



ANTIMICROBIAL RESISTANCE AND MOLECULAR CHARACTERIZATION OF ACINETOBACTER BAUMANNII ISOLATES FROM HOSPITAL ACQUIRED INFECTION IN INTENSIVE CARE UNIT

Hemant Kumar^{1*}, Dr. Harshada Shah², Neha Rani³

¹PhD Scholar, Department of Microbiology, Index Medical College, Hospital & Research Centre, Indore, (M.P) India (Malwanchal University) Email – hk824500@gmail.com

²Research supervisor, Department of Microbiology, Index Medical College, Hospital & Research Centre, Indore (M.P)India. (Malwanchal University) sharshada1955@yahoo.com

³PhD Scholar, Department of Microbiology, Index Medical College, Hospital & Research Centre, Indore, India. (Malwanchal University) Email- neharani.7931@gmail.com

***Corresponding author:** Hemant Kumar

*PhD Scholar, Department of Microbiology, Index Medical College, Hospital & Research Centre, Indore, (M.P) India (Malwanchal University) Email – hk824500@gmail.com

Abstract

Background: In intensive care units (ICUs), hospital-acquired infections (HAIs) are a major contributor to morbidity and mortality. Because *A. baumannii* can thrive in a hospital setting, form biofilms, and quickly develop multidrug and carbapenem resistance, which limits treatment options, it has become a prominent ICU infection.

Aims: The present study aimed to determine the prevalence, antimicrobial resistance pattern, and molecular characteristics of *Acinetobacter baumannii* isolates recovered from hospital-acquired infections in ICU patients.

Materials and Methods: A cross-sectional observational study was carried out in the Department of Microbiology at the Index Medical College Hospital and Research Center in Indore (M.P). Standard microbiological techniques were used to process 360 clinical samples from adult ICU patients with suspected HAIs (≥ 48 hours of admission). PCR targeting the blaOXA-51 gene and phenotypic testing were used to validate the identification of *A.baumannii*. According to CLSI recommendations, the Kirby-Bauer disk diffusion method was used for antimicrobial susceptibility testing, and MIC determination was used to confirm carbapenem resistance. Assays for biofilm formation and molecular detection of the carbapenemase genes blaIMP, blaVIM, and blaNDM were carried out.

Results: Out of 360 ICU samples analyzed, 126 (35.0%) were positive for *Acinetobacter baumannii*, with respiratory and wound-related specimens being the most common sources of isolation. Infections were more frequent in male patients and predominantly affected individuals aged 41–60 years. Mechanical ventilation was the major risk factor, followed by urinary catheterization and prolonged ICU stay.

Conclusion: Multidrug-resistant *A.baumannii* is highly prevalent in ICU-associated HAIs, which emphasizes the critical need for ongoing antimicrobial resistance surveillance, molecular monitoring, and strict infection control measures to stop its spread and enhance clinical outcomes.

Introduction

A significant worldwide health concern, hospital-acquired infections (HAIs) are most common in intensive care units (ICUs), where patients are immunocompromised, severely ill, and frequently exposed to invasive procedures and long-term antimicrobial medication.¹ Because of its remarkable capacity to endure in the hospital setting and quickly develop antibiotic resistance, *A. baumannii* has become a prominent pathogen among the organisms responsible for ICU-associated HAIs.²

A variety of diseases, such as ventilator-associated pneumonia, bloodstream infections, urinary tract infections, and surgical site infections, are caused by the aerobic, non-fermenting, Gram-negative coccobacillus *A. baumannii*.³

Its ability to build biofilms on medical equipment, last on dry surfaces for extended periods of time, and withstand standard disinfectants makes it easier for it to spread within intensive care units (ICUs) and increases the frequency of nosocomial outbreaks.⁴

Treatment options have been severely limited due to the rising frequency of *A. baumannii* that is both carbapenem-resistant and multidrug-resistant (MDR). In the past, carbapenems were thought to be the best treatment for serious infections brought on by this organism; nevertheless, resistance has quickly increased globally.⁵ Along with additional mechanisms like efflux pumps, decreased outer membrane permeability, and changes in penicillin-binding proteins, the formation of carbapenemases, such as metallo- β -lactamases and class D oxacillinases, is the main cause of carbapenem resistance in *A. baumannii*.⁶ These resistance determinants can spread quickly in hospital settings because they are frequently carried on mobile genetic elements.⁷

Understanding resistance mechanisms, monitoring the development of epidemic clones, and directing infection control tactics all depend on the molecular characterization of *A. baumannii* isolates.⁸ In ICU settings, thorough surveillance that combines molecular analysis and antibiotic susceptibility testing is therefore essential. The goal of this study is to assess the molecular traits and antibiotic resistance patterns of *A. baumannii* isolates that cause hospital-acquired infections in the intensive care unit.

Materials and Methods

A cross-sectional observational study was conducted in the Department of Microbiology, Index Medical College Hospital and Research Centre, Indore, involving adult ICU patients with suspected hospital-acquired infections developing after 48 hours of admission. The study was carried out over 18 months, including a data analysis period, after obtaining ethical approval from the Institutional Ethics Committee.

Clinical specimens received for culture and sensitivity testing were processed for the isolation and identification of *A. baumannii*. Patients with ICU stay of less than 48 hours and pediatric or neonatal patients were excluded. The sample size was calculated assuming an 18% prevalence of *A. baumannii* infections, resulting in an estimated requirement of approximately 340 isolates.

Specimens such as respiratory samples, catheterized urine, blood, pus, and wound swabs were collected aseptically and cultured using standard microbiological techniques. Identification was based on Gram staining, colony morphology, biochemical tests, and molecular confirmation by PCR targeting the blaOXA-51 gene.

Antimicrobial susceptibility testing was performed by the Kirby–Bauer disk diffusion method and interpreted according to CLSI guidelines, with carbapenem resistance confirmed by MIC determination. Molecular characterization included real-time PCR for blaOXA-51 and multiplex PCR for detection of blaIMP, blaVIM, and blaNDM genes. Biofilm production was assessed using tube and microtiter plate assays. Data were analyzed using SPSS and Excel, and a p-value <0.05 was considered statistically significant.

Result –

Out of 360 ICU samples analyzed, 126 (35.0%) were positive for *Acinetobacter baumannii*, while 234 (65.0%) were negative. This indicates that more than one-third of ICU patients were affected, highlighting the significant burden of *A. baumannii* infections in the intensive care setting.

Table 1 - Overall Prevalence of *A.baumannii* Among ICU Samples

Total samples	Positive (35%)	Negative
360	126 (35.0%)	234 (65.0%)

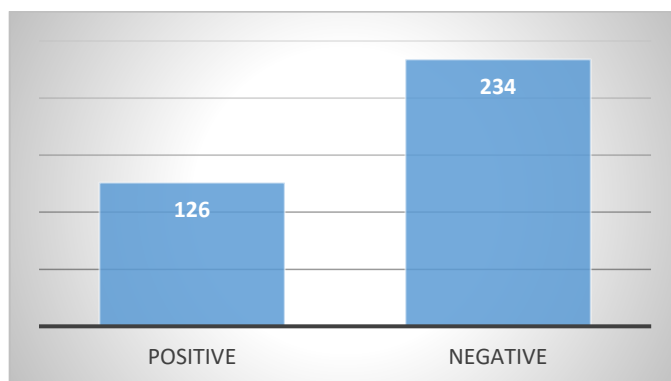


Figure 1- Overall Prevalence of *A.baumannii* among ICU Samples

A total of 360 clinical samples from ICU patients were processed for the isolation of *A. baumannii*, of which 126 (35.0%) were positive and 234 (65.0%) were negative. The highest isolation rate was observed in respiratory samples, with 42 of 108 specimens (38.9%) yielding *A.baumannii*. This was followed by pus, wound swabs, and surgical site samples, where 30 of 72 samples (41.7%) were positive. Urine samples showed 20 positives out of 72 (27.8%), while blood cultures were positive in 16 of 54 cases (29.6%). Among body fluids such as pleural fluid, cerebrospinal fluid, and drain fluids, 10 of 36 samples (27.8%) were positive. In the remaining category, including catheter tips, bronchoalveolar lavage, and miscellaneous swabs, 8 of 18 samples (44.4%) yielded *A.baumannii*. Overall, respiratory and wound-related specimens constituted the major sources of *A.baumannii* isolation in ICU patients.

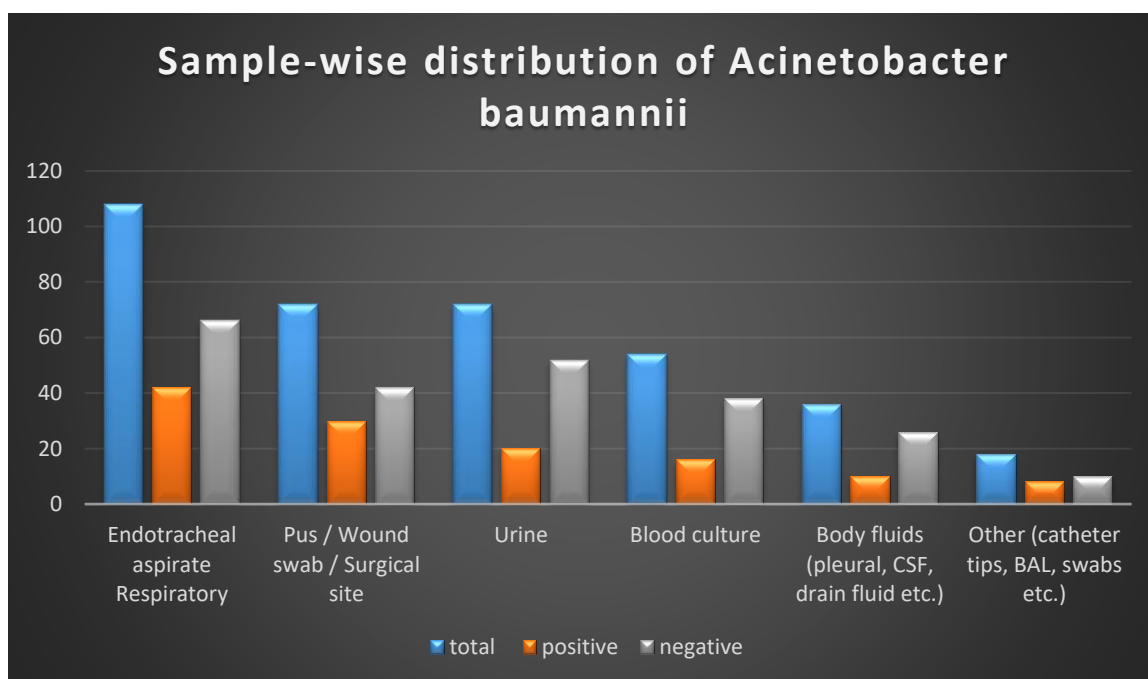


Figure 2-Sample-wise distribution of *Acinetobacter baumannii*

Of the 126 samples positive for *Acinetobacter baumannii*, 76 (60.3%) were obtained from male patients and 50 (39.7%) from female patients, indicating a higher prevalence among males. Among

the 234 culture-negative samples, 124 were from males and 110 from females, reflecting a comparable gender distribution in the overall ICU population.

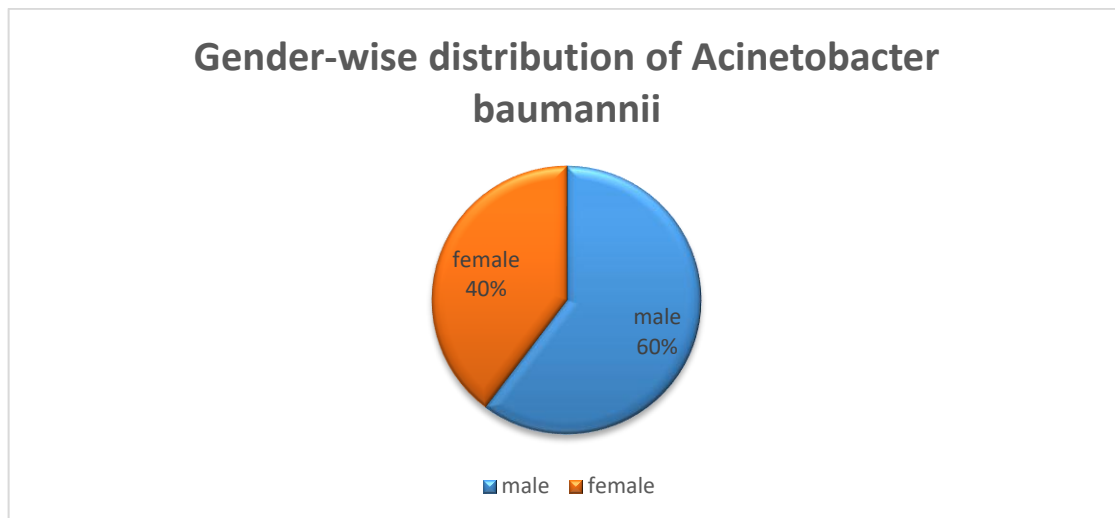


Figure 3 -Gender-wise distribution of *A.baumannii*

Out of 360 ICU patients, *Acinetobacter baumannii* was isolated in 126 cases. The highest proportion of infections was observed in patients aged 41–60 years (43.7%), followed by those older than 60 years (34.0%). Patients aged 21–40 years accounted for 17.5% of cases, while those under 20 years comprised only 4.8%. The remaining 234 patients had negative culture results.

Table- 2 Age-wise distribution of *A.baumannii*

Age group (years)	Positive isolates (n = 126)	Percentage (%)	Negative isolates (from total 360)
<20	6	4.8%	24
21–40	22	17.5%	58
41–60	55	43.7%	71
>60	43	34.0%	81
Total	126	100%	234

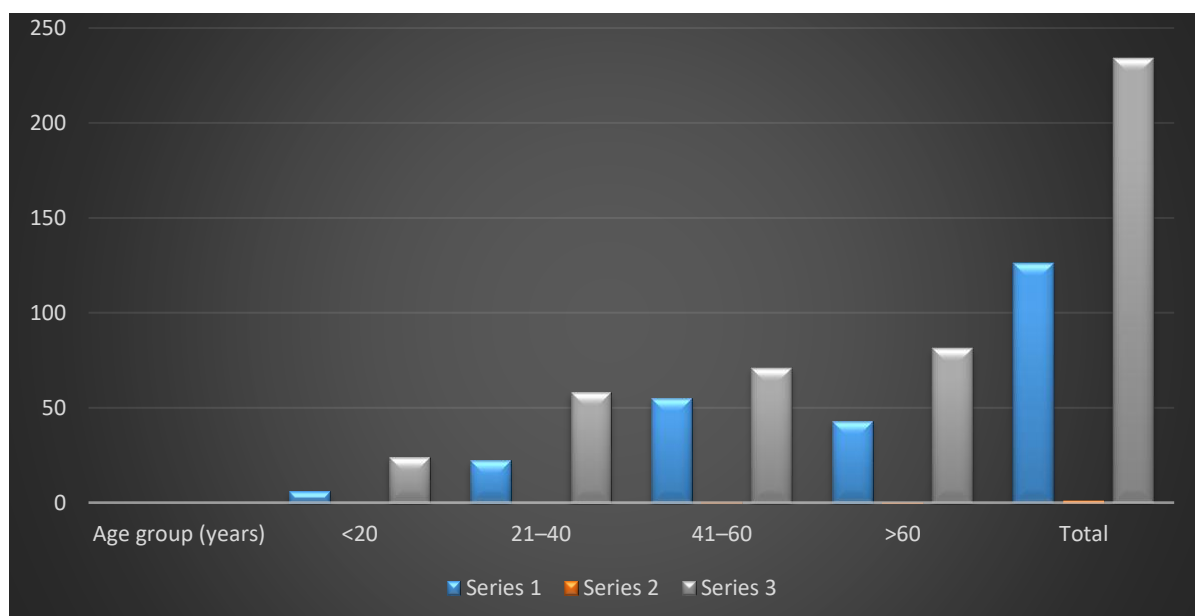


Figure 4- Age-wise distribution of *A.baumannii*

Among the 126 patients with *Acinetobacter baumannii* infection, mechanical ventilation or endotracheal intubation was the most common risk factor, present in 50 patients (39.7%). Urinary catheterization was observed in 32 patients (25.4%), while 25 patients (19.8%) had a prolonged ICU stay of more than seven days. Recent surgery or wound exposure was noted in 13 patients (10.3%), and comorbid conditions were present in 6 patients (4.8%).

Table- 3 Risk Factor Distribution in *Acinetobacter baumannii* Positive Patients

Risk Factor	Number of patients	Percentage (%)
Mechanical ventilation / ET tube	50	39.7%
Urinary catheter	32	25.4%
Prolonged ICU stay (>7 days)	25	19.8%
Recent surgery / wounds	13	10.3%
Comorbidities	6	4.8%
Total	126	100%

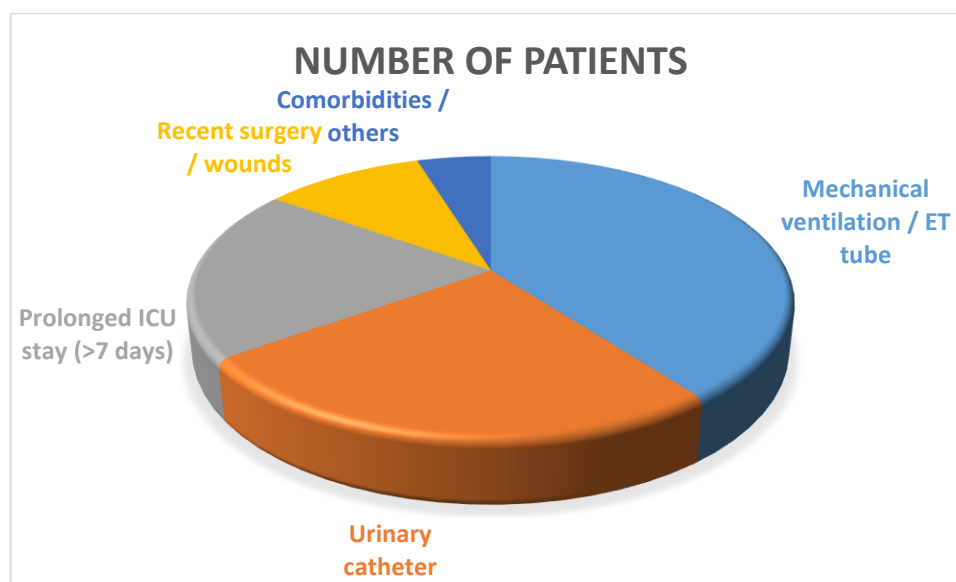


Figure 5-Risk Factor Distribution in *Acinetobacter baumannii* Positive Patients

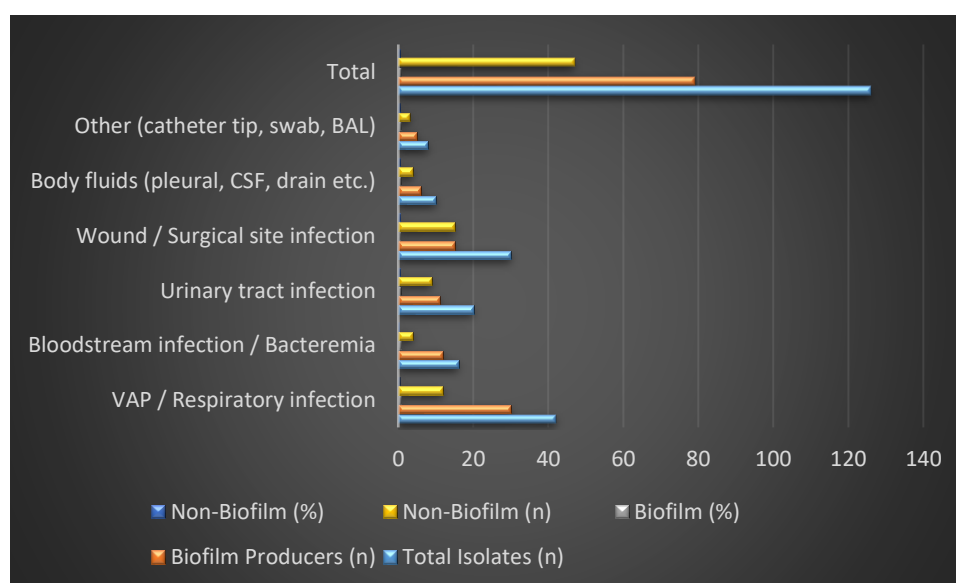


Figure 6- Biofilm Production in Different Clinical Infections

Out of 126 *Acinetobacter baumannii* isolates, 79 (63%) were biofilm producers, while 47 (37%) did not produce biofilms. Biofilm production varied by the source of infection:

- Bloodstream infections: 12/16 isolates (75%)
- Respiratory infections (including ventilator-associated pneumonia): 30/42 isolates (71%)
- Catheter tips, swabs, and BAL samples: 5/8 isolates (62.5%)
- Body fluids (pleural fluid, CSF, drain samples): 6/10 isolates (60%)
- Urinary tract infections: 11/20 isolates (55%)
- Wounds and surgical sites: 15/30 isolates (50%)

Conclusion

Acinetobacter baumannii showed a high prevalence in ICU-acquired infections, especially among ventilated patients. Respiratory samples were the most common source of isolation. Strengthened infection control and antimicrobial stewardship are essential to reduce its burden in ICUs.

DISCUSSION -

Acinetobacter baumannii was recovered from 126 out of 360 ICU samples in this investigation, resulting in a 35% prevalence rate. The increasing significance of *A. baumannii* as a predominant ICU infection is highlighted by this high prevalence. Sarkar et al. (2014)⁹ in Eastern India and Garnacho-Montero et al. (2015)¹⁰ in European intensive care units observed similar prevalence rates, with isolation rates ranging from 30% to 40%. Agarwal et al. (2012)¹¹ and Rao et al. (2016),¹² on the other hand, observed lower prevalence rates, ranging from 15% to 25%. These discrepancies could be explained by disparities in infection control procedures, antimicrobial stewardship initiatives, ICU infrastructure, and patient populations.

In the current investigation, respiratory samples were the most frequent source of *A. baumannii* isolation, followed by pus and wound swabs. The substantial correlation between *A. baumannii* and ventilator-associated respiratory infections is demonstrated by the approximately 39% of respiratory samples that tested positive. *A. baumannii* was found to be a major cause of ventilator-associated pneumonia (VAP) by Peleg et al. (2008)¹³ and Chastre and Fagon (2002)¹⁴. The organism's endurance and spread in intensive care units are greatly increased by its capacity to stick to endotracheal tubes and create biofilms on ventilator circuits. More than one-third of all *A. baumannii* isolates were found in respiratory specimens, according to Singh et al. (2020).¹⁵

The second most frequent source in the current investigation was wound-related samples, such as pus and surgical site specimens. This result is consistent with findings by Howard et al. (2012)¹⁶ and Manchanda et al. (2010)¹⁷ who noted significant rates of *A. baumannii* isolation from surgical wounds, particularly in trauma and postoperative intensive care unit patients. Wound contamination and nosocomial transmission are made easier by the organism's resistance to standard disinfectants and capacity to live on dry surfaces.

A. baumannii infections were more common in male patients (60.3%) than in female patients (39.7%), according to the gender distribution. Mohanty et al. (2021),¹⁸ Kumar et al. (2017)¹⁹ have all reported similar male predominance. Males are more likely to be admitted to the intensive care unit (ICU), are more likely to undergo invasive procedures, and are more likely to have comorbid diseases including diabetes and chronic lung disease. However, some research, such as Falagas et al. (2006),²⁰ found no discernible gender difference, indicating that gender might not be a risk factor in and of itself.

According to the current study's age-wise analysis, individuals between the ages of 41 and 60 accounted for 43.7% of *A. baumannii* infections, followed by those over 60 (34%). These results are in line with reports by Falagas et al. (2016)²⁰ and Dash et al. (2018),²¹ who found that middle-aged and older ICU patients were more susceptible. Patients who are older are more likely to develop nosocomial infections due to decreased immunity, numerous comorbidities, extended hospital stays, and higher exposure to intrusive devices. Patients under the age of 20 may have a comparatively reduced prevalence due to either lower rates of invasive interventions or exclusion from lengthy ICU exposure.

According to the current study's risk factor analysis, mechanical ventilation—which was present in 39.7% of patients who tested positive for *A.baumannii*—was the most important predisposing factor. This result is consistent with research by Vincent et al. (2009)²² and Kollef et al. (2012)²³ that found mechanical ventilation to be a significant independent risk factor for *A.baumannii* infections acquired in intensive care units. Bacterial adhesion and biofilm formation are facilitated by endotracheal tubes, which shield the organism from antimicrobial agents and host defences.

The second most frequent risk factor was urinary catheterization (25.4%), which was followed by an ICU stay longer than seven days (19.8%). Similar correlations have been documented by Bergogne-Bézín and Towner (1996) and Garcia-Garmendia et al. (2001),²⁴ who highlighted the significance of invasive equipment and extended hospital stays in promoting *A.baumannii* colonization and infection. Multidrug-resistant strains are more likely to arise and persist during extended stays in the intensive care unit (ICU) due to increased exposure to resistant hospital flora and selective antibiotic pressure. According to research by Manchanda et al. (2010)¹⁷ and Howard et al. (2012),¹⁶ 10.3% of cases involved recent surgery and wound exposure. Normal skin barriers are disrupted during surgical operations, creating entrance points for opportunistic organisms like *A.baumannii*. Even though they affect a lower percentage of patients (4.8%), comorbid disorders increase vulnerability by compromising host immune responses.

Conclusion

Particularly in ventilated patients, *Acinetobacter baumannii* was found to be highly prevalent in ICU-acquired illnesses. The most frequent source of isolation was respiratory samples. To lessen its burden in intensive care units, improved infection control and antimicrobial stewardship are crucial.

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