2. REVIEW OF LITERATURE

2.1. Nosocomial infections from ICUs

Nosocomial infections occur worldwide and mainly affect developed countries. Nosocomial infections are the major causes of death and increased morbidity among hospitalized patients. It is estimated that 80% of all hospital deaths are directly or indirectly related to nosocomial infections.\(^{36}\) Studies have shown that nosocomial prevalence varies from 3.8% to 18.6% depending on the population surveyed.\(^{37}\) Other studies particularly in developing countries, Jroundi et al, 2007 found prevalence of nosocomial infections was 17.8% and higher in ICUs (50%).\(^{38}\) Allegranzi et al., 2011 also documented 15.5% of prevalence, but is higher when compared with studies from developed countries (5.7%–6.8%).\(^{39}\)

Intensive care units are a specialized section of the hospital that provides continuous care for persons who are critically ill. More than 20% of all nosocomial infections are acquired in ICUs.\(^{40}\) The frequency on infections at different anatomic sites and the risk of infection vary by the type of ICUs like burn ICUs, surgical ICUs. ICU patients are very susceptible to infections due to acquired defects in host defence mechanisms from the immune suppressive affect of for example the underlying disease, recent surgery, trauma and concurrent drug therapy.\(^{41}\) This is a population of patients which often have multi-organ failure (MOF) and as a consequence of this, are exposed to invasive procedures.

The main studies on the incidence of ICU acquired infections are presented in infection rates vary between hospitals and according to the type of population studied, being highest in burn units and surgical and trauma ICUs and lowest in coronary care units \(^{42,43,44}\)

2.2. Nonfermenting Gram Negative Bacilli

The causal microorganisms differ, depending on the origin and source of infection. A study by Alberti et al, 2002 found gram-negative bacilli predominated (49% of isolates), followed by gram positive cocci (37%) and fungi (9.7%) in ICU-acquired infections.\(^{45}\) In a recent multicentre sepsis study, 62% of infections are
microbiologically documented, while gram-positive cocci accounted for 47% of ICU-acquired infections and gram-negative bacilli for 53%. NFGNB are known to account for about 15% of all bacterial isolates from a clinical microbiology laboratory. NFGNB, that were previously considered to be contaminants but now emerged as important nosocomial pathogens. Rits K in 2013 studied the prevalence of nonfermenters isolated from different clinical samples along with their susceptibility profile by Conventional bacteriological methods and found Pseudomonas aeruginosa and Acinetobacter baumannii most predominant pathogen along with Acinetobacter lowffii, Pseudomonas fluorescens, Pseudomonas stutzeri, Burkholderia cepacia, Stenotrophomonas maltophilia, Achromobacter xylosoxidan and Shewanella putrefaciens. Most of the isolated organisms were multi drug resistant. NFGNB isolated from in-patient was more than out-patient as seen in studies mainly ICUs and most common non fermenters were Pseudomonas and Acinetobacter. Soad AA et.al. 2010 did a retrospective study and documented the rate of Hospital acquired infections in ICUs in three consecutive years (2006-2008) and they found Acinetobacter was second most common pathogenic organism (rate of infection >20%) in all three years. The most frequently isolated microorganism isolated from ICUs was Acinetobacter, 21% isolates of which were MDR. Samanta P et.al. 2013 most commonly isolate NFGNB in their study and found Acinetobacter spp. followed by Pseudomonas spp., Burkholderia cepacia complex, S. maltophilia.

2.3. *A.baumannii*: Nosocomial pathogen

*Acinetobacter* spp. has been growing for the past 30 years. *Acinetobacter* spp. is emerging as an important pathogen in healthcare settings. *A.baumannii* is a common cause of nosocomial infections worldwide. The genus *Acinetobacter* has a complex history, and only recently it has become possible to distinguish various species within the genus. Phylogenetic classification places the genus *Acinetobacter* in the group of *Gammaproteobacteria* of the *Proteobacteria*. The history of the genus *Acinetobacter* begins in 1911 when the Dutch microbiologist Beijerinck isolated a micro-organism from soil by enrichment in calcium acetate containing minimal medium and was named as *Micrococcus calcoaceticus* and assigned to different genera and species, e.g; *Achromobacter anitratus*. It belongs to the family Neisseriaceae. It can be
misidentified as *Neisseria* or *Moraxella* species on gram staining. In 1954 Brisou and Prévot proposed the genus *Acinetobacter* (greek- akinetos, i.e nonmotile) to separate the non-motile micro-organisms from the motile genus *Achromobacter*. Twelve *Acinetobacter* genospecies were delineated on the basis of DNA-DNA hybridization.\[^{57}\] In which *Acinetobacter baumannii* belong to genospecies 2. Further work done by Bouvet and Jeanjean, Tjernberg and Ursing, added to the description of further *Acinetobacter* genomic species.\[^{58,59}\]

*Acinetobacter* is a gram-negative coccobacillus that has emerged as an important nosocomial pathogen. It is non-motile, encapsulated, non-fermentative, indole negative, oxidase negative and catalase positive. *Acinetobacter* is ubiquitous in the outside environment and has been isolated from hospital personnel, and hospital equipments.\[^{60,61}\] It is strictly aerobic, and does not require unusual nutrients to survive in the environment. *Acinetobacter* is easily grown on routine laboratory media (e.g. Tryptic soy agar). A propensity to tolerate drying and resistance to multiple classes of antibiotics are the key factors in enabling the organism to survive and spread in the nosocomial environment.

The incidence of *A. baumannii* infections has risen over the past decades \[^{62,63}\], and recent studies indicate that this pathogen is more resistant and virulent, and becoming a serious nosocomial threat, likely because of patient-to-patient transmission via the hands of health care workers from a common environmental source.\[^{64,65}\] Vincent JL et.al., 2009 analyse global epidemiology of infection in ICU and found most common gram negative organism were *Pseudomonas*, *E.coli* and *Acinetobacter* species.\[^{10}\] There was considerable variation in the type of organisms isolated from different geographical regions; rates of infection of *Acinetobacter* differed most markedly, ranging from 3.7% in North America to 19.2% in Asia.\[^{10}\]

In 2009, the Infectious Diseases Society of America (IDSA) set the acronym ESKAPE, which lists the groups of pathogens that pose the highest threat to patients’ safety and to public health \[^{66}\], one of which is *Acinetobacter baumannii*. Jaggi N,2012 analyse nosocomial status and pathogenic potential of *A.baumannii*, found 9.4% prevalence and pathogenic potential of around 54.6%.\[^{67}\] Dash M. 2013 isolate only 3% of *Acinetobacter*\[^{68}\] while in Alsehlawi ZS et.al.,2014 identified 6.8% *A.baumannii* among GNB\[^{69}\]. Mixed infections are frequent in cases of *Acinetobacter* infection, and
this observation has opened a debate on the importance of bacterial synergy in cases of bacteraemia.\(^{[70]}\)

### 2.4. *A. baumannii*: Risk factors

*A. baumannii* can survive on various surfaces within hospital settings, including catheters and other medical equipments. Thus environmental contamination is an important source of infection as pathogens are spread directly from surfaces or through the hands of healthcare workers to patients.\(^{[60]}\) The main risk factors of *Acinetobacter* infections are invasive procedure e.g. catheterization, mechanical ventilation, nasogastric tube,\(^{[71,72]}\) another major risk factors are prolonged hospital stay, ICU stay, widespread use of broad spectrum antibiotics or previous use of third generation cephalosporins.\(^{[73]}\) The risk factors within the ICU’s concern the immunosuppressed patients, patients previously exposed to antimicrobial therapy, high invasive procedures and patients who suffered from previous sepsis.\(^{[5]}\) Other risk factors include pneumonia as a source of infection, inappropriate empirical treatment and prior treatment with carbapenems.\(^{[74]}\) Surgical procedures performed within the emergency operating theatre is another major risk factor contributing to the spread of *A. baumannii*, however Villers et.al. found the previous use of fluoroquinolones as a main risk factor.\(^{[75]}\)

### 2.5. *A. baumannii*: Virulence Factors

*A. baumannii* have very few virulence factors\(^{[73]}\), however some strains have virulence factors associated with invasiveness, transmissibility or the enhanced ability to colonise immunocompromised patients.\(^{[76]}\) Adhesion to surfaces is the capacity that secures the anchoring of the cell to mucosal surfaces and to cells of the host. However, the ability of *A. baumannii* to adhere to or even invade cells was generally found to be lower compared to other microorganisms.\(^{[77,78,79]}\) McConnell MJ et.al. characterised virulence factors of *A. baumannii* that contribute to its pathogenesis, with a focus on motility, adherence, biofilm formation, and iron acquisition. In addition, the outer membrane protein OmpA, phospholipases, membrane polysaccharide components, penicillin-binding proteins and outer membrane vesicles also identified the virulence factors.\(^{[80]}\) Outer membrane proteins have been shown to contribute to the virulence of *A. baumannii*. OmpA was implicated to facilitate adhesion \(^{[79]}\), particularly to epithelial cells from the airways \(^{[78]}\).
In a study by Boujaafar et. al. (1990), it was demonstrated that *A. baumannii* strains isolated from catheters and tracheal devices expressed higher surface hydrophobicity than strains obtained from normal skin.\(^8\) Rasha JM et al. found all the *A. baumannii* have the ability to form biofilm.\(^8\) Gelatinase enzyme secreted by *A. baumannii* which can cross cell membrane and hydrolyse collagen in subcutaneous tissue and certain bioactive peptides also produced by *Acinetobacter* which participated in inflammatory reactions.\(^8\)

### 2.6. *A. baumannii*: Clinical manifestations of infections

The outcomes of patients with *A. baumannii* infections seem to be poorer if caused by isolates with resistance to multiple classes of antimicrobial agents.\(^8\) Patients at risk of developing *A. baumannii* infections are those with immunosuppression, prolonged hospitalization and critically ill patients in ICUs.\(^7\) *Acinetobacter* spp. has been implicated in ventilator-associated pneumonia, catheter related blood stream infections, urinary tract infections, cerebrospinal-shunt-related meningitis, and wound infections. Pneumonia, mainly ventilator-acquired pneumonia (VAP), is the most commonly identified clinical manifestation of *A. baumannii*.\(^7,8\) This could be attributed to the colonization of airways by *A. baumannii* which soon develop into true pneumonia upon prolonged hospital admission and extensive administration of antibiotics. Furthermore, data from the National Nosocomial Infections Surveillance (NNIS) System in USA show that ICU VAP rates due to *A. baumannii* reached 7% in 2003.\(^8\) *Acinetobacter* spp. represent one of the most important agents causing VAP together with *S. aureus*, *P. aeruginosa* and Enterobacteriaceae.\(^7,8,9\) Munoz-Price et.al found 17% -19% of VAP caused by *A. baumannii* as well as *Pseudomonas*, in an ICU.\(^9\) And according to Rocha LA et.al. (2008) and Jaggi N 2012 documented 18% \(^7\) and 30.4% \(^9\) respectively VAP was due to *A. baumannii*. A recent study by Lee et al. (2012) was studied the different clinical manifestations associated with *A. baumannii* vs *A. nosocomialis* in pneumonia and revealed that *A. baumannii* pneumonias seem to be more severe and more likely to have associated abnormal haematological findings.\(^9\) Jaggi N 2012 also documented that *A. baumannii* contributed 35.2% Catheter Associated Blood Stream Infections, 12.5% Surgical Site Infections and 2.94% Catheter Associated Urinary Tract Infections.\(^9\) Pradhan NP, 2014 found respiratory infections were the commonest (65.8%), followed by urinary infections (17.1%) and
dual infections (urinary plus respiratory) (17.1%) While Rajmane VS 2015 isolate *Acinetobacter* predominantly (55.81%) from pus followed by sputum (19.76%), urine (6.97%), blood (4.65%), endotracheal secretion, IV catheter (3.48% each) and CSF (2.32%).\[^{93,94}\]

Bacteraemia is currently one of the infections with the highest mortality rate in hospitals. The most common source of *Acinetobacter* BSI is respiratory tract.\[^{95}\] In the USA, *A. baumannii* was found as the 10th most common aetiologic agent in nosocomial blood-stream infections.\[^{85}\] *A. baumannii* bacteraemia can be secondary to pneumonia, and can also result from central-venous line catheters, which act as a main route for dissemination of organisms into the blood stream.\[^{96,97}\] One study reported sepsis and/or septic shock in 19% of patients with *Acinetobacter* bacteraemia.\[^{98}\] This observation highlighted the true pathogenicity of a few strains, with a crude mortality rate of c. 42%. An attributable mortality rate of 7.8% found in one survey was related to a delay in the initiation of appropriate therapy.\[^{99}\] According to Falagas ME, 2007 the mortality rate of 7.8% to 23% accounted for hospitalised patients with *A. baumannii* infection and the rate was 10% to 43% among ICU admitted patients with *A. baumannii* infection.\[^{100}\]

*A. baumannii* also causes urinary tract infection, often related to indwelling Foley catheters.\[^{76,101}\] *A. baumannii* may cause wound colonization and infection in patients with severe burns or trauma. *A. baumannii* also related to cerebrospinal shunt-related meningitis in neurosurgical patients.\[^{102}\]

Another pathway that can lead to the development of nosocomial pneumonia or bacteraemia involves bacterial overgrowth in the stomach. This process may occur under conditions of reduced acid secretion in the stomach, such as occurs in many ICU patients. *Acinetobacter* has been shown to grow under these conditions.\[^{103}\]

### 2.7. *A. baumannii* : Identification

Identification of Acinetobacter species are done by phenotypicaly which is quite inadequate and insufficient. For the identification of *Acinetobacter* species, several genotypic methods have been shown to be adequate.\[^{104}\]
Furthermore, phenotypic identification by commercial colorimetric systems has been associated with poor accuracy.\cite{76} Many studies identify *Acinetobacter* phenotypic method described by Gerner-Smidt et al. and Bouvet and Grimont using a panel of 25 carbon assimilation tests. These phenotypic tests are able to identify most of the genomic species except isolates belonging to DNA group 2 and 13. Gerner-Smidt and Bouvet were further biotype *Acb*-complex by additional 5 carbon assimilation tests of carbon sources.\cite{105,106} Many studies identified *Acinetobacter* spp. by simplified phenotypic identification test which is cost effective.\cite{107,108,109} Ajao AO compare different media CHROMagar Acinetobacter when compared to sheep blood agar, MacConkey agar and MacConkey agar with 6 μg/ml of imipenem for isolation and identification and reveal Sheep blood agar and CHROMagar detected all *Acinetobacter* and MDR-*A. baumannii*. CHROMagar may be useful for rapid detection of MDR-Acinetobacter.\cite{110} *Acinetobacter* spp. can be identified by conventional methods such as Vitek \cite{69,104,111} and API20E \cite{112,113} and gave sufficient results but Bergogne Berezin, 2001\cite{114} not considered a reliable identification method with API 20E and NE and in 2009. Therefore, genotypic methods have been proposed for fast and yet accurate identification of *Acinetobacter* species. After phenotypic and conventional method of identification, *Acinetobacter* spp. were confirmed by genotypic method.\cite{111,108} Among the genotypic methods, 16S rRNA gene sequencing is one of the most commonly used for bacterial identification.\cite{115} The RNA polymerase β-subunit (rpoB) gene sequences are one of the most useful tools for the identification and taxonomic classification of various bacterial species, including *Acinetobacter* spp.\cite{116,117} Khosravi AD et al. (2015) revealed the sequence analysis of the 16S rRNA and rpoB spacer simultaneously was able to do identification of Acinetobacter spp. to species level.\cite{118} Sequence analysis of the rpoB gene has been found to be a reliable and rapid method for identifying *Acinetobacter* species. For 16S rRNA gene sequencing, universal PCR primer pairs that enable PCR amplification from all bacteria are available. By contrast, rpoB gene sequencing uses a PCR primer pair specific to the genus of bacteria to be identified most of the time. If the bacteria to be identified are not *Acinetobacter* species, they cannot be identified because they give negative results in *Acinetobacter*-specific rpoB PCR. Therefore, when the genus of the bacteria to be identified is unknown, it would be more convenient to first identify it by 16S rRNA gene sequencing.\cite{119} Newer method for identification of different species includes High
resolution fingerprinting with AFLP PCR, RFLP with digestion of PCR amplified sequence and analysis of various DNA sequences.

However these genotypic methods have high sensitivity and advantageous but not appropriate for routine clinical laboratory because of certain limitations such as Cost: more expensive than other methodologies and molecular methods require dedicated space that may not be available in some clinical laboratories.

While automated methods give save labor cost, greater speed, more reproducible results over a long period of time thus there is increasingly used in routine analyses.

### 2.8. *A.baumannii*: Antimicrobial Resistance pattern

Initial Indian studies in the 21st century showed that *Acinetobacter* species were fairly sensitive. For instance, Suri et al. (2000) demonstrated *Acinetobacter* in patients from a neurosurgical unit and it was sensitive to ciprofloxacin, amikacin cefotaxim and ceftriaxone. Prashanth and Badrinath (2004) isolated *Acinetobacter* which was sensitive to amikacin and ceftazidime and resistant to ciprofloxacin and cefotaxime. Gladstone et al. (2005) from Vellore reported a prevalence of 14% carbapenem-resistant *Acinetobacter* spp., isolated from tracheal aspirates. Banerjee et al. (2005) isolated *Acinetobacter* from different body fluids which have good sensitivities for gentamycin. Prashanth and Badrinath (2006) showed gradually increasing resistance of *Acinetobacter*.

*A.baumannii* has caused numerous global outbreaks and displayed ever increasing rates of resistance. The general mechanisms of antibiotic resistance in *Acinetobacter* spp. include enzyme-mediated resistance, genetic adaption, efflux pumps, porin mutations, changes in the structure of outer membrane components and production of acquired carbapenem-hydrolyzing class D β-lactamases (CHDLs). *Acinetobacter* spp. have acquired a variety of β-lactamases, the production of which affects porins in the outer membrane, making *A.baumannii* impermeable and, thus, resistant to antibiotics due to this impermeability of outer membrane cause difficulty in treatment.

*A.baumannii* has become resistant to many classes of antibiotics and is well suited for genetic exchange. *A.baumannii* is among a unique class of Gram-negative
bacteria that are described as “naturally transformable”.[125,126,127] As a result of the rapid acquisition of resistance genes to different and multiple classes of antibiotics, several drugs have already been eliminated from treatment options for *A.baumannii* infections such as β-lactamase (penicillins, cephalosporins), aminoglycosides, quinolones and tetracyclines.[128]

ESBL associated resistance among *Acinetobacter* species is now known. Sinha M 2007 speciate clinical isolates of *Acinetobacter*, identify the production ESBLs and compare the role of different cephalosporins in detecting ESBL production in the isolates and found relatively high levels (28%) of ESBL in *Acinetobacter* and may reflect the scenario in India.[129] ESBL production in *Acinetobacter* has been found to vary from 46% in Turkey[130] to 54.6% in Korea.[131]

The CLSI method for ESBL detection consists of the initial screen test and phenotypic confirmatory test.[24] In initial screening there are evaluation of susceptibilities to more than one of cefpodoxime, ceftazidime, ceftriaxone, cefotaxime, and aztreonam by using disk diffusion or broth dilution method. A decrease in susceptibilities to one or more antibiotics tested may indicate production of ESBLs and warrant performance of the subsequent phenotypic confirmatory tests by determining susceptibilities to cefotaxime and ceftazidime alone and those with clavulanate are compared using disk diffusion or broth dilution method.[124]

In between Double disk diffusion synergy test (DDST) as a screening method and cephalosporin/clavulanate combination discs as phenotypic confirmatory test for ESBL detection, Disc diffusion method is easy to perform and it is comparatively simple and cost effective.[132]

