

Prevalence of *Acinetobacter* spp. and Antibiotic Susceptibility Pattern in Various Clinical Samples in a Tertiary Care Hospital

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ABSTRACT

Background: *Acinetobacter* spp. are opportunistic gram-negative pathogens responsible for severe hospital-acquired infections, particularly in immunocompromised and device-dependent patients. *A. baumannii* is the most clinically significant species, exhibiting remarkable environmental persistence and extensive antimicrobial resistance, including carbapenem resistance. Its diverse resistance mechanisms drive multidrug resistance, making treatment challenging and heightening morbidity, especially in ICU settings, thereby necessitating continuous surveillance and susceptibility profiling.

Methods: A retrospective study (January–June 2025) was conducted in the Microbiology Laboratory, D. Y. Patil University, Navi Mumbai. Clinical samples (urine, CSF, pus, pleural fluid, endotracheal secretions, BAL) were processed, and *Acinetobacter* spp. were identified using standard methods; antimicrobial susceptibility testing was performed according to CLSI guidelines.

Result: During the study period, a total of 2,582 clinical samples were processed, of which 69 (2.67%) yielded *Acinetobacter* spp. isolates. The highest prevalence was observed in endotracheal (ET) secretions (28.1%, 9/32). This indicates that *Acinetobacter* spp. was most frequently isolated from respiratory specimens. Across all specimens, colistin showed 100% sensitivity, making it the most effective drug. In urine isolates, the highest susceptibility was to colistin (100%). CSF isolates demonstrated 50% sensitivity to cefepime and carbapenems. Pus isolates showed the best response to colistin (100%).

Conclusion: The study concluded that the prevalence of *Acinetobacter* spp. varies considerably across clinical specimens, with the highest detection observed in endotracheal aspirates and markedly lower rates in urine, cerebrospinal fluid, and pleural fluid samples.

Key-words: *Acinetobacter baumannii*, Antimicrobial resistance, Multidrug resistance, Carbapenem resistance, Antibiotic susceptibility pattern, Colistin

INTRODUCTION

Acinetobacter spp. is a gram-negative bacterium, mainly classified as an opportunistic infection. Recently, they have been termed nosocomial pathogens, which are quite common in patients with a weak immune system [1].

Patients receiving respiratory therapy and catheters are susceptible to the pathogen, leading to infections such as pneumonia, septicemia, wound sepsis, urinary tract infection, endocarditis, and meningitis.

The *Acinetobacter baumannii* complex comprises several species, most notably *Acinetobacter baumannii*, *Acinetobacter nosocomialis*, and *Acinetobacter pittii*, which together represent the most clinically significant members of the *Acinetobacter* genus. *A. baumannii* is the most significant among all of the species. Most clinical infections have been caused by *Acinetobacter* spp. among humans [2].

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A. baumannii can tolerate harsh conditions, showing resistance to other disinfectants. Strains are resistant to a broad range of antimicrobials, which include carbapenems, tetracyclines, colistin, and polymyxins. This is a major medical concern in the treatment of infections caused by pathogens, largely due to antimicrobial resistance mechanisms. The mechanism of *A. baumannii* against antimicrobial agents includes aminoglycoside-modifying enzymes, which alter penicillin-binding proteins to produce broad-spectrum β -lactamases, and changes in outer membrane proteins. This reduces the permeability of the drug to the cell membrane and upregulates the efflux pump, thereby increasing the prevalence of MDR *A. baumannii*^[3].

Treatment for *Acinetobacter* species is very difficult due to the intrinsic antibiotic resistance of these organisms, which confers resistance to other antibiotics^[4]. The transmission of antimicrobial resistance is stimulated by various factors, including the use of antibiotics, infection control practices, and increased international travel.

WHO reports that carbapenem-non-susceptible *Acinetobacter* spp. is the most antimicrobial-resistant pathogen, resulting in severe morbidity among patients admitted to the ICU. *A. baumannii* can cause life-threatening infections such as meningitis, ventilator-associated pneumonia, bacteremia, wound, and urinary tract infections. Antibiotic resistance in *Acinetobacter* spp. has developed through exposure to soil microbes during antibiotic production. They are most likely to be present in damp and dry areas^[5]. Among *Acinetobacter* spp., *Acinetobacter baumannii* has developed multiple resistance mechanisms to commonly used antimicrobial classes, including penicillins, cephalosporins, tetracyclines, aminoglycosides, and fluoroquinolones, complicating its clinical management. Many studies have reported an increased prevalence of MDR strains of *A. baumannii*. This has been increasing in South Asia, the Arabian Peninsula, and other parts^[6]. The prevalence of infections has been increasing due to drug resistance in *Acinetobacter* spp., posing challenges for treatment, and increasing the economic burden. Also, limited studies have been conducted to identify the incidence and drug resistance in *Acinetobacter* spp. More research studies have been conducted to develop strategies and treatments for resistant cases^[7]. *Acinetobacter* spp. has been associated with a high rate of mortality of 32 to

52% for the bloodstream infection. Also, 70% of mortality has been observed in the critical care cases. The identification of *Acinetobacter* spp. from clinical samples is essential. The emergence of multidrug-resistant variants of *Acinetobacter* spp., which cause nosocomial infections, has been a global concern among the scientific community. Eradication of nosocomial outbreaks is challenging due to the intrinsic and acquired resistance of *Acinetobacter* species to multiple antibiotics, which facilitates persistent colonisation and hospital transmission. Consequently, systematic evaluation of *in-vitro* antimicrobial susceptibility patterns and accurate identification of antibiotic resistance mechanisms are essential for guiding appropriate therapy and improving clinical outcomes in infections caused by *Acinetobacter*^[8]. The study aims to investigate the antibiotic susceptibility patterns of *Acinetobacter* and the prevalence of the microbe in various clinical samples obtained from a tertiary care hospital.

MATERIALS AND METHODS

Research design- This is a retrospective study to determine the prevalence of *Acinetobacter* species and their susceptibility patterns across different clinical samples. The study was conducted in the Microbiology Laboratory of D. Y. Patil Medical College and Hospital, Navi Mumbai, over 6 months from January to June 2025. Various clinical samples were received, including urine, cerebrospinal fluid (CSF), pus, pleural fluid, endotracheal secretions, and bronchoalveolar lavage (BAL). Ethics approval was obtained, and written and verbal informed consent were obtained from the ethics board. Data analysis was performed to determine the prevalence of *Acinetobacter* spp., assess antibiotic sensitivity, and identify resistance patterns. Patients were selected according to specific inclusion and exclusion criteria.

Inclusion criteria

- Patients of above 18 years of age were selected for the study.
- Both male and female patients were selected for the study.

Exclusion criteria

- Any repeated samples of the same patients were not included in the study.

Procedure- The study procedure involved a retrospective evaluation of clinical samples collected from various hospital departments. All specimens underwent initial microscopic examination and Gram staining. Following this, samples were inoculated onto Blood agar and MacConkey agar media for bacterial isolation and analysis. The inoculated culture plates were incubated aerobically at 37 °C for 18–24 hours.

Bacterial isolates were identified using standard microbiological techniques based on colony morphology and preliminary tests, including Gram staining, catalase test, oxidase test, and motility assessment. Further biochemical characterization was carried out using indole production, citrate utilisation, urease test, triple sugar iron agar reaction, and phenylalanine deaminase test to confirm the identification of *Acinetobacter* spp. Additional confirmatory tests specific for *Acinetobacter baumannii* included oxidative/ fermentation glucose reaction, arginine decarboxylation, and growth at 42 °C. Antibiotic susceptibility testing was performed using the Kirby–Bauer disc diffusion method on Mueller–Hinton agar, in accordance with Clinical and Laboratory Standards Institute (CLSI) guidelines. A panel of antibiotic discs representing major antimicrobial classes was used, including cephalosporins, aminoglycosides, fluoroquinolones, beta-lactam/beta-lactamase inhibitor combinations, and carbapenems. Isolates exhibiting resistance to three or more antimicrobial classes were classified as multidrug-resistant (MDR), while those resistant to all tested antibiotics were designated as pan-resistant.

Minimum inhibitory concentration (MIC) testing for colistin and minocycline was performed and interpreted according to CLSI guidelines. Colistin susceptibility was defined as an MIC \leq 2 µg/mL and resistance as \geq 4 µg/mL. Minocycline susceptibility was defined as an MIC \leq 4 µg/mL, intermediate susceptibility as 8 µg/mL, and resistance as \geq 16 µg/mL. These agents were evaluated as reserve antibiotics for MDR *Acinetobacter* species, with MIC-based interpretation emphasized due to the limited reliability of disc diffusion for colistin and the clinical importance of accurate susceptibility assessment.

Outcome assessment- The outcome assessment shows the prevalence of *Acinetobacter* spp. across different clinical samples and has identified *A. baumannii* as the predominant strain. Antibiotic susceptibility tests have

been performed by observing resistance patterns to different antibiotic discs, and isolates have been categorised as multidrug-resistant (MDR) or pan-resistant.

Statistical Analysis- Data analysis was entered and recorded in Microsoft Excel. Descriptive statistical procedure was used, and proportions and percentages were used for data analysis. Tables and graphs were used for the representation of data. Generation of data was performed by the use of Microsoft Word and Microsoft Excel.

Ethical Approval- The ethical approval was procured from the Institutional Ethics Board.

RESULTS

A total of 2,582 clinical samples were processed in this study, of which 69 (2.67%) yielded *Acinetobacter* spp. Isolates. The age distribution shows a broad and continuous spread of cases across the adult lifespan, with notable clustering in the middle and early older age groups. The highest frequencies appear in the 45–49 and 60–64-year intervals, indicating two prominent peaks in midlife and the early elderly population. Substantial representation is also evident in the 35–39 and 55–59-year groups, suggesting sustained case concentration across the mid-adult to pre-senior age range. Beyond 70 years, the frequency decreases steadily between 70 and 79 years, and there is only sporadic representation in those aged 80 and above. Overall, the pattern demonstrates a clear predominance of middle-aged and early older adults within the study sample (Fig. 1).

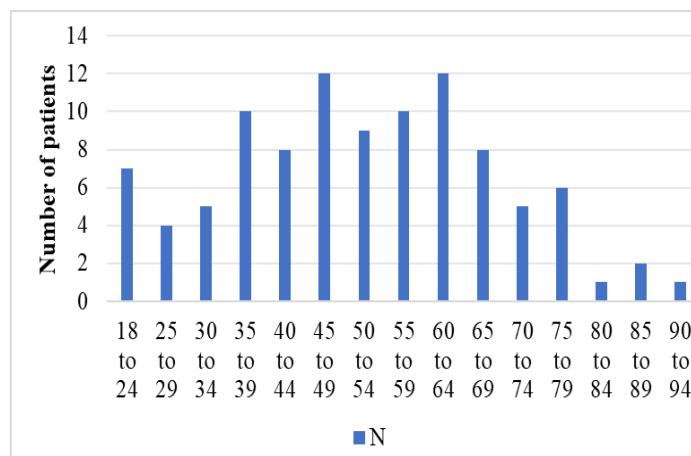


Fig. 1: Age distribution of the study sample

The distribution of sample types across sexes reveals substantial variation in representation. Urine constitutes the majority of all samples at 52.18%, with a markedly higher proportional contribution from females, who account for 65.52% of all urine specimens compared with 42.50% among males. Pus samples from the second-largest category at 24.64%, with predominance among males (27.50%) relative to females (20.69%). Endotracheal secretions represent 13.05% of the dataset, with males contributing 15.00% and females 10.35%. Bronchoalveolar lavage, comprising 4.35% of

total specimens and with a male predominance of 7.50%, while cerebrospinal fluid samples show near-equivalent representation between sexes, contributing 2.90% overall. Pleural fluid, also at 2.90%, is observed exclusively among males. Collectively, these findings indicate that females show a higher rate in urine samples. In contrast, males show higher representation in pus, endotracheal, pleural, and BAL samples, reflecting underlying clinical and anatomical influences on sampling patterns (Table 1).

Table 1: Distribution of sample types among males and females

Sample Type	Total	Total (%)	Males	Males (%)	Females	Females (%)
BAL	3	4.35	3	7.50	0	0
CSF	2	2.90	1	2.50	1	3.45
ET Secretion	9	13.05	6	15.00	3	10.35
Plural fluid	2	2.90	2	5	0	0
Pus	17	24.64	11	27.50	6	20.69
Urine	36	52.18	17	42.50	19	65.52
Total	69	100	40	100	29	100

A total of 69 *Acinetobacter* spp. Isolates were analysed from urine (36), CSF (2), pus (17), and respiratory samples (14). Fig. 2 shows variation in positivity across specimen types, with endotracheal aspirates exhibiting an exceedingly high prevalence of 28.10%, far exceeding that of any other sample category. In contrast, urine and cerebrospinal fluid exhibit nearly identical prevalence values of 2.07% and 2.08%, respectively, indicating comparable pathogen detection levels in these samples. Pleural fluid and bronchoalveolar lavage also fall within a

narrow range, with prevalences of 1.92 and 2.21%, suggesting minimal variation from the baseline pattern seen in urine and CSF. Pus samples, however, present a noticeably higher prevalence of 3.60%, almost double that of the low-prevalence groups, reflecting the inherently infectious nature of purulent material. When the dataset is examined collectively, the overall prevalence is 2.67%, but this aggregate is substantially influenced by the disproportionately elevated rate seen in endotracheal samples.

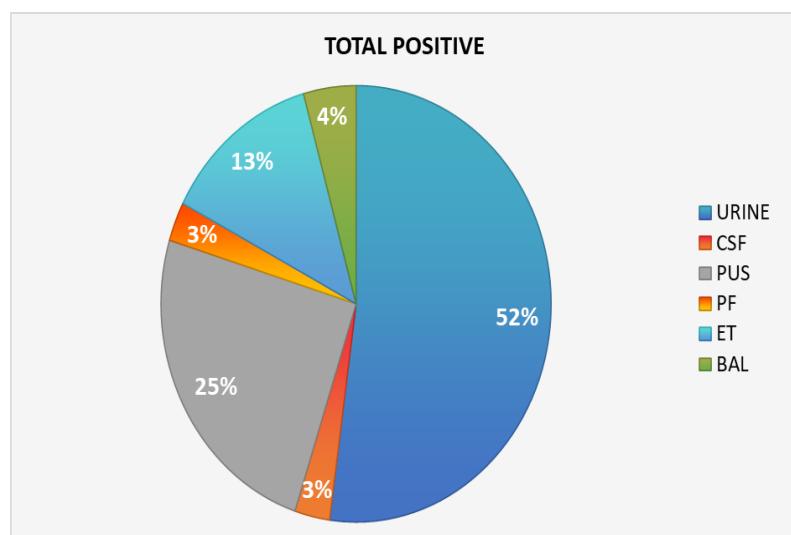


Fig. 2: Distribution of Positive Samples in this study

The antimicrobial sensitivity pattern shows a wide range of effectiveness among the tested drugs, with clear distinctions in activity against the isolates. Among the β -lactam antibiotics, piperacillin-tazobactam shows a modest sensitivity of 33%, while cefepime and ceftazidime exhibit sensitivities of 30% and 28% respectively. First- and third-generation cephalosporins, such as cefuroxime and ceftriaxone, perform poorly, with each demonstrating only 12% sensitivity. Carbapenems show comparatively higher activity, with imipenem at 35% and meropenem at 38%, though still far below expected efficacy. Doxycycline presents a sensitivity of 41%, like ciprofloxacin at 41% and levofloxacin at 43%,

