

Molecular Profiling and Phenotypic Characterisation of Carbapenem Resistant *Acinetobacter Baumannii* Isolates in Invasive Clinical Infections in A Tertiary Care Hospital.

Mahenaz Khan¹, Dr. Ramanath K²

¹Research Scholar, Department of Microbiology, Index Medical College Hospital & Research Centre, Indore, Malwanchal University. Madhya Pradesh- 452016

²Professor, Department of Microbiology, Index Medical College Hospital & Research Centre, Indore, Malwanchal University. Madhya Pradesh- 452016

ABSTRACT

BACKGROUND: The *Acinetobacter baumannii* has become a key nosocomial pathogen, especially within intensive care units, which is a critical cause of morbidity and mortality amongst the patients with critical conditions. They are linked with severe invasive infections like the septicemia and ventilator-associated pneumonia and sometimes urinary tract and soft tissue infections. Carbapenem-resistant *A. baumannii* (CRAB) is an increasing human health issue with the main cause being the synthesis of carbapenemase enzymes that degrade the efficacy of the last-resort antibiotics. These resistant strains can be detected early in order to provide effective treatment and avoid hospital outbreaks. Although the phenotypic assay such as the CarbAcineto NP test has been used to support diagnosis quickly, molecular techniques used in detecting the resistance genes (PCR) are more accurate.

OBJECTIVES:

- 1 To identify the occurrence of carbapenem resistance in clinical isolates of *A. baumannii* by the phenotypic CarbAcineto NP test.
- 2 Molecular diagnosis of OXA-type and MBL-type carbapenimase gene (blaOXA-51, blaOXA-23, blaOXA-24, blaOXA-58, blaNDM, blaVIM) by molecular technique (PCR).

METHODOLOGY: The present study included a cross-sectional study at IMCHRC, Indore, using 123 non duplicate isolates of *A. baumannii*, mostly blood and endotracheal aspirates. The production of carbapenemase was measured through CarbAcineto NP test and molecular analysis was conducted through the use of PCR.

RESULTS: Of the isolates, 64 percent were males with the majority of them being aged between 60-80 years. Most of them (73.2) were bloodstream infections. The phenotypic result was positive in 91.1 percent (112/123) with carbapenemase activity. The most frequently identified genotype was blaOXA-23, next was blaOXA-24, blaOXA-58 and MBL genes (blaNDM, blaVIM), and multiple genes were found to co-exist in several isolates.

CONCLUSION: CRAB is a very difficult problem in hospitals, particularly in ICUs. The strong percentage of carbapenemase detection highlights the need to incorporate a rapid phenotypic test such as CarbAcineto NP and molecular diagnostic to administer timely and specific therapy. Monitoring of resistance genes is a critical area in terms of informing antimicrobial stewardship and in the control of infection management practices.

Keywords: *Acinetobacter baumannii*, carbapenem resistance, CarbAcineto NP test, blaOXA-23, blaNDM, molecular diagnostics, ICU infections, antimicrobial resistance, phenotypic detection, multidrug resistance.

INTRODUCTION

One of the most alarming healthcare pathogens has been *Acinetobacter baumannii*, which is a gram-negative microbe that causes serious invasive infections such as Sepsis, Ventilator associated pneumonia, Urinary tract infections and Skin and Soft tissue infections [1]. This is a very dangerous pathogen because it can develop resistance to most antibiotics including carbapenems that are used as the last line of therapy against multidrug-resistant infections. The attention of numerous people has been on carbapenem-resistant *Acinetobacter baumannii* (CRAB) due to its

complicated treatments and increased mortality rates associated with the disease, particularly in critically ill patients or those with compromised immune systems or other underlying health complications [2].

CRAB causes severe complications in the ICUs, where patients are most susceptible, thus posing a major challenge on the part of healthcare providers. Not only are these infections more difficult to treat because of the limited antibiotic solutions, but they also are also a significant threat to patients with compromised immune defence. CRAB infections are known to be extremely fatal, which is why the additional treatments are necessary, to improve the state of infection control and further investigation of novel treatment methods [2].

Treatment of CRAB infections is usually initiated with first-line antibiotics including ampicillin-sulbactam, carbapenems, fluoroquinolones, aminoglycosides, and trimethoprim-sulfamethoxazole with high-dose ampicillin-sulbactam being particularly effective in serious infections. In the event that the bacteria are resistant to these drugs, second-line agents such as colistin, minocycline, or high-dose tigecycline may be used, usually in combination therapy, depending on the patient and on the prevalence of antibiotic resistance in a locality [3]. Cefiderocol is a new siderophore cephalosporin which is not particularly superior to the currently existing ones, but is one of the few agents that can be used in XDR CRAB, particularly those with OXA-type β -lactamases. Eravacycline which is normally used for intra-abdominal infections has shown promising in vitro activity against CRAB but its role in clinical situations is questionable due to the absence of well-defined breakpoints. Bacteriophage therapy is a customized treatment of resistant infections, and lytic phages may be used to complement antibiotics, although it is not routinely used. Sulbactam-durlobactam combination has demonstrated better survival in patients with CRAB pneumonia and could be used as a safer substitute to colistin.

Carbapenem resistance in *Acinetobacter baumannii* is largely explained by the expression of carbapenemases which are the enzymes that are able to hydrolyze carbapenem antibiotics and, therefore, render them ineffective. The most significant way through which *A. baumannii* develops resistance to carbapenems is through carbapenemase production [4]. There are many different carbapenemases which have been described as major contributors to this resistance: OXA-type carbapenemases (especially OXA-23 and OXA-24), KPC (*Klebsiella pneumoniae* carbapenemase), and NDM (New Delhi metallo- β -lactamase).

Their transfer is further promoted in the environment of the hospitals, when the transfer of these carbapenemase genes can occur either through plasmids or other mobile genetic elements [5]. The Indian literature has shown that 40-75 percent of *Acinetobacter baumannii* isolates are resistant to carbapenems antibiotics. The reported resistance rates have varied worldwide with the highest resistance rate being in Asia, Eastern Europe and Latin America with 48-85 being the reported resistance rates [6]. Rossi I et al. [7] found a rate of 0.7/1000 patient-day CRAB infections among adult Intensive care unit (ICU) patients.

According to the SENTRY Antimicrobial Surveillance Program (1997-2016), the prevalence of extensively drug-resistant (XDR) *Acinetobacter baumannii* was the highest in Europe (66.4%), Latin America (61.5%), Asia-Pacific (56.9%), and North America (38.8%). Colistin was the most effective agent as 95.9% of the isolates were susceptible [8]. According to the ATLAS Program (2018-2022) there was a steady high proportion of carbapenem-resistant *A. baumannii* (CRAB) and difficult-to-treat resistance (DTR) rate and these rates were over 25% in all regions except North America. It is believed to have been particularly high in the Asian-Pacific, Europe, Latin America and Middle East-Africa regions [9].

Carbapenem resistance in *A. baumannii* is an important phenomenon to be detected in order to receive timely and proper antimicrobial treatment. Nonetheless, the conventional algorithms of detecting the carbapenem resistance, including disk diffusion or automated, can require 24-48 hours to deliver the results, which can delay the process of effective treatment. In order to solve this, a number of phenotypic and molecular diagnostic techniques have been established to detect carbapenemase-producing organisms (CPOs) quickly.

Out of them, one of the most effective phenotypic tests to identify carbapenemase production in *Acinetobacter baumannii* is the CarbAcineto NP test that is both a very accurate and relatively inexpensive technique [10]. CarbAcineto NP test has a higher sensitivity of detection due to use of hyperosmotic 5 M NaCl solution to lysate bacteria as well as the elevation of bacterial inoculum. These changes make it easier to release and detect carbapenemase activity making it sensitive at 94.7% and specific at 100% in determining the presence of carbapenemase-producing *Acinetobacter* spp. [11]. The method of direct clinical sample detection of carbapenemase activity by the detection of carbapenemase activity on clinical samples: the Carbapenem Inactivation Method (CIM) and its modifications, including the Blood-Modified Carbapenem Inactivation Method (Blood-MCIM) confirmed itself to be more dependable and cost-effective based on the CLSI guidelines 2024 [12, 13].

Molecular procedures especially polymerase chain reaction (PCR) provides accurate detection of resistance genes and do give information about the genetic nature of carbapenem resistance. Such tools as PCR make it possible to detect

certain carbapenemase genes and promote the molecular characterization of the resistant strains and the further understanding of the genetic diversity of carbapenem-resistant *A. baumannii* [14].

Acinetobacter baumannii that is carbapenem-resistant (CRAB) has relatively wide genetic diversity, whose specific sequence types (STs) and clonal complexes (CCs) play a role in its spread and resistance characteristics on the global scale. Multilocus sequence typing (MLST) has established a number of high-risk clones. ST2 is the commonest among them, and it is highly related to the blaOXA-23 gene encoding the blaOXA-23, which is the class D carbapenemase protein that causes high-level resistance. Other well-known types of sequences are ST208 and ST369, which are members of Clonal Complex 92 (CC92), ST229 has been associated with localized outbreaks in some regions of India, and this points to regional differences in the distribution of strains.

Multiple resistance determinants, particularly, the class D b-lactamase genes like blaOXA-23, blaOXA-24, and blaOXA-58 are common in these high-risk clones. These genes can also be further improved through the insertion sequences such as the ISAbal which is a potent promoter when it is placed upstream of the resistance genes [15].

The genetic mechanisms of carbapenem resistant can help in the designing of specific therapeutic interventions, as well as contribute to preventing the further evolution of this extremely resistant pathogen. With the current increase in antimicrobial resistance in the world, the results of this study may give significant information on how to deal with the issue of CRAB and the improvement of patient outcomes in clinical practices.

Due to the dire need of quick detection techniques, this research will examine the phenotypic and molecular features of carbapenem-resistant *Acinetobacter baumannii* strains on invasive infections in a tertiary care hospital, Indore, India. The study is aimed at comparing the effectiveness of the CarbAcineto NP, which is a phenotypic assay, and molecular methods like PCR in detecting carbapenemase producing *A. baumannii*.

OBJECTIVES

- To identify the resistance of carbapenems of clinical isolates of *Acinetobacter baumannii* using a phenotypic test CarbAcineto NP test.
- Identify OXA type and MBL type of carbapenem-resistant *Acinetobacter baumannii* genes.

METHODOLOGY

The study was conducted as a cross-sectional study at Index Medical College Hospital & Research Centre, Indore where 123 non-duplicate isolates of *A. baumannii* of invasive nature were involved with majority of the isolates being of blood and endotracheal aspirates. The production of carbapenemase was evaluated by the CarbAcineto NP test and PCR was conducted as an evaluation of molecular analysis.

PHENOTYPE IDENTIFICATION OF CRAB: CARBACINETO NP TEST

There are 123 *Acinetobacter baumannii* isolates tested, of them 91.1% (n=112) were found to be carriers of carbapenemase, 74.1% of them (n=83) were found in blood samples and the rest of 25.9% (n=29) in endotracheal aspirates. The rest 8.9% (n=11) were non-carbapenemase producers with 63.6 percent (n=7) of them being of blood samples and 36.4 percent (n=4) of them being of endotracheal aspirates.

Of the isolates, with carbapenemase production:

- 14 isolates (11.3%) changed their colour rapidly (within 15 minutes) to yellow, which is characteristic of a high carbapenemase activity.
- There was a delayed but conclusive enzyme activity with 98 isolates (79.6%) becoming positive after 2 hours of incubation.

These results support the fact that carbapenemase producing *Acinetobacter baumannii* is highly prevalent in the environment, and the detection time difference may reflect the difference in the intensity of enzyme activity within the isolates.

Phenotypic detection of carbapenemase production by the Carbacineto NP test of the CRAB isolates tested was found to be statistically significant with the sample type ($p = 0.032$), with a significantly greater rate of detection in blood isolates than endotracheal isolates.

CARBAPENIN-RESISTANT ACINETOBACTER BAUMANNII MOLECULAR CHARACTERISATION.

Carbapenem-resistant *Acinetobacter baumannii* (CRAB) isolates were also analysed by molecular techniques which showed that they have Class D oxacillinases with the blaOXA-51 gene being present in all 123 isolates. Also, blaOXA-

23 was found in 104 isolates, blaOXA-24 in 26 isolates and blaOXA -58 in 54 isolates. There was the presence of the NDM metallo-b-lactamase genes in 93 isolates and VIM was confirmed in 22 isolates.

Co-existence of OXA & MBL genes
Genes Positive percentage (n=123)

blaOXA-23	104 (84.6%)
blaOXA-24	26 (21.1%)
blaOXA-58	54 (43.9%)
blaNDM	93 (75.6%)
blaVIM	22 (17.9%)

Genes co-existence with blaOXA-23 positives (n=104)

Genes Positive percentage (n=104)

blaOXA-24	26 (25%)
blaOXA-58	54 (51.9%)
blaNDM	93 (89.4%)
blaVIM	22 (21.2%)

Blastomeres with blaOXA-24 positives (n=26) co-existed with genes.

Genes Positive percentage (n=26)

blaOXA-58	2 (7.7%)
blaNDM	21 (80.8%)
blaVIM	4 (15.4%)

Co-existence between genes and blaOXA-58 positives (n=54)

Genes Positive percentage (n=54)

blaNDM	37 (68.5%)
blaVIM	11 (20.4%)

Out of 93 blaNDM positive isolates, none of them contained blaVIM gene.

A. baumannii Oxacillinases genes, Class D.

Acinetobacter baumannii has the blaOXA-51 gene that is intrinsically located there and is used as a species-specific marker to identify the bacteria. Its association with Acinetobacter baumannii is also intrinsic as its blaOXA-51 gene is present in all of the 123 isolates related to blood and endotracheal aspirates. The blaOXA-51 gene was observed in all the 123 CRAB isolates and its existence was statistically significant (p value is 0.049).

blaOXA-23 gene: blaOXA-23 gene was identified in 104 CRAB isolates and the presence of it was found statistically significant (p value is 0.016). It was found in 73 of 90 (81.1) blood isolates and 31 of 33 (93.9) endotracheal aspirate isolates.

blaOXA-24 gene the blaOXA-24 gene was detected in 26 of CRAB isolate containing statistically significant value (p value is 0.01.0) It was observed in 17/90 (18.9) and 9/33 (27.3) blood and endotracheal isolates respectively.

blaOXA-58 gene: The blaOXA-58 gene was found in 54 CRAB isolates and its presence was statistically significant (p value is 0.026). It was found in 39/90 (43.3%) blood isolates and 15/33 (45.4%) endotracheal aspirate isolates.

A. baumannii with A. baumannii Class B metallo-beta-lactamase genes in A. baumannii.

NDM gene: The MBL Carbapenemase - NDM gene was observed in 93 (CRAB) isolates and its occurrence was found to be statistically significant (p value is 0.021). NDM gene prevalence in 74/90 (82.2) blood isolates and 19/33(57.5) endotracheal aspirate was observed.

VIM gene: VIM gene was identified in 22 CRAB isolates and the presence of the gene was statistically significant (p value is 0.049). VIM gene was found in 16 of 90 (17.7) blood isolates and 6 of 33 (18.1) endotracheal isolates.

DISCUSSION

Acinetobacter baumannii is a pathogen that has a significant clinical implication because it contributes a big part to hospital-acquired infections. The fact that it can withstand the environment of several years and develop resistance to most of existing antibiotics makes it a challenging microorganism to deal with in particular. *A. baumannii* infections have been linked with high morbidity and mortality and can be a major therapeutic problem [16].

The resistance to carbapenem antibiotics has been rising drastically in the past few years and it has been reported that the resistance goes beyond 90 percent. *A. baumannii* is a major cause of serious nosocomial infections such as bacteraemia and ventilator-associated pneumonia. Although carbapenems were considered as one of the most effective treatment options available when treating these infections, the emerging resistance has severely restricted this treatment option.

Carbapenemase enzymes are the major determinants of carbapenem resistant in *A. baumannii* since the enzymes degrade these antibiotics. These are Class D oxacillinases (blaOXA-23-like, blaOXA-24-like, and blaOXA-58-like), Class B metallo-β-lactamases (blaNDM-like, blaVIM-like, blaIMP-like, and blaSIM-like) and Class A carbapenemases (blaGES-like and blaKPC-like). Also, it has the possibility of resistance via non-carbapenemase mechanisms, such as efflux pump genes overexpression and decreased membrane permeability, which complicates treatment approaches.

Out of the 123 clinical isolates of the *Acinetobacter baumannii* that we studied, 64 percent of the isolates were male and 36 percent were female patients, giving a male to female ratio of 1.8:1. This distribution is in agreement with a study conducted by Matsui et al. which revealed the same gender distribution amongst *A. baumannii* isolates [17]. Research of 676 *Acinetobacter* patients reported that, 60.2 percent of the patients were male and 39.8 percent were female giving a male-to-female ratio of about 1.5:1.

Our study showed that the age distribution of clinical isolates was such that the highest percentage of patients with carbapenem-resistant *Acinetobacter baumannii* (CRAB) were 60 to 80 years old, and then followed 45 to 60 years old. This trend is similar to the available literature, which says that the CRAB infection is more common in older adults. According to a study conducted by Said et al. [18] the proportions of CRAB were significantly lower in the young patients where the proportion was only 0.6% in patients under one year of age and only 1.3% in patients aged 1-19 years. Their highest proportions were found among adult patients aged 20 to 79 years with a 7.7 percent proportion between 20 and 39 years, 6.2 percent between 40-59 years, and 5.8 percent between 60 to 79 years. Surprisingly, there was a reduction in the rate of CRAB infection in patients above 79 years of age (1.9%).

These data indicate that the tradition of CRAB infections is the greatest in older adults, that is, individuals aged 60-80 years, which is probably associated with the underlying comorbidities, the presence of frequent hospitalization, the duration of exposure to healthcare settings, as well as the extensive use of antibiotics [18].

The sample-wide distribution of CRAB isolates in our study, majority of the carbapenem-resistant *Acinetobacter baumannii* (CRAB) isolates were found in blood samples (90 isolates), mostly in patients with septicaemia and 33 in patients with pneumonia endotracheal aspirates. The study by Adejei et al. also indicated the same finding as this work, showing a domination of CRAB isolates in blood and tracheal aspirate samples. Their paper, which centered on the phylogenetic study of *Acinetobacter baumannii* strains, also provides an argument in favor of the importance of these clinical sources in CRAB infections. The emphasis on the crucial role of the pathogen in severe infections is made by the high prevalence of CRAB in blood and respiratory samples, especially in the critically ill and hospitalized patients [19].

Study Blood ET Aspirates Others.

Vijaykumar et al. [20]	30	73	NIL
Shi et al. [21]	44	NIL	50
Kalal et al. [22]	2	60	38
El-Badawy et al [23]	12	NIL	20
Strateva et al. [24]	NIL	45	55

In the current study the *Acinetobacter baumannii* was most commonly isolated in the blood samples (n = 90) and other isolates were also made in the endotracheal aspirates (n = 33) indicating that it is the cause of not only bloodstream infections but also ventilator-associated respiratory infections. This occurrence is in line with the findings of previous studies, with the article by Shi et al. found more of the isolates in blood (n = 44) as compared to the cerebral fluid (n = 50), showing the role of the organism in systemic infections in critically ill and immunocompromised patients.

Likewise, the study by El-Badawy et al. has found *A. baumannii* isolates produced by cerebrospinal fluid and blood (n = 20 and n = 12, respectively), supporting the notion that it is a leading agent of healthcare-associated invasive infections.

On the other hand, Vijaykumar et al. demonstrated that more isolates were obtained as a result of the ET aspirates (n = 73) than blood (n = 30), which highlights the clinical importance of *A. baumannii* in the respiratory diseases, especially in patients who are mechanically ventilated. Kalal et al. also reported that there was a considerable preponderation of the isolates of ET aspirates (n = 60) and only minimal numbers of blood (n = 2), implying that it was nosocomial transmission between intubated patients. Also, isolates of respiratory (n= 45) and other clinical (n= 55) specimens were reported by Strateva et al. (no respiratory blood sources). This also indicates the broad clinical spectrum and tissue predilection of the organism.

All these results support the pivotal role of *A. baumannii* in respiratory and bloodstream infections, especially in the tertiary care and intensive care units. The fact that the blood isolates prevail in our current study can either indicate that there is an increased burden of the invasive disease among our patient group or that our current patient group may be a result of strict diagnostic measures, e.g., regular blood culture surveillance among suspected sepsis. This flexibility of sample distribution in various studies has highlighted the flexibility of the pathogen, its tendency to transmit in a nosocomial setting, and its capacity to impose serious infections on susceptible populations of patients.

PATTERN OF ANTIMICROBIAL SUSCEPTIBILITY.

In the current study, 123 clinical isolates of *Acinetobacter baumannii* were all resistant to a wide spectrum of commonly used antibiotics, which demonstrates the highly threatening Extensively drug-resistant (XDR) phenotype of the pathogen. It is important to note that all the isolates were resistant to ceftazidime, cefepime, meropenem, imipenem, amikacin, gentamicin, ciprofloxacin, levofloxacin, and polymyxin B. The carbapenem resistance was further described by the existence of different carbapenemase genes as presented in the molecular characterization section.

CONCLUSION

CRAB is a tough issue to deal with in the hospital environment, particularly the ICUs. The reason, which stems the high rates of detecting carbapenemases, is the need to combine speedy phenotypic tests such as CarbAcineto NP and molecular diagnostics to administer therapy in a timely and tailored manner. Resistance gene surveillance is imperative in the creation of antimicrobial stewardship and informing infection control practices.

REFERENCES

- [1]. Wong D, Nielsen TB, Bonomo RA, Pantapalangkoor P, Luna B, Spellberg B, Clinical and Pathophysiological Overview of *Acinetobacter* Infections: a Century of Challenges. *Clin Microbiol Rev.* 2017 Jan;30(1):409-47.
- [2]. Cisneros JM, Rodriguez-Bano J. Nosocomial bacteremia caused by *Acinetobacter baumannii*: epidemiology, clinical presentation and treatment. *Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis.* 2002 Nov;8(11):687-93.
- [3]. Bassetti M, Righi E, Esposito S, Petrosillo N, Nicolini L. Drug treatment of *Acinetobacter baumannii* multidrug-resistant infections. *Future Microbiol.* 2008 Dec;3(6):649-60.
- [4]. Paul DR, Singh DK. Basic principles of Molecular Diagnostics in Clinical Microbiology. Dentomed Publication House, 2024. 151 p.
- [5]. Lynch JP, Zhanel GG, Clark NM. *Acinetobacter baumannii* Infection in the ICU: Treatment. *Semin Respir Crit Care Med.* 2017 Jun;38(3):311-25.
- [6]. Pathogenic *Acinetobacter* species such as the new *Acinetobacter dijekshoorniae* in market meat in Peru - PubMed [Internet]. [cited 2025 Mar 19]. Retrieved at:<https://pubmed.ncbi.nlm.nih.gov/31226568/>
- [7]. Dancer SJ. Hospital-acquired infection control: pay attention to the part of the environment and emerging technologies in the process of decontamination. *Clin Microbiol Rev.* 2014 Oct;27(4):665-90.
- [8]. Polyzou E, Schinas G, Spernovasilis N, Gogos C, Dimopoulos G, Akinosoglou K. Preventing Multidrug-Resistant Bacterial Transmission in the Intensive Care Unit with a Comprehensive Approach: A Policymaking Manual. *Antibiotics.* 2023 Jul 30;12(8):1255.
- [9]. Risk-factors of acquisition of imipenem-resistant *Acinetobacter baumannii* in Spain: a study nationwide - PubMed [Internet]. [cited 2025 Mar 21]. Referred to:<https://pubmed.ncbi.nlm.nih.gov/16216101/>
- [10]. Lee SO, Kim NJ, Choi SH, Hyong Kim T, Chung JW, Woo JH, et al. Risk factors in the acquisition of imipenem-resistant *Acinetobacter baumannii*: a case-control study. *Antimicrob Agents Chemother.* 2004 Jan;48(1):224-8.
- [11]. Ye JJ, Huang CT, Shie SS, Huang PY, Su LH, Chiu CH, et al. Multidrug resistant *Acinetobacter baumannii*: risk factors to emergence of imipenem resistant strains on patients who had previously had susceptible strains. *PLoS One.* 2010 Apr 1;5(4):e9947.
- [12]. Thacharodi A, Vithlani A, Hassan S, Alqahtani A, Pugazhendhi A. Carbapenem-resistant *Acinetobacter baumannii* concerns the world with new antibiotic regimens. *iScience.* 2024 Dec 20;27(12):111367.

