and Pt. B. D. Sharma PGIMS, Rohtak, were included.

Data collection

Clinical samples, including blood, urine, respiratory, and wound swabs, were collected from patients. Gram-negative bacterial isolates from these samples was cultured and identified using standard microbiological techniques. Biochemical tests such as citrate utilization, Oxidative-Fermentative (dextrose), catalase, arginine dihydrolase, and growth at 44°C was performed to confirm the identity of *Acinetobacter* species, following established microbiological guidelines. Blood samples were processed by inoculating them into glucose broth. The broth was sub-cultured onto blood agar and MacConkey agar plates after 24 hours, 48 hours, 72 hours, and on the 7th day. Colony morphology was observed for any growth, and further identification was performed using the VITEK automated system. All confirmed *A. baumannii* isolates were undergo antimicrobial susceptibility testing. The antimicrobial susceptibility of *A. baumannii* isolates were tested using the Kirby-Bauer disc diffusion method on Mueller-Hinton Agar (MHA), following Clinical and Laboratory Standards Institute (CLSI) guidelines. Control strains, including *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853, were used for quality assurance.[13,14]

The following antimicrobial discs, procured from Hi-Media Laboratories, Mumbai, were used in the testing:

- Gentamicin (10µg)
- Amikacin (30µg)
- Tobramycin (10µg)
- Ampicillin/sulbactam (10µg/10µg)
- Ticarcillin/clavulanic acid (75µg/10µg)

- Cefepime (30µg)
- Ceftriaxone (30µg)
- Cefotaxime (30µg)
- Ciprofloxacin (5µg)
- Levofloxacin (5µg)
- Ertapenem (10µg)
- Imipenem (10µg)
- Meropenem (10µg)
- Trimethoprim/sulfamethoxazole (1.25µg/23.75µg)
- Aztreonam (30µg)
- Ceftazidime (30µg)
- Piperacillin/tazobactam (100µg/10µg)

Isolates that demonstrate resistance to imipenem, meropenem, or ertapenem will be further processed for colistin resistance.

Statistical analysis

The data collected is analyzed with the help of a software package (SPSS version 21.0). Frequency distribution and cross-tabulation were used to create summary tables and compare items within and across various categories. Association was tested using Chi-square and a "p" value of <0.05 was considered critical for statistical significance.

Results

During the 24-month study period, a total of 200 non-replicative clinical isolates of *Acinetobacter baumannii* were collected from various clinical samples, including blood, respiratory secretions, urine, and wound swabs. The study included 200 patients, with a male predominance (71%) compared to females (29%). The majority of patients fell within the 15-64 years age group (62%), followed by the 0-14 years age group (24%) and patients aged 65 and above (14%). Age-wise distribution by sex showed that males were predominant in all age categories, with 91 males and 34 females in the 15-64 age group. The distribution of isolates by sample type revealed that blood samples accounted for the largest proportion (56%), followed by respiratory samples (34%), body fluid (8%), and HVS (2%).

Table 1: Distribution of patients

Category	Subcategory	No. of Patients/Isolates	Percentage (%)
Distribution of Patients by Sex	Male	142	71
	Female	58	29
Age-Wise Distribution of Patients	0-14 years	48	24
	15-64 years	125	62
	65 years and above	27	14
Age-Wise Distribution by Sex	0-14 years (Male)	35	13
	0-14 years (Female)	13	5
	15-64 years (Male)	91	34
	15-64 years (Female)	34	14
	65 years and above (Male)	16	11
	65 years and above (Female)	11	5
Distribution of Isolates by Sample Type	Blood	112	56
	Respiratory	68	34
	Body Fluid	17	8
	HVS	3	2

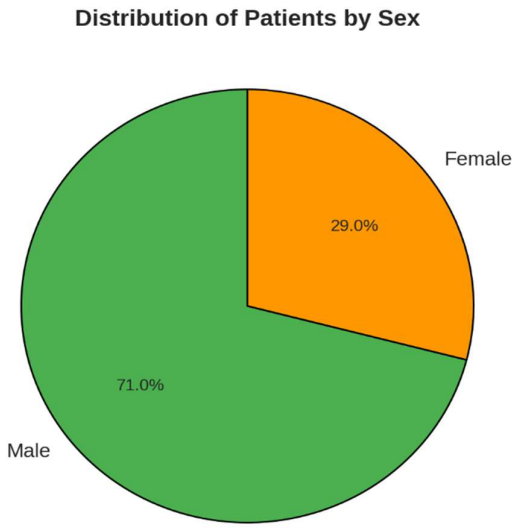

Figure 1: Distribution of isolates by gender

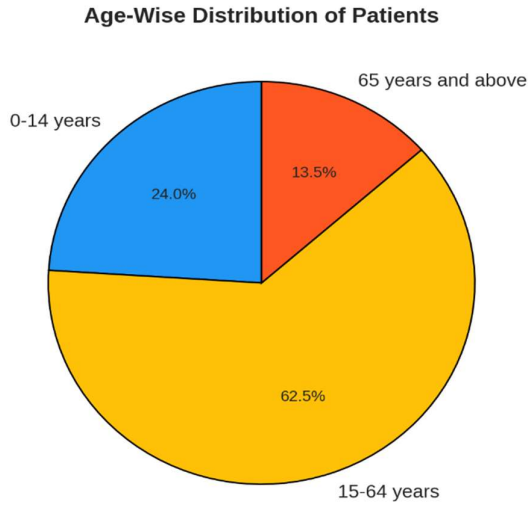

Figure 2: Age-wise distribution of patients

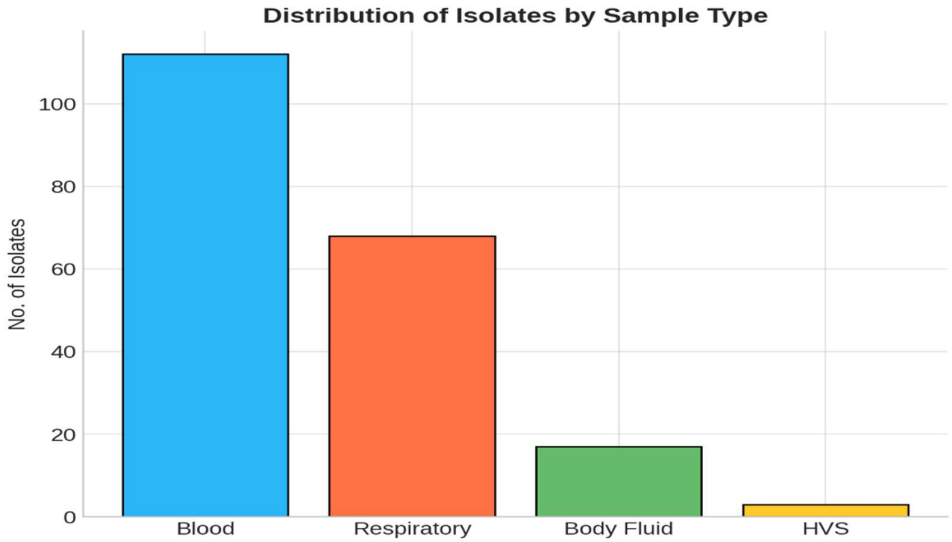

Figure 3: Distribution of isolated by sample type

The antibiogram results show a high level of resistance among *Acinetobacter baumannii* isolates to multiple antibiotics. Amikacin exhibited the highest resistance rate at 85%, followed by imipenem at 82%. Aztreonam, cefotaxime, and ceftazidime showed resistance levels of 80%, 78%, and 75%, respectively. Ciprofloxacin and gentamicin had lower resistance rates, with 65% and 60% resistance. Sensitivity rates were consistently low, with the highest sensitivity observed for gentamicin at 40%, indicating a significant challenge in treating these infections with standard antibiotics.

Table 2: Antibiogram of Acinetobacter baumannii

Antibiotic	Resistant (%)	Sensitive (%)
Amikacin	85	15
Aztreonam	80	20
Cefotaxime	78	22
Ceftazidime	75	25
Ciprofloxacin	65	35
Gentamicin	60	40
Imipenem	82	18

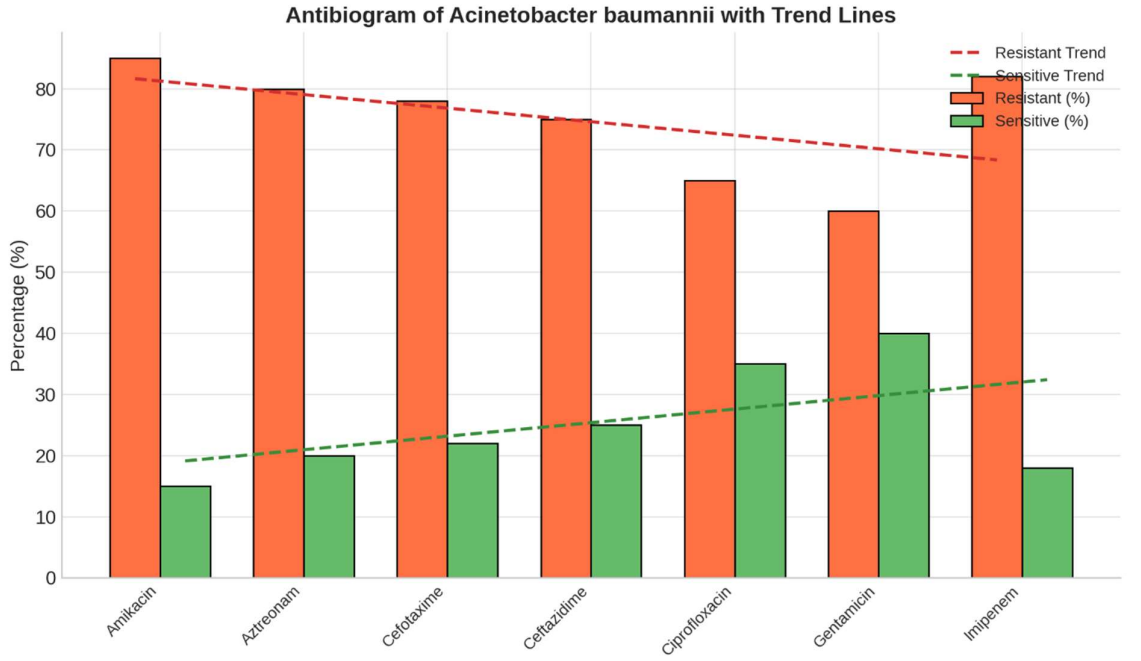

Figure 4: Antibiogram of Acinetobacter baumannii

Discussion

The emergence of antibiotic resistance in *Acinetobacter baumannii*, particularly carbapenem resistance, poses a significant threat to global health. Carbapenem-resistant *A. baumannii* (CRAB) has been a challenge to manage due to its ability to develop multidrug resistance (MDR), leaving clinicians with limited therapeutic options. In this study, we observed a concerning prevalence of carbapenem resistance among clinical isolates, with a particular focus on colistin resistance trends.[15] Colistin has long been considered the last line of defense against MDR Gram-negative pathogens, including CRAB. In our study, **4%** of CRAB isolates demonstrated resistance to colistin, with an MIC value of ≥ 4 µg/ml, highlighting a small but significant emergence of colistin-resistant strains. While the majority of isolates remained susceptible to colistin (96%), this resistance rate indicates the slow erosion of colistin's efficacy in treating these infections[16].The development of colistin resistance can be attributed to various factors, including its increased usage as a last-resort antibiotic, particularly in regions with high rates of carbapenem resistance. Resistance mechanisms in *A. baumannii* often involve alterations in the bacterial outer membrane or modifications in lipopolysaccharides, which reduce the binding efficacy of colistin. Additionally, the horizontal transfer of colistin-resistance genes, such as **mcr-1**, has been implicated in the spread of colistin-resistant *A. baumannii* in several regions of the world.[15,16] The presence of colistin resistance significantly complicates the treatment landscape for CRAB infections. Colistin resistance in *A. baumannii* is associated with higher mortality rates, increased length of hospital stays, and greater healthcare costs due to the need for alternative and often more toxic or less effective treatments, such as polymyxins or experimental therapies. In cases where colistin is ineffective, clinicians may resort to combination therapies, utilizing drugs like tigecycline or fosfomycin alongside colistin, though this approach remains controversial and not universally effective[16].Our findings suggest that,

while colistin resistance is still relatively uncommon, its emergence highlights the need for vigilant antibiotic stewardship.[17] The misuse or overuse of colistin, particularly in healthcare settings, can accelerate the development of resistance and lead to outbreaks of colistin-resistant *A. baumannii* strains. The prevalence of CRAB and the emergence of colistin resistance are not isolated to this study but are part of a larger global trend.[18] Countries in Asia, Europe, and the Middle East have reported similar or higher rates of colistin resistance among CRAB isolates. This reinforces the urgent need for global surveillance programs to track resistance patterns and inform public health interventions.[19] Despite the valuable findings of this study, certain limitations should be acknowledged. First, the study was conducted in two specific hospitals, limiting the generalizability of the results. Further multicentric studies across diverse regions are needed to better understand the spread of colistin resistance in different healthcare settings.

Conclusion

It is concluded that colistin remains an effective treatment for most carbapenem-resistant *Acinetobacter baumannii* (CRAB) isolates, though a concerning 4% have developed resistance. The emergence of colistin-resistant strains highlights the need for vigilant antibiotic stewardship and continued surveillance.