whereas gentamicin performs slightly better at 43% and amikacin reaches 46%, indicating superior aminoglycoside activity. Trimethoprim-sulfamethoxazole demonstrates a moderate sensitivity of 49%, while norfloxacin used for urinary isolates yields 35%. The most active agent among all tested drugs is minocycline, with a striking 68% sensitivity, and colistin shows complete sensitivity at 100%. These findings collectively illustrate substantial variation in drug performance, with minocycline and colistin emerging as the most effective agents, while traditional β -lactams and several carbapenems demonstrate markedly reduced activity (Fig. 3).

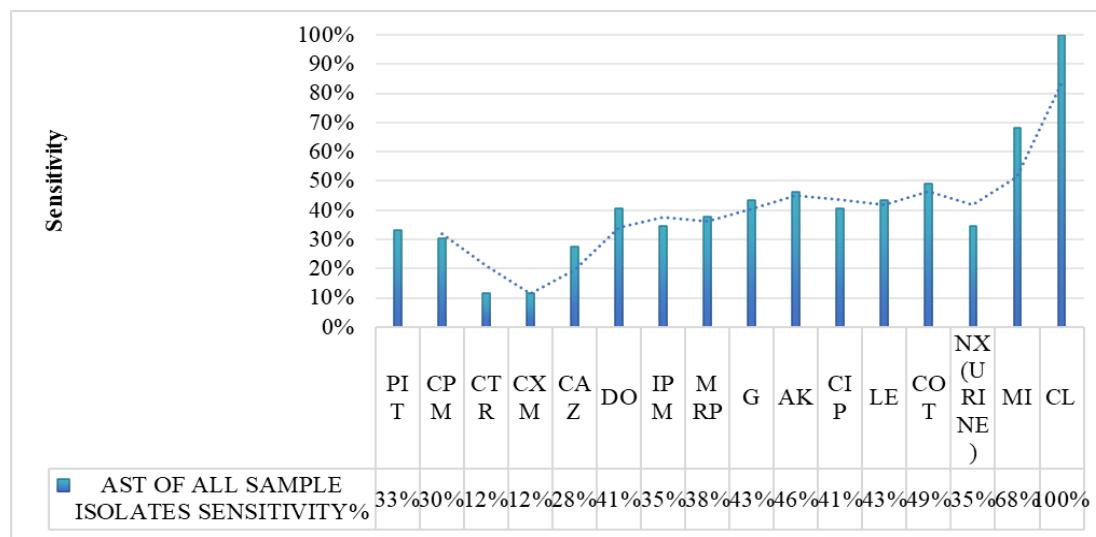


Fig. 3: Findings related to the Antibiotic Sensitivity Test

The antimicrobial susceptibility patterns across sample types reveal pronounced differences in drug performance and highlight clear gradients of sensitivity among the isolates. Across all samples, colistin demonstrates the highest activity with 100% sensitivity, followed by minocycline with 68%, establishing these as the most effective agents. In contrast, ceftriaxone and cefuroxime record the lowest sensitivities at 12%, reflecting minimal therapeutic value against the isolates. Drugs such as doxycycline, ciprofloxacin, levofloxacin, trimethoprim-sulfamethoxazole, gentamicin, and amikacin exhibit moderate effectiveness, ranging from 41% to 49% for the former group and 46% to 72% for aminoglycosides, depending on the specimen. When comparing drug responses across samples, urine isolates display comparatively high sensitivities to minocycline (75%), amikacin (72.2%), levofloxacin and gentamicin (69.4%), and colistin (100%). Cerebrospinal fluid isolates