However, now a day's carbapenem resistance in *Acinetobacter* spp is observed increasingly worldwide. Metallo-β-lactamases (MBLs) confer a high level of resistance to carbapenem as well as all β-lactam except aztreonam, because of their strong hydrolytic activity against these antibiotics. MBL producing isolates also have the propensity to show concomitant resistance to multiple antimicrobial agents thus further limiting therapeutic options.[133] In India, it has been documented that approx 35% *Acinetobacter* sp. are found to be carbapenem resistance, the prevalence of carbapenem resistance was increasing greatly in *Acinetobacter* sp.[108,127,134] A study by Weinbren et
al., detected that isolates were resistant to carbapenem by disk diffusion method and revealed MICs of 0.5-2 µg/mL, which is below the recommended MIC breakpoint for resistant isolates.\textsuperscript{[135]} While a study by Sinha M (2007), majority of the isolates detected resistant by disk diffusion method were found to have MICs in the sensitive range.\textsuperscript{[136]}

There are various methods of detecting MBL; the most widely accepted standardized MBL functional screen is the MBL Etest. However, due to the high cost and unavailability of Etest strips, many clinical microbiology laboratories use alternative screening methods, such as the double-disk synergy test (DDST) and the combined disk (CD) assay.\textsuperscript{[137]} John S, 2011 Compared the phenotypic tests for MBL detection and DDST was reliable and reproducible, with separate EDTA and Zn disks it was also identification of ambler class B MBLs.\textsuperscript{[138]} Lee K. et.al. 2003 evaluate different test for differentiating MBL producing isolates of Pseudomonas and Acinetobacter spp. and they documented that MBLs can distinguished from other B-lactamase is done by inhibiting the activity of enzyme by EDTA(chelating agents).\textsuperscript{[139]} Hodge test can be used to screen carbapenamase producing gram negative bacilli but DDST [IPM-EDTA] can distinguish MBL-producing from MBL non-producing Gram negative bacilli.\textsuperscript{[140]} Lee K, 2003 reported that IPM-EDTA DDST detected all MBL producing Pseudomonas, while IPM-SMA(sodium mercaptoacetoacitic acid) DDST detect all MBL producing Acinetobacter.\textsuperscript{[139]} Yong et.al. also reported IPM-EDTA disk produces smaller inhibition zone for Acinetobacter then Pseudomonas.\textsuperscript{[141]} Sinha N, 2013 found MBLs was produced by 16% of \textit{Acinetobacter} isolates which detected by ethylene-diamine-tetra-acetic acid disc synergy test (EDTA-DST) and found better method then Modified Hodge Test (MHT).\textsuperscript{[142]} Noyal MJ, 2009 also found among various detection methods EDTA-disk synergy test is a relatively simple and sensitive method for MBL detection.\textsuperscript{[143]}

CLSI has recommended MHT for detection of carbapenemases activity in Enterobacteriaceae, but not in non-fermenters.\textsuperscript{[124]} Detection of genes coding for carbapenem resistance by PCR, usually give reliable and satisfactory results. A study by Shahcheraghi et. al. (2011) detect $\text{bla}_{\text{SPM-1}}, \text{bla}_{\text{GES-1}}, \text{bla}_{\text{OXA-51}}, \text{bla}_{\text{OXA-23}}$ genes among 6, 2, 94 and 84 isolates of the \textit{Acinetobacter} bacterium, respectively.\textsuperscript{[144]} Alsultan AA et al 2015 conclude $\text{blaOXA-23}; \text{blaOXA-24/40}, \text{blaVIM}$ and $\text{blaSPM}$ were the most prevalent genes in the carbapenem resistant \textit{A. baumannii}.\textsuperscript{[69]}
Ting et al. (2013) investigated the drug resistance genes of imipenem-resistant *A. baumannii* and detected TEM and OXA-23 genes among all the isolates, but the other genes such as SHV, CTX-M, DHA, CIT, IMP, VIM and KPC could not be detected from imipenem-resistant *A. baumannii*.\(^{[145]}\) Another research by Rezaee et al. (2013) revealed 37% of isolates carried at least one of the *bla*\(_{\text{PER1}}\) or *bla*\(_{\text{TEM-1}}\) genes and 13.15% of their studied isolates reported to harbor *bla*\(_{\text{TEM-1}}\) gene. *A. baumannii* isolates were harboring for *bla*\(_{\text{PER1}}\), *bla*\(_{\text{SHV}}\), *bla*\(_{\text{CTX-M-2}}\) and *bla*\(_{\text{GES-1}}\).\(^{[146]}\) Safari M (2015) found some of the genes including SHV (58%), TEM (20%) and VIM (30%).\(^{[147]}\) Alsultan AA et al. 2015 conclude *bla*\(_{\text{OXA-23}}\), *bla*\(_{\text{OXA-24/40}}\), *bla*\(_{\text{VIM}}\) and *bla*\(_{\text{SPM}}\) were the most prevalent genes in the carbapenem resistant *A. baumannii*.\(^{[69]}\)

New Delhi metallo-β-lactamase (NDM) mediated carbapenem resistance has rapidly disseminated globally. Gautam V, 2014 evaluated presence of NDM in *Acinetobacter calcoaceticus*-*A. baumannii* complex from medical intensive care unit (ICU), neurosurgery ICU, respiratory ICU, cardio-thoracic vascular surgery ICU, emergency medical wards and out-patient departments (OPDs).\(^{[9]}\)

Multidrug resistant *A. baumannii* (MDR-AB) is fast becoming a global threat, having developed resistance to major classes of antibiotics and carbapenem-resistant isolates have increasingly been reported worldwide as a cause of nosocomial outbreaks. Approx 20 to 30% were resistant to β-lactam and β-lactamase inhibitor combinations.\(^{[148]}\) Some studies indicated that up to 80% were resistant to all aminoglycosides \(^{[149]}\) and ciprofloxacin.\(^{[19]}\) MDR Acinetobacter infection has also become a serious problem in the ICU located in several Asian countries.

Despite intensive efforts, nosocomial acquisition of MDR-AB is still a problem due to the organism's great ability to colonize human and environmental reservoirs. This prompted several microbiological and epidemiological studies leading to the formulation of interventions and infection control measures to prevent the impact of this organism on health care.\(^{[150]}\) MDR *Acinetobacter* infections have become increasingly difficult to treat because of the emergence of resistant to all drugs or commonly prescribed antimicrobial drug. These MDR strains are sometimes susceptible only to polymyxins (colistin and polymyxin B), a class of antimicrobial drugs that has not been in widespread use for several decades and is more toxic than most currently used antimicrobial drugs.
2.9. Treatment options for *A. baumannii* infections

The resistance patterns observed for *A. baumannii* in the clinical setting is leaving very few treatment options. Combination therapy is relied on in many centres to treat resistant strains of *A. baumannii*, where a significant synergy is observed in vitro. Combination therapy relies on an aminoglycoside with a 3rd generation cephalosporin, or colistin combined with rifampicin, ceftazidime or imipenem.[76,151] Combination therapy aims to prevent the emergence of resistance when using two different compounds, as well as provide coverage of a broad spectrum of pathogens in the case of mixed or unidentified infections.[152,85]

Most importantly, antibiotic selection must rely on the antibiotic susceptibility of individual.[85] Due to *A. baumannii* being resistant to many antibiotics, carbapenems are the ideal drugs in treating *A. baumannii* infections, but resistance is emerging very rapidly, that leaving few available treatment options.[153] Fluoroquinolones were used to treat sporadic cases of *A. baumannii*, but resistance is also now widespread in endemic strains.[76,152] In some countries sulbactam, a β-lactamase inhibitor, is used as a β-lactam drug by itself, and it has shown activities against some MDR strains.[154, 155]

The use of polymyxins and tigecycline has emerged in recent years to overcome carbapenem and multi-drug resistance, and have proven success in treating severe *A. baumannii* infections.[76,151] Resistance is still relatively rare for these compounds. Colistin (polymyxin E) has proven clinical efficacy in treating carbapenem and multi-drug resistant strains of *A. baumannii*, but the main issues of concern are nephrotoxicity and heteroresistance.[156,85] Interestingly, colistin is combined with rifampicin or a carbapenem in treating metallo-β-lactamase producing *A. baumannii*.[152] The strategy of antibiotic usage in neutropenic patients is that it should start immediately with the onset of febrile neutropaenia and first-line antibiotics should cover the most probable pathogens, based on the local epidemiology and patient history.[152] Patient history is particularly important because the carriage of MDR strains may last several weeks or months, and may be latent until neutropenia.

There is an urgent need for the development of novel antibacterial agents to combat the MDR organisms in the healthcare setting. However the pharmaceutical industry has reduced antibacterial drug development programs due to the low profit margins, and the
current ‘novel’ antibacterial are merely a modification of already existing
compounds.\textsuperscript{157,158}

There is an immense public health importance of antibiotic resistance, which
transcends national borders. The clock is ticking and the time is not too far when there
will be no drug available to treat serious infections, lest we enhance our infection
control practices and implement the use of strong antibiotic policies urgently. And the
analysis of susceptibility pattern will be useful in understanding the epidemiology of
this organism in different hospital setup, which will help in treating individual cases
and controlling the spread of resistant isolates to other individuals.

2.10. \textit{Acinetobacter} infections in India

There are wide ranges of NFGNB - \textit{Acinetobacter} infections are documented
from India. Gladstone \textit{et al.} from Vellore, India (2005), reported a prevalence of 14%
carbapenem-resistant \textit{Acinetobacter} spp., isolated from tracheal aspirates.\textsuperscript{121} Sinha M
2007 in Bangalore detected a high level of resistance in \textit{Acinetobacter} species to most
antibiotics tested. \textit{Acb} complex was the predominant species of \textit{Acinetobacter} isolated
and was also found to be more resistant than the other species. Cefepime and
cefotaxime along with the clavulanate disk in the DDST detected most of the ESBLs in
\textit{Acinetobacter}.\textsuperscript{129} Patwardhan RB in Pune studied nosocomial pathogen ICU and
isolate NFGNB, \textit{Acinetobacter} by API20 from Urinary tract predominantly.\textsuperscript{159} In
Rajasthan \textit{Acinetobacter baumanii} (83.2\%) was reported as the most common
nosocomial pathogen organism specially from ICUs.\textsuperscript{3}

Ravi et al, 2013 found \textit{Acinetobacter} species, one of commonest isolated
pathogen followed by E.coli and Pseudomonas in ICU-acquired infections.\textsuperscript{160} Sinha
N,2013 found predominant multidrug resistant \textit{Acinetobacter} from
Lucknow.\textsuperscript{142} Pradhan NP 2014 in Pune reported 9.6\% prevalence of nosocomial
infections, of which respiratory infections were the commonest (65.8\%), followed by
urinary infections dual infections (urinary plus respiratory). The most frequently
isolated microorganism causing respiratory infections was \textit{Acinetobacter} (40.4\%),in
which one fourth isolates of which were multidrug resistant. Overall ICU mortality was
19.9\%.\textsuperscript{93} In new Delhi  HAI prevalence was 8.78\% with highest in ICUs (33.3\%)
followed by paediatric wards (12.5\%) and surgical wards (10.3\%). Surgical procedures,
mechanical ventilation, urinary catheters, or intravascular devices were independent risk factors for HAI. The most common HAI category was urinary tract infection followed by respiratory tract infections, and surgical site infection. *Acinetobacter* was second most common pathogen. [14] Srirangaraj et al.2015 [16] in Pondicheri gave a case report on multidrug-resistant *Acinetobacter baumannii* from nosocomial urinary tract infection and describe clinical presentation, risk factors, laboratory evaluation and therapeutic outcome of a 70-year-old patient and conclude that *A.baumannii* is an important opportunistic agent of nosocomial UTI, especially in patients with longer hospitalization, antibiotic exposure, urinary catheterization and decreased immunity. High antimicrobial resistance and patient co-morbidities limit therapeutic choices.

Thus, for developing countries like India, surveillance of antimicrobial resistance is essential for preventing the emergence and transmission of multidrug-resistant pathogens in healthcare facilities.

This study, describes findings on the *A. baumannii* from intensive care units, particularly against matelio β-lactamase producers among strains isolated in Kanpur, Uttar Pradesh.