- [13]. Falagas ME, Kopterides P. Risk factors of the isolation of multi-drug-resistant *Acinetobacter baumannii* and *Pseudomonas aeruginosa*: a literature systematic review. *J Hosp Infect*. 2006 Sep;64(1):7-15.
- [14]. Bartal C, Rolston KV, Nesher L. Carbapenem-resistant *Acinetobacter baumannii*: colonization, infection and existing treatment options. *Infectious diseases and therapy*. 2022 Apr;11(2):683-94.
- [15]. Lye DC, Earnest A, Ling ML, Lee TE, Yong HC, Fisher DA, et al., Effects of multidrug resistance in healthcare associated and nosocomial Gram-negative bacteraemia on length of stay and mortality: cohort study. *Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis*. 2012 May;18(5):502-8.
- [16]. Bonomo RA, Szabo D. Multidrug resistance in *Acinetobacter* species and *Pseudomonas aeruginosa*: mechanisms. *Clin Infect Dis Off Publ Infect Dis Soc Am*. 2006 Sep 1:43 Suppl 2:S49-56.
- [17]. Matsui M, et al. Distribution and Molecular Characterization of *Acinetobacter baumannii* International Clone II Lineage Japan. *Antimicrob Agents Chemother*. 2018 Feb;62(2):e02190-17.
- [18]. Said D, Willrich N, Ayobami O, Noll I, Eckmanns T, Markwart R. The epidemiology of carbapenem resistance in *Acinetobacter baumannii* complex in Germany (2014–2018): an analysis of data from the national Antimicrobial Resistance Surveillance system. *Antimicrob Resist Infect Control*. 2021 Dec;10(1):45.
- [19]. Adjei AY, Vasaikar SD, Apalata T, Okuthe EG, Songca SP. Phylogenetic analysis of carbapenem-resistant *Acinetobacter baumannii* isolated from different sources using Multilocus Sequence Typing Scheme. *Infect Genet Evol*. 2021 Dec 1;96:105132.
- [20]. Vijaykumar S, Mathur P, Kapil A, Das BK, Ray P, Gautam V, et al. Molecular characterization & epidemiology of carbapenem resistant *Acinetobacter baumannii* isolated in India. *Indian J Med Res*. 2019 Feb;149(2):240-6.
- [21]. Shi X, Wang H, Wang X, Jing H, Duan R, Qin S, et al. Molecular characterization and antibiotic resistance of *Acinetobacter baumannii* in cerebrospinal fluid and blood. *PloS One*. 2021;16(2):e0247418.
- [22]. B.S. Kalal, et al. Molecular characterization of carba-penem-resistant *Acinetobacter baumannii* strains from a tertiary care center in South India. *Infectio* 2020; 24(1):27-34
- [23]. El-Badawy MF, Abdelwahab SF, Alghamdi SA, Shohayeb MM. Characterization of phenotypic and genotypic traits of carbapenem-resistant *Acinetobacter baumannii* clinical isolates recovered from a tertiary care hospital in Taif, Saudi Arabia. *Infect Drug Resist*. 2019 Oct 3;12:3113-3124. doi: 10.2147/IDR.S206691.
- [24]. Strateva TV, Sirakov I, Stoeva TJ, Stratev A, Peykov S. Phenotypic and Molecular Characteristics of Carbapenem-Resistant *Acinetobacter baumannii* Isolates from Bulgarian Intensive Care Unit Patients. *Microorganisms*. 2023 Mar 29;11(4):875.