

Isolation and Prevalence of Multi-Drug Resistant Acinetobacter from Various Clinical Samples in a Tertiary Care Hospital

Qursheed Sultana¹, R. Saraswati Jayanthi², K. Pavani³

^{1,2}Professor, ³Professor & HOD,
Department of Microbiology, RVM Institute of Medical Sciences and Research Centre, Mulugu, Telangana

Corresponding Author: Qursheed Sultana

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ABSTRACT

BACKGROUND: Acinetobacter species are emerging pathogens in healthcare settings, responsible for many infections, including bacteraemia, pneumonia, meningitis, peritonitis and wound infections. Their ability to develop multidrug resistance through various resistance determinants poses significant challenges for Treatment. Timely detection and reporting of these multidrug-resistant pathogens would be useful not only for guiding an appropriate antibiotic therapy but also in controlling the spread of these drug-resistant strains in the hospital.

OBJECTIVES:

- 1) To isolate and study the prevalence of Acinetobacter species from all clinical samples received in the microbiology lab from inpatient wards and ICUS OF a tertiary care hospital for a period of one year.
- 2) To determine the Antibiotic resistance pattern of Acinetobacter species.
- 3) To determine the prevalence of multidrug-resistant and carbapenem-resistant Acinetobacter species.

MATERIALS AND METHODS: A retrospective observational study was conducted in the microbiology department of RVMIMS&RC, MULUGU MANDAL of Siddipet district for a period of one year.

Samples were taken from patients admitted to different ICUs and wards of the hospital and processed according to the standard microbiological procedures. Identification and antibiotic susceptibility testing were done in a fully automated Vitek 2 system. All clinical samples and patients of all age groups were included, and duplicate isolates from the same patient were excluded from the study.

RESULTS: During this study, a total of 7855 samples were processed and 1171(14.90%) samples were culture positive, out of which 136 (11.61%) were Acinetobacter species, and Acinetobacter baumannii (92.6%) was the commonest of Acinetobacter species. Acinetobacter baumannii was the most prevalent organism in ICUS (58.08%), affecting elderly patients above 60 years (40.44%) and most commonly affecting Males (74.26%). Acinetobacter baumannii was most commonly isolated from pus samples (36.03%), and they were resistant to commonly used drugs. They were multidrug-resistant and carbapenem-resistant. Colistin was 100% sensitive, although it was not confirmed by the MBC method. Good sensitivity was seen to the combination drug, like Ampicillin-sulbactam (57.57%) and 54% to minocycline.

CONCLUSION: The present study showed the alarming trends of resistance of Acinetobacter strains isolated from clinical specimens to the various classes of antimicrobials. The improvement of microbiological techniques for quick and more accurate identification of bacteria is necessary for the selection of appropriate treatments.

Keywords: Acinetobacter, Multidrug resistant, Carbapenem resistant

INTRODUCTION

Acinetobacter species are small, Gram-negative, strictly aerobic, and non-fermenting bacteria that are oxidase-negative and non-motile. Commonly found in moist environments such as soil and water, these organisms are notable for their ability to survive on various surfaces, which enables their persistence in hospital settings^[1]. Acinetobacter species can act as opportunistic pathogens, causing a range of infections including those of the respiratory tract, skin, soft tissue, bloodstream, and urinary tract, especially in hospital environments and immunocompromised patients.

The most clinically significant member of this genus is *Acinetobacter baumannii*, followed by species such as *A. lwoffii*, *A. haemolyticus*, *A. junii*, and *A. ursingii*^[2]. Infections are often associated with various risk factors, including underlying diseases like diabetes, climatic conditions, and the use of invasive medical devices such as ventilators and catheters. The rise of antibiotic resistance among Acinetobacter species, particularly multidrug resistance, is a major concern in healthcare institutions, with resistance rates varying across settings and regions. Prolonged ICU stays and frequent exposure to antibiotics contribute significantly to this problem. Most infections result from nosocomial spread and colonization rather than new infections.