REFERENCES

1. Rossi F, Girardello R, Cury AP, Di Gioia TS, Almeida JN Jr, Duarte AJ. Emergence of colistin resistance in the largest university hospital complex of São Paulo, Brazil, over five years. Braz J Infect Dis.2017;21(1):98-101.
2. Mohanty S, Maurya V, Gaind R, Deb M, Phenotypic characterization and colistin susceptibilities of carbapenem resistant of *Pseudomonas aeruginosa* and *Acinetobacter spp*. J Infect Dev Citries 2013;7(11):880-7.
3. Montefour K, Frieden J, Hurst S, Helmich C,Headley D, Martin M, et al. *Acinetobacter baumannii* An emerging multidrug resistant pathogen in critical care. Aacnjournals 2008;28(1):15-25.
4. Ko KS, Choi Y, Lee JY. Old drug, new findings: Colistin resistance and dependence of Acinetobacter baumannii. Pfm journal2017;1(4):159-67.
5. Cai Y, Chai D, Wang R, Liang B, Bai N. Colistin resistance of *Acinetobacter baumannii*: clinical reports, mechanisms and antimicrobial strategies. J Antimicrobial Chemother2012;67(7):1607-15.
6. Qureshi ZA, Hittle LE, O'Hara JA, Rivera JI, Syed A, Shields RK, et al. Colistin-resistant Acinetobacter baumannii: beyond carbapenem resistance. Clin Infect Dis 2015;60: 1295-303.
7. Kempf M, Rolain JM. Emergence of resistance to carbapenems in Acinetobacter baumannii in Europe: clinical impact and therapeutic options. Int J Antimicrob Agents2012;39(2):105-14.
8. Boucher HW, Talbot GH, Benjamin KD Jr, Bradley JS, Guidos RJ, Jones RN, et al. 10 X '20 progress-Development of new drugs active against gram-negative bacilli: An update from the Infectious Disease Society of America. Clin Infect Dis 2013;56(12):1685-94.
9. Sinirtas M, Akalin H, Gedikoglu. Investigation of colistin sensitivity via three different methods in *Acinetobacter baumannii* isolates with multiple antibiotic resistance. Int J Infect Dis 2009;13(5):217-20.
10. Lesho E, Yoon EJ, McGann P. Emergence of colistin-resistance in extremely drug-resistant Acinetobacter baumannii containing a novel pmrCAB operon during colistin therapy of wound infections. J Infect Dis 2013; 208(7):1142–51.
11. Lopez-Rojas R, McConnell MJ, Jimenez-Mejias ME, Dominguez-Herrera J, Fernandez-Cuenca F, Pachon J. Colistin resistance in a clinical Acinetobacter Baumannii strain appearing after colistin treatment: effect on virulence and bacterial fitness. Antimicrob Agents Chemother 2013; 57(9):4587–9.
12. Caniaux I, van Belkum A, Zambardi G, Poirel L, Gros MF. MCR:modern colistin resistance. Eur J Clin Microbiol Infect Dis.2016;36(3):415-42.
13. Clinical and laboratory standard institute (CLSI). Performance standards for antimicrobial susceptibility testing; 28the. CLSI M100-S28. Wayne, PA: CLSI; 2018:30-8
14. Clinical and laboratory standard institute (CLSI). Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; 10the. CLSI document M07-A10.Wayne, PA: CLSI; 20159.
15. The Infectious Diseases Society of America. 10 X '20 Initiative: Pursuing a global commitment to develop 10 new antibacterial drugs by 2020.Clin Infect Dis 2010; 50(8):1081–3.
16. Who.int [Internet].Geneva; World Health Organization: WHO publishes list of bacteria for which new antibiotics are urgently needed, 2018;[cited 2018]. Available from: http://www.who.int/news-room/detail/27-02-2017-who-publishes-list-of-bacteria-for-which-new-antibiotics-are-urgently-needed.

17. Taneja N, Singh G,Singh M, Sharma M. Emergence of tigecycline & colistin resistant *Acinetobacter baumanii*in patients with complicated urinary tract infections in north India. Ind. J Med. Res.2011;133(6):681-4.
18. Al-Swein N, Al-Hubil MA, Rotimi VO. Emergence of tigecycline and colistin resistance in acinetobacter species isolated from patient in Kuwait. Jn of chemo.2011;23(1):13-6.
19. Chang KC, Lin MF, Lin NT, Wu WJ, Kuo HY, Lin TY et al. Clonal spread of multidrug-resistant Acinetobacter baumannii in eastern Taiwan. J Microbiol Immunol Infect2012; 45(1):37–42.