remain uniformly distributed with 50% sensitivity to most agents and complete susceptibility to amikacin and colistin. Pus isolates demonstrate markedly low sensitivity to most beta-lactams such as ceftriaxone, cefuroxime, and cefepime (0–6%), but respond better to doxycycline (29%), trimethoprim-sulfamethoxazole (41%), minocycline (65%), and colistin (100%). Respiratory isolates, comprising bronchoalveolar lavage, endotracheal aspirates, and pleural fluid, exhibit inferior sensitivity to most drugs, with only levofloxacin and ciprofloxacin reaching 7%, trimethoprim-sulfamethoxazole reaching 14%, and minocycline achieving 57%; colistin again shows complete sensitivity at 100%. The study found that colistin is the most sensitive agent across all specimen types, while ceftriaxone and cefuroxime are consistently the least effective. Minocycline appears as a highly active 2nd line agent, particularly in urine, pus, and respiratory samples.

In clinical settings, beta-lactams such as piperacillin-tazobactam and cefepime are frequently used but exhibit modest activity, underscoring their limited utility against these isolates. Collectively, the data indicate that therapeutic effectiveness varies substantially by

specimen type, with urinary isolates showing comparatively favourable drug susceptibility and respiratory isolates demonstrating the greatest resistance (Table 2).

Table 2: Antimicrobial susceptibility patterns of *Acinetobacter* spp. isolates

	Pus	CSF						Urine						MRP	CIP	LE	COT	NX (URINE)
		%	Total	I	R	S	%	Total	I	R	S	%	Total	I	R	S		
24	17	1	13	4	50	2	0	1	1	50	36	0	18	18				PIT
6	17	0	16		50	2	0	1	1	52.8	36	0	17	19				CPM
0	17	0	17	0	50	2	0	1	1	19.4	36	0	29	7				CTR
0	17	0	17	0	50	2	0	1	1	19.4	36	0	29	7				CXM
12	17	0	15	2	50	2	0	1	1	41.7	36	0	21	15				CAZ
29	17	0	12	5	50	2	0	1	1	55.6	36	0	16	20				DO
18	17	0	14	3	50	2	0	1	1	55.6	36	1	16	20				IPM
24	17	0	13	4	50	2	0	1	1	58.3	36	0	15	21				
24	17	0	13	4	50	2	0	1	1	69.4	36	1	11	25				G
24	17	0	13	4	50	2	0	0	2	72.2	36	0	24	26				AK
24	17	0	13	4	100	2	0	1	1	61.1	36	1	14	22				
18	17	0	13	3	50	2	0	1	1	69.4	36	0	11	25				
41	17	1	10	7	50	2	0	1	1	66.7	36	0	12	24				
24	17	0	13	4	50	2	0	1	1	52.8	36	0	17	19				
65	17	0	6	11	50	2	0	1	1	75	36	0	9	27				MI
100	17	0	0	17	100	2	0	0	2	100	36	0	0	36				CL

All sample	Respiratory (BAL, ET, PF)						
	%	Total	I	R	S	%	Total
33	69	0	46	23	0	14	0
30	69	0	48	21	0	14	0
12	69	1	60	8	0	14	0
12	69	0	61	8	0	14	0
28	69	0	50	19	0	14	0
41	69	0	41	28	0	14	0
35	69	1	44	24	0	14	0
38	69	0	43	26	0	14	0
43	69	2	37	30	0	14	1
46	69	0	37	32	0	14	0
41	69	1	40	28	7	14	0
43	69	1	38	30	7	14	0
49	69	0	35	34	14	0	12
35	69	0	44	24	0	14	0
68	69	0	22	47	57	14	0
100	69	0	0	69	100	14	0
						0	14

DISCUSSION

Ahmad *et al.* have observed *Acinetobacter* spp. for 5 years. *A. baumannii* is the most common species isolated from the samples of the ICU, which consist of the respiratory secretions and wound swabs. The study has concluded that there is a high rate of multidrug resistance (MDR) to carbapenems, cephalosporins, and fluoroquinolones. Colistin and tigecycline are the most potent antibiotics, with the least ability to develop resistance. An increase in the trend of carbapenem-resistant *Acinetobacter* (CRAB) strains among the ICU patients has been noted. The results have revealed a strong association between long-term hospitalization and the use of invasive procedures. The study highlighted the need for surveillance and infection control, including antibiotic use, in tertiary care ^[9]. The study observed that *Acinetobacter* spp. causing blood infection is associated with ICU admission, invasive procedures, and a severe stage of illness. *A. baumannii* is the most common species with a high rate of multidrug resistance, for carbapenems, fluoroquinolones, and cephalosporins. The study has investigated the increase in the carbapenem-resistant strains, with tigecycline having important activity. Patients with carbapenem-resistant infection have shown a high rate of mortality.

The study has revealed a delay in the effective use of antibiotics as a common risk factor. The study has highlighted the need for continuous resilience to strengthen control strategies.

Raheja *et al.* reported that infections caused by *Acinetobacter baumannii* were identified among patients admitted to the ICU, in respiratory samples, wound swabs, and blood samples. Isolates have shown high multidrug resistance, including carbapenem, cephalosporin, and aminoglycoside resistance. Colisin is the most potent antibiotic with a low rate of resistance, but tigecycline has mild sensitivity. Infections were commonly observed among long-term hospitalized patients, mechanically ventilated patients, and those exposed to antibiotics. Major cases have led to a high morbid stage. Altogether, the study has revealed the rising concern for MDR *A. baumannii*. This has underscored the need for antibiotics and robust infection control practices ^[10].

Alibrahim *et al.* reported a long-term increase in the rate of *A. baumannii* among ICU and mechanically ventilated patients. Also, the isolates showed high multidrug resistance to carbapenems, cephalosporins, and fluoroquinolones, reducing their efficacy. The crucial drug was Colistin, but tigecycline showed moderate

activity. Infections have been associated with a longer duration of sleep, more invasive procedures, and the use of a broader spectrum of antibiotics. The study also highlighted the need for robust antimicrobial stewardship programs and regulations to address the growing challenge of MDR *A. baumannii* [11].

Sannathimappa *et al.* reported that *Acinetobacter baumannii* is the most common isolate from samples associated with respiration and wound secretions [12]. High levels of multidrug resistance, including carbapenems, cephalosporins, and fluoroquinolones, were associated with poor activity. Carbapenem-resistant strains were obtained from the isolates, limiting the CRAB. Colistin has the greatest potential for antibiotic resistance. The study also highlights the long hospital stay and the need for a control strategy. This also enhances antimicrobial stewardship to prevent the spread of MDR *Baumannii*.

CONCLUSIONS

The study concluded that the prevalence of *Acinetobacter* spp. in this sample was 2.67% and varied considerably across clinical specimens, with the highest detection in endotracheal aspirates and markedly lower rates in urine, cerebrospinal fluid, and pleural samples. The antimicrobial susceptibility analysis reveals extensive resistance to multiple first-line agents, particularly against cephalosporins and several β -lactams. At the same time, colistin and minocycline emerge as the most effective agents across isolates. These findings highlight both the heterogeneous distribution of *Acinetobacter* spp. in a tertiary care setting and the substantial therapeutic limitations posed by prevailing resistance patterns, reinforcing the need for evidence-guided antimicrobial stewardship.

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Materials— Priyakshree Saikia

Data collection— Priyakshree Saikia

Data analysis and interpretation— Priyakshree Saikia

Literature search— Priyakshree Saikia

Writing of the article— Priyakshree Saikia

Critical review— Keertana Shetty

Article editing— Keertana Shetty

Final approval— Keertana Shetty

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