These organisms can be isolated from a variety of clinical sources, such as sputum, pus, wound swabs, tissue, endotracheal secretions, bronchoalveolar lavage, blood, urine, and other body fluids. Acinetobacter is recognised as one of the six key pathogens in the “ESKAPE” group, making it an increasingly important and difficult-to-treat. Acinetobacter species exhibit resistance to

nearly all commonly prescribed antibiotics, including aminoglycosides, fluoroquinolones, broad-spectrum β -lactams, and cephalosporins. While carbapenems were once considered the most effective treatment option, resistance to these drugs has risen significantly in recent years, leading to the emergence of multidrug-resistant (MDR), extensively drug-resistant (XDR), and even pan-drug-resistant (PDR) strains,^[3].

A growing concern is the increasing global resistance to colistin, a last-resort antibiotic, with resistance rates approaching 4% internationally. This trend further limits available therapeutic choices for Acinetobacter infections. The resistance mechanisms in these organisms involve multiple pathways, such as β -lactamase production, target site alteration, efflux pump overexpression, reduced membrane permeability, and biofilm formation, all of which contribute to the pathogen's persistence and adaptability in healthcare environments.

Antibiotic susceptibility patterns among Acinetobacter species vary considerably across geographical regions and even between different hospital units, making continuous local surveillance essential. Regular monitoring helps determine appropriate antibiotic therapies and supports effective infection control measures.

Acinetobacter infections have been linked to high mortality rates, reaching up to 70% in critically ill patients. This study, therefore, aims to achieve the following objectives:

- 1) To isolate and evaluate the prevalence of Acinetobacter infections from various clinical samples.
- 2) To determine the antibiotic susceptibility profiles of the isolates within the hospital setting.

- 3) To estimate the proportion of carbapenem-resistant and multidrug-resistant *Acinetobacter* species, thereby identifying the most effective treatment strategies.

MATERIALS AND METHODS

This study was designed as a retrospective cross-sectional analysis conducted in the Department of Microbiology at RVM IMS & RC, Mulugu mandal, Siddipet district, Telangana, spanning July 2024 to July 2025. Clinical specimens—including pus, wound swabs, tissue, endotracheal secretions, sputum, bronchoalveolar lavage, blood, urine, and body fluids (pleural, peritoneal, and cerebrospinal)—were collected from inpatients across ICUs, postoperative, and general wards. The primary objective was to isolate *Acinetobacter* species and determine their antibiotic susceptibility profiles.

Inclusion criteria encompassed all clinical samples from patients of any age group, provided that *Acinetobacter* species were identified, while duplicate isolates and samples from the same patient were excluded. Ethical clearance for the study was obtained from the college ethics committee, and relevant patient information (age, sex, admitting ward, identification of organism, and susceptibility pattern) was documented from departmental records.

Aseptic sterile containers were used for sample collection and sent immediately for laboratory processing. All samples were subjected to Gram staining and also inoculated on blood agar, MacConkey agar, CLED agar (for urine) and incubated at 37°C for 24-48 hours. Isolates were identified based on colony morphology, non-lactose fermentation, pigmentation, hemolysis, and a battery of biochemical tests (catalase, oxidase, motility, Indole, citrate, urease, TSI, and glucose utilization). Suspected *Acinetobacter* isolates were further processed and identified using the BIOMERIEU VITEK 2 system.

Antibiotic susceptibility testing was performed using the VITEK-2 automated system and followed CLSI,^[4] (2025)

guidelines for interpretation. An N406 panel of antibiotics were used, like ceftazidime, cefepime, ciprofloxacin, levofloxacin, gentamicin, tobramycin, amikacin, imipenem, meropenem, piperacillin-tazobactam, cotrimoxazole, minocycline, colistin, and cefoperazone-sulbactam. Results for multidrug resistance (MDR) and carbapenem resistance (CR) were documented in microbiology records, and treatment recommendations were made to clinical teams based on these findings.

RESULTS

During the one-year study period, a total of 7,855 clinical samples were processed in the Department of Microbiology. Out of these, 1,171 samples (14.9%) were culture positive, and 136 isolates (11.61%) were identified as *Acinetobacter* species.

