

Isolation of *Acinetobacter baumannii* and its Antimicrobial Resistance Pattern in an Intensive Care Unit (ICU) of a Tertiary Care Hospital

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ABSTRACT

Introduction: *Acinetobacter*, once considered as an opportunistic pathogen has recently emerged as an important nosocomial pathogen worldwide, mostly involving patients with impaired host defenses. Critically ill patients acquire an infection during their stay in Intensive Care Unit (ICU) and the frequency of these infections varies considerably in different populations and clinical setting. The purpose of this study was to know Antimicrobial sensitivity pattern of *A.baumannii* from various clinical samples collected from patients admitted in ICU at Adesh Institute of Medical Sciences and Research, Bathinda, Punjab over a period of one year six months from July 2014 to December 2015.

Material and methods: A total of 48 *A.baumannii* were obtained from 545 samples (8.8%). Antimicrobial susceptibility testing of all *A.baumannii* isolates was done using Kirby Bauer's disc diffusion technique as per recommendations of Clinical Laboratory Standards Institute (CLSI).

Results: Maximum number of *A.baumannii* were isolated from respiratory samples-tracheal aspirate, Endotracheal secretions and sputum (68.7%) followed by pus (12.5%), blood (8.3%), Intercostal drain tube and CSF (4.2%), urine (2.08%). All *A.baumannii* isolates were resistant to ceftazidime and cefepime. Higher level of resistance was also recorded for piperacillin-tazobactam (95.8%) gentamicin and amikacin (93.7%), ciprofloxacin (91.6%), cotrimoxazole (91.6%) ampicillin-sulbactam (75%). Resistance towards imipenem was recorded as 47.9% and meropenem as 58.2%. Minimum resistance was shown towards polymixin B (2.08%) and colistin (4.1%).

Conclusion: *A.baumannii* is emerging as a predominant healthcare associated multidrug resistant pathogen, especially in the ICU's. The findings of this study will help our clinicians to apply adequate antibiotics for treatment of patients admitted in ICU.

Keywords: *A.baumannii*, Intensive Care Unit (ICU), respiratory samples, antimicrobial resistance

genus but only three species i.e - *A.baumannii*, *A.calcoaceticus* and *A.lwoffii* appear to be of clinical importance. These species have been included under the term *A.calcoaceticus-A.baumannii* complex and are usually reported as *Acinetobacter*.^{6,7} Outbreaks of *Acinetobacter* are linked to contaminated respiratory equipment, intravascular access devices, bedding materials and transmission via hands of hospital personnel.⁸ An increase in antibiotic resistance among the isolates of organism during recent years, has made these infections difficult to treat.⁹ The resistance mechanisms of *Acinetobacter* are multiple. They include production of beta lactamases, alterations in cell wall channels and efflux pumps by which it becomes resistant to beta-lactam antibiotics, production of aminoglycoside modifying enzymes and mutations in genes *gyrA* and *parC* mediate resistance to aminoglycosides and quinolones respectively.¹⁰ The success of antimicrobial therapy depends upon the appropriateness of choice of antibiotics that should be used prior on basis of prior knowledge of susceptibility pattern of the agent. Therefore, the purpose of the study was to examine antimicrobial sensitivity pattern of *A.baumannii* isolates obtained from various clinical samples collected from patients admitted in ICU at Adesh Institute of Medical Sciences and Research, Bathinda over a period of one year six months, from July 2014 to December 2015

MATERIAL AND METHODS

A total of 545 clinical samples which included Tracheal aspirate, Endotracheal secretions, sputum, urine, blood, pus, intercostal drain tubes, CSF were collected from patients admitted in ICU of AIMSRS, Bathinda after getting clearance from the Institutional Ethics Committee of AIMSRS. The samples were collected from patients of all age groups, both sexes, who were critically ill and suspected for pneumonia, urinary tract infection, septicaemia, skin and soft tissue infection and meningitis. The samples were inoculated on Blood Agar and MacConkey Agar plates under strict aseptic conditions. Plates were incubated at 37C for 24-48 hrs. *A.baumannii* was identified and confirmed by Gram

INTRODUCTION

Members of Genus *Acinetobacter* have emerged as organisms of questionable pathogenicity and pan resistant nosocomial pathogens worldwide in past two or three decades; especially since 2005-2006.¹ Critically ill patients acquire an infection during their stay in an Intensive care unit (ICU) and the frequency of these infections varies considerably in different populations in clinical settings.^{2,3} The increased risk of infection is associated with severity of patient's illness, length of exposure to invasive and procedures, increased patient contact with healthcare personnel and length of stay in ICU.⁴ It can colonise the respiratory, urinary and gastrointestinal tract and wounds of patients and can cause infections in burn, trauma, mechanically ventilated and immune compromised patients as it shows a special predilection for ICU.⁵ There are many species in this