Overall, 136 non-duplicate isolates of *Acinetobacter* were obtained from various clinical specimens. Within the spectrum of culture-positive Gram-negative bacteria, *Acinetobacter* ranked as the fourth most common isolate, following *Escherichia coli*, *Klebsiella* species, and *Pseudomonas* species. This distribution pattern aligns with findings from previous studies, where pus and respiratory samples were consistently reported as the predominant sources of *Acinetobacter* isolates.

Table 1: Sample-wise distribution of *Acinetobacter* isolates

Sample	%	Number
Pus	36.03	49
Et secretions	30.15	41
Sputum	7.35	10
BAL	5.88	8
Urine	10.30	14
Blood	5.88	8
Body fluids	4.41	6

Acinetobacter species were most frequently isolated from pus samples (49 isolates, 36.03%), followed by respiratory specimens such as endotracheal secretions, bronchoalveolar lavage, and sputum, which together accounted for 59 isolates (43.38%). Additionally, isolates were recovered from

urine (14 isolates, 10.30%), blood (8 isolates, 5.88%), and body fluids, including pleural and peritoneal fluids (6 isolates, 4.41%).

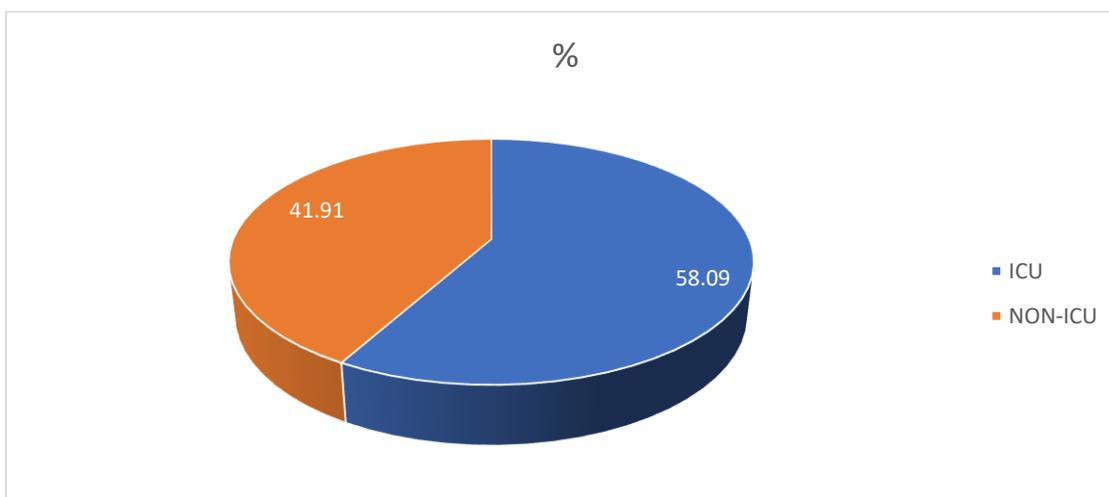


Fig.1. ICU/NON-ICU Distribution of Acinetobacter isolates

Table 2: ICU/Ward-wise distribution of Acinetobacter isolates

ICU/WARD	NO. OF CASES	PERCENTAGE
RICU	62	45.58%
MICU	17	12.5%
ORTHO	22	16.17%
SURGERY	16	11.76%
RESPIRATORY	6	4.45%
MEDICAL	5	3.67%
ONCOLOGY	3	2.20%
CTVS	2	1.47%
UROLOGY, ENT, GYN	3	2.20%
		100%

Out of the total 136 Acinetobacter isolates obtained, 79 (58.08%) were recovered from various Intensive Care Units (ICUs), with the Respiratory Intensive Care Unit (RICU) being the predominant source, contributing 62 isolates (45.58%). The Medical Intensive Care Unit (MICU) accounted for 17 isolates (12.5%).

The remaining 41.91% (57 isolates) originated from different hospital wards. Among these, post-operative orthopaedic wards reported 22 isolates (16.17%), post-operative surgical wards 16 (11.76%),

respiratory wards 6, medical wards 5, oncology 3, cardiothoracic and vascular surgery (CTVS) 2, and departments such as ENT, gynaecology, and urology contributed one isolate each.

This distribution emphasises the predominance of Acinetobacter infections in ICU settings, particularly in respiratory units, due to factors such as mechanical ventilation, invasive procedures, and prolonged hospitalisation, which are known to facilitate colonisation and infection by Acinetobacter species.

Table 3: Speciation of Acinetobacter isolates

Species	Number	Percentage
Acinetobacter baumannii	126	92.64%
Acinetobacter Iwoffii	6	4.41%
Acinetobacter haemolyticus	2	1.47%
Acinetobacter ursingii	1	0.74%
Acinetobacter junii	1	0.74%

Out of the 136 *Acinetobacter* isolates obtained, 126 (92.6%) were identified as belonging to the *Acinetobacter baumannii* complex. The remaining isolates included *Acinetobacter lwoffii* (6 isolates, 4.41%), *Acinetobacter hemolyticus* (2 isolates, 1.47%), and single isolates of *Acinetobacter ursingii* and *Acinetobacter junii* (1 each, 0.74%).

This distribution aligns with global and regional findings indicating that *A. baumannii* remains the predominant clinical isolate, typically representing over 90% of *Acinetobacter* infections in hospital and ICU

settings. Its high prevalence is attributed to its ability to survive on dry surfaces, form biofilms on medical devices, and develop resistance to multiple antibiotic classes.

Non-*baumannii* species such as *A. lwoffii*, *A. hemolyticus*, *A. junii*, and *A. ursingii* are less common and are usually associated with community-acquired or opportunistic infections in immunocompromised patients. However, even these species have shown emerging multidrug resistance in some healthcare environments, making continuous monitoring essential.

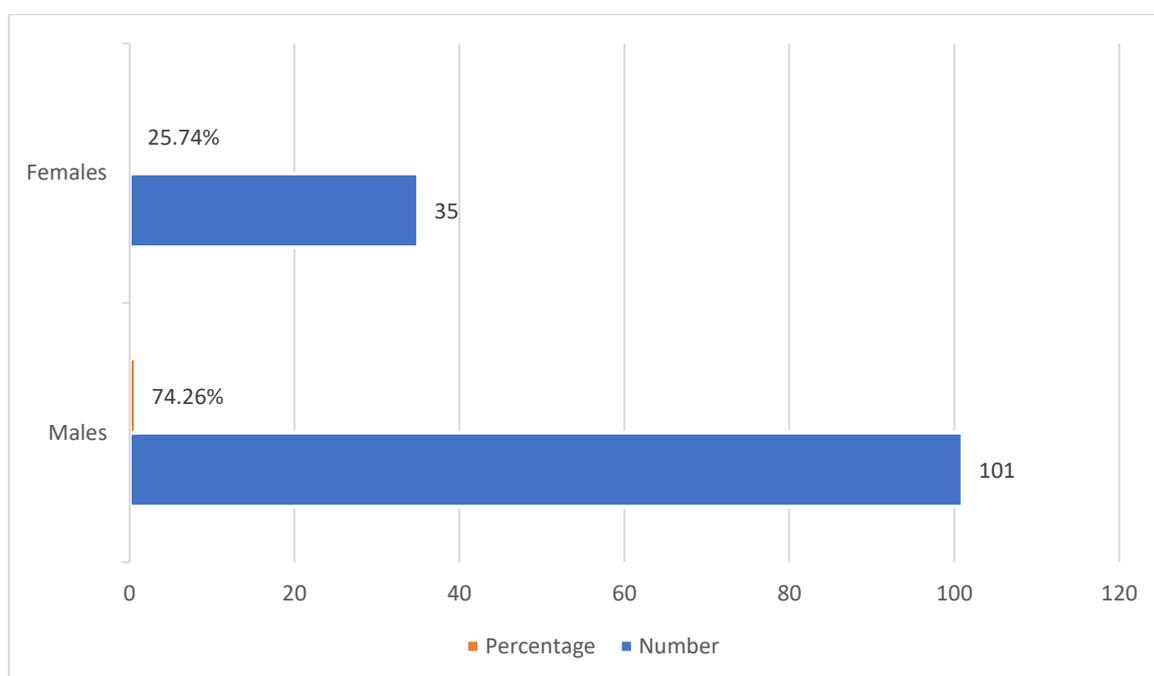


Fig.2: Gender wise distribution of *Acinetobacter* isolates

Table 4: Age-wise distribution of *Acinetobacter* species

Age	Males	Females	Total
<30yrs	13	3	16
31-40yrs	12	3	15
41-50yrs	11	9	20
51-60yrs	24	6	30
>60yrs	41	14	55
Total	101(74.26%)	35(25.74%)	136(100%)

The majority of *Acinetobacter* infections in this study occurred among patients aged above 60 years (55 cases), followed by those aged 51–60 years (30 cases), 41–50 years (20 cases), and below 30 years (16 cases). The lowest number of cases was observed in the 31–40 years age group (15 cases). This age

distribution indicates that older adults were significantly more vulnerable to *Acinetobacter* infections, likely due to compromised immunity, multiple comorbidities, and prolonged hospital stays. Recent studies support this trend, showing that *Acinetobacter* infections, particularly

multidrug-resistant (MDR) strains, are most prevalent in elderly populations, especially those aged 60 years and above. These studies have demonstrated that age correlates positively with both infection rates and mortality, as advanced age is often accompanied by weakened immune defence and frequent exposure to invasive procedures and antibiotics.

In contrast, younger patients exhibited comparatively lower infection rates, suggesting that host-related factors such as immunity, underlying chronic illnesses (e.g., diabetes, hypertension), and duration of hospitalisation are pivotal in determining susceptibility to *Acinetobacter* infections.

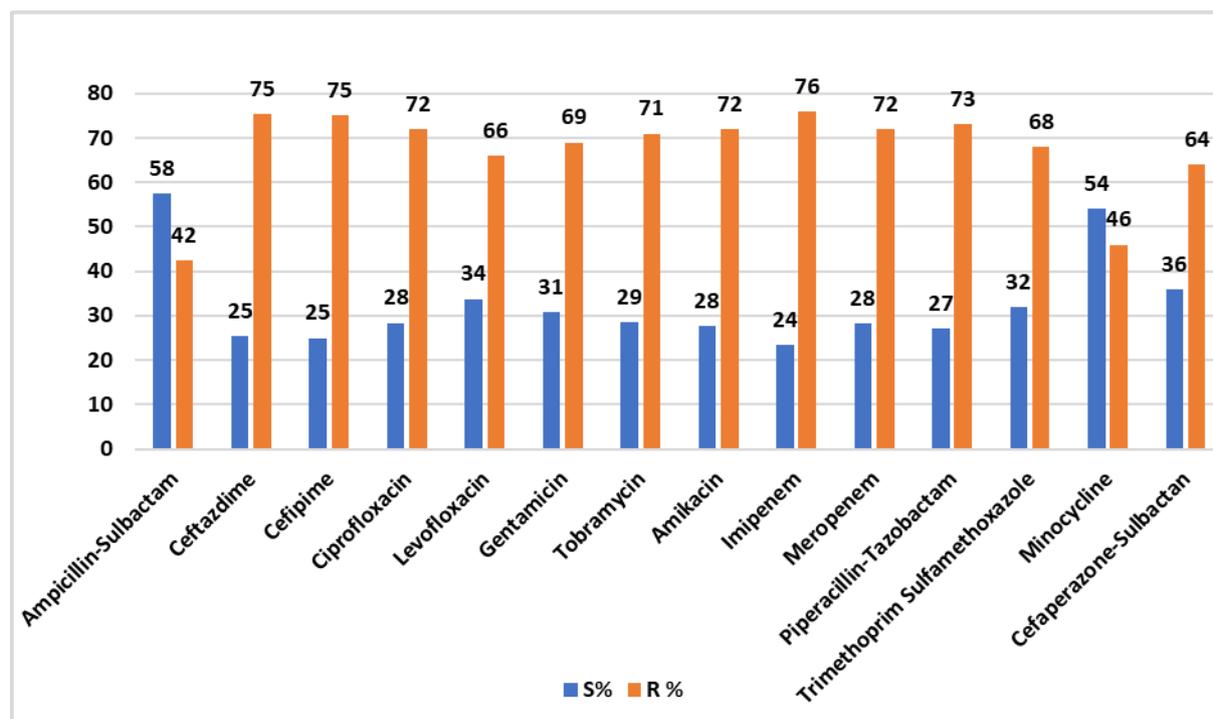


Fig.3: sensitivity and resistance pattern of antibiotics.

Antibiotic resistance profiling of *Acinetobacter* isolates in this study showed substantial resistance to widely used antibiotics. The resistance rates for cephalosporins—specifically ceftazidime and cefepime—were 75%. Aminoglycosides also exhibited high resistance levels: 71% to tobramycin, 72% to amikacin, and 69% to gentamicin. Fluoroquinolones such as ciprofloxacin and levofloxacin had resistance rates of 72% and 66%, respectively. Piperacillin-tazobactam (a β -lactam/ β -lactamase inhibitor combination) showed a 73% resistance rate, whereas cefoperazone-sulbactam and ampicillin-sulbactam demonstrated better efficacy, with 36% and 58% sensitivity, respectively. Cotrimoxazole had a sensitivity of 32%. The highest sensitivity rates were observed for

minocycline (54%) and ampicillin-sulbactam (58%), while carbapenems like imipenem and meropenem showed only 24% and 28% sensitivity, respectively. Notably, all isolates were sensitive to colistin.

Out of the 136 isolates, 89 (65.44%) demonstrated multidrug resistance (resistant to at least three groups of antibiotics). Carbapenem resistance was particularly high, with 95 isolates (69.85%) being resistant to both imipenem and meropenem. Carbapenem-resistant *Acinetobacter* species were detected mainly in ICUs, accounting for 61 isolates (64.21%) compared to 34 isolates (35.78%) from other wards.

These findings emphasise the pressing need for consistent antimicrobial stewardship, infection control interventions, and periodic local surveillance to guide effective

therapeutic strategies, especially given the limited treatment options and the high prevalence of multidrug and carbapenem resistance among hospital isolates.

DISCUSSION

Acinetobacter infections have become a major challenge in modern healthcare, particularly in intensive care units (ICUs). Among the various species, *Acinetobacter baumannii* stands out as one of the most difficult pathogens to manage due to its ability to acquire multidrug resistance (MDR) and survive on hospital surfaces under both dry and moist conditions.

The present study aimed to isolate *Acinetobacter* species and analyse their antibiotic susceptibility profiles from multiple clinical samples. Here, 136 isolates (11.61%) of *Acinetobacter* were identified from 1,171 culture-positive samples. This prevalence was comparable to findings by Rajkumari et al,^[5](2020), in Nepal (11.49%), while Madhavi et al,^[6](2022), reported a lower rate of 8.9%. Slightly higher prevalence values (16.2%) were observed in studies by Swathi Akula et al. 2017, likely due to methodological and population differences.

In the current study, 58.09% of isolates originated from ICUs, which is similar to Sana et al,^[7](2020) and contrasting with Wankhade et al,^[8](2016) study that reported an ICU isolation rate of 77%. The high ICU prevalence in this study might be attributed to prolonged hospital stays, invasive interventions (e.g., ventilation, catheterization), and compromised immunity among critically ill patients. Similar trends were seen in tertiary care hospitals, where referred patients often carry drug-resistant strains.

Gender-wise, male patients (74.26%) were more frequently affected than females (25.74%), in agreement with observation from Ahmed et al,^[9](2023). However, opposite trends showing female predominance have been reported in Rabic V et al,^[10](2018) study, possibly linked to higher healthcare access among women. Age

distribution analysis revealed that individuals over 60 years had the highest infection rates (40.44%), consistent with reports from Asghar et al,^[11](2024) study, which showed 40.59% in patients above 60 years. Comorbidities, such as diabetes and hypertension, likely increase susceptibility in elderly populations.

Regarding sample distribution, pus (36.03%) and endotracheal secretions (30.15%) were the most common sources, a pattern similar to findings from Asghar et al. 2024. In contrast to our study, Guddetti et al. ^[12](2023) found most isolates in Endotracheal aspirates(50%), Rajkumari et al 2020 found most isolates in sputum(31.88%), and Gupta et al,^[13](2015) reported most isolates in blood(36.9%). Lower detection rates in urine (10.30%), sputum (7.35%), BAL (5.88%), blood (5.88%), and other fluids (4.41%), in contrast with Tewari R et al,^[14](2018), who reported urine as the predominant source of isolates. The variation in the prevalence of *Acinetobacter* species can be attributed to differences in geographical distribution and antibiotic policies adopted by different institutions.

Acinetobacter baumannii was the predominant species, accounting for 92.64% of all isolates, followed by *A. lwoffii* (4.41%). These findings closely reflect studies by Pragya Rani et al,^[15](2015), where *Acinetobacter baumannii* represented 93.16% of isolates. Osman et al, ^[16](2023) isolated *Acinetobacter baumannii* with less percentage of about 87.5%. The persistence of *Acinetobacter baumannii* in hospitals is likely due to its notable resistance against desiccation, disinfectants, and major antimicrobial drugs.

The antibiotic resistance profile from this study revealed a high resistance rate to cephalosporins (ceftazidime & cefepime) of 75%, consistent with Sannathimmappa's study ^[17], and lower than the koul et al,^[18](2022), where it was 100%. In our study, resistance to aminoglycosides (69–72%) and fluoroquinolones (66–72%), which is less than Asghar et al study, where it was 98.36% and 100%.

Piperacillin-tazobactam showed 72.09% resistance consistent with sannathimmappa study(72%) and less than Asghar et al study(98.36%)

Carbapenem resistance rates (76.4% for imipenem; 71.77% for meropenem) were in consistent with Yadav et al,^[19](2023) and comparatively lower than those reported by Asghar et al. (2025) and Sohail et al,^[20](2016), who recorded resistance above 90%. The relatively lower carbapenem resistance in this study likely reflects judicious antibiotic use and stringent infection-control measures.

Notably, colistin showed a 100% sensitivity rate, consistent with recent global data confirming it as the most effective remaining drug against MDR and XDR *Acinetobacter baumannii* strains. However, emerging reports of colistin resistance worldwide underscore the need for continuous surveillance and antibiotic stewardship.

In conclusion, the study highlights a persistently high prevalence of MDR and carbapenem-resistant *Acinetobacter* infections, especially in ICUs. The findings warrant stricter infection control, rational antibiotic prescription, and continuous surveillance to curb the spread of resistant strains and preserve the effectiveness of last-line drugs.

CONCLUSION

A total of 136 *Acinetobacter* isolates were obtained from 1171 culture positive clinical samples, yielding an overall prevalence rate of 11.61% across the hospital. Among inpatients, the majority of isolates were recovered from ICU patients, accounting for 58.09% while the remaining isolates were mainly from ward patients.

Species identification revealed that *Acinetobacter baumannii* was the predominant strain, comprising 126 isolates (92.64%). The less frequently isolated species included *Acinetobacter lwoffii* (6 isolates, 4.41%), *Acinetobacter hemolyticus* (2 isolates, 1.47%), and *Acinetobacter junii* and *ursingii* (1 isolate each, 0.74%). This species distribution reflects the strong

dominance of *A. baumannii* in hospital-acquired infections, consistent with international studies and current epidemiological trends in critical care settings. In our study, 65.44% of the isolates were multidrug resistant and 69.85% were carbapenem resistant, which is less than the Vadodara, Gujarat,^[21] (2024) study and Asghar et al, 2025 study. In our study, Ampicillin-sulbactam showed good sensitivity of 57.57% and emerged as the preferred therapeutic option for serious carbapenem-resistant infections, followed by Minocycline 54%, cotrimoxazole, levofloxacin, gentamicin, cefoperazone-sulbactam and Tobramycin showed around 30% sensitivity each. Colistin showed 100% sensitivity in the Vitek 2 system.

Our study lacks molecular detection methods which would be beneficial for timely detection of these multidrug-resistant organisms by molecular detection methods for effective antibiotic therapy.

The high resistance to commonly used antibiotics highlights the need for antibiotic testing with new antibiotics like Ervacycline and combination drugs like ceftazidime-avibactam, sulbactam-durlobactam, Aztreonam, are urgently needed for effective treatment and to prevent multidrug resistance.

Declaration by Authors

Ethical Approval: Approved

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