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staining as Gram negative cocci or coccobacilli in pairs, non-motile, oxidase negative, Alkaline/Alkaline (K/K) reaction in Triple sugar Iron (TSI) slant, catalase positive, Indole negative,

Samples	Total number processed	Positive for <i>A.baumannii</i>
Tracheal aspirate	98	16
Endotracheal secretions	83	13
Sputum	23	4
Urine	162	1
Blood	146	4
Pus	17	6
Intercostal drain tubes	6	2
CSF	10	2
Total	545	48

Table-1: *Acinetobacter baumannii* isolates from various samples

Specimen	<i>A.baumannii</i> isolated (%)
Tracheal aspirate	16 (33.3%)
Endotracheal secretions	13 (27.08%)
Sputum	4 (8.3%)
Urine	1 (2.08%)
Blood	4 (8.3%)
Pus	6 (12.5%)
Intercostal drain tubes	2 (4.2%)
CSF	2 (4.2%)

Table-2: Number of *A.baumannii* isolated from various samples

Antimicrobial agent	Sensitive n (%)	Resistant n (%)
Ceftazidime	0 (0%)	48 (100%)
Cefepime	0 (0%)	48 (100%)
Ampicillin-sulbactam	12 (25%)	36 (75%)
Imipenem	25 (52.1%)	23 (47.9%)
Meropenem	21.8 (43.8%)	27 (58.2%)
Piperacillin-tazobactam	2 (4.2%)	46 (95.8%)
Cotrimoxazole	4 (8.4%)	44 (91.6%)
Ciprofloxacin	4 (8.4%)	44 (91.6%)
Gentamicin	3 (6.3%)	45 (93.7%)
Amikacin	3 (6.3%)	45 (93.7%)
PolymixinB	47 (97.9%)	1 (2.1%)
Colistin	46 (95.8%)	2 (4.2%)

Table-3: Sensitivity pattern of *A.baumannii* to different antimicrobial agents (N=48)

Name of Antibiotic	Tracheal aspirate (16)	ET secretion (13)	Sputum (4)	Urine (1)	Blood (4)	Pus (6)	ICD tube (2)	CSF (2)
Ceftazidime	16 (100%)	13 (100%)	4 (100%)	1 (100%)	4 (100%)	6 (100%)	2 (100%)	2 (100%)
Cefepime	16 (100%)	13 (100%)	4 (100%)	1 (100%)	4 (100%)	6 (100%)	2 (100%)	2 (100%)
Ampicillin-sulbactam	12 (75%)	8 (61.5%)	4 (100%)	1 (100%)	2 (50%)	5 (83.3%)	2 (100%)	2 (100%)
Imipenem	7 (43.7%)	4 (30.7%)	3 (75%)	0 (0%)	2 (50%)	5 (83.3%)	2 (100%)	2 (100%)
Meropenem	10 (62.5%)	6 (46.1%)	3 (75%)	0 (0%)	2 (50%)	2 (33.3%)	2 (100%)	2 (100%)
Piperacillin-tazobactam	15 (93.7%)	13 (100%)	4 (100%)	0 (0%)	4 (100%)	6 (100%)	2 (100%)	2 (100%)
Cotrimoxazole	16 (100%)	11 (84.6%)	4 (100%)	1 (100%)	2 (50%)	6 (100%)	2 (100%)	2 (100%)
Ciprofloxacin	14 (87.5%)	12 (92.3%)	4 (100%)	0 (0%)	4 (100%)	6 (100%)	2 (100%)	2 (100%)
Gentamicin	16 (100%)	12 (92.3%)	4 (100%)	1 (100%)	2 (50%)	6 (100%)	2 (100%)	2 (100%)
Amikacin	16 (100%)	12 (92.3%)	4 (100%)	1 (100%)	2 (50%)	6 (100%)	2 (100%)	2 (100%)
Polymixin B	1 (6.25%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Colistin	2 (12.5%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)

Table-4: Antibiotic resistance pattern of *A.baumannii* isolated from different sites of infection

Citrate utilization test positive, Nitrate reductase negative, urease test negative. It showed Oxidative –Fermentative (O/F) test –oxidative and growth at 44 C.^{11,12}

Antimicrobial susceptibility testing of all *A.baumannii* isolates was done using Kirby Bauer disc diffusion technique as per recommendations of Clinical Laboratory Standards (CLSI).^{13,14} Antimicrobial discs used for sensitivity testing were- ceftazidime (30 µg), cefepime (30 µg), piperacillin-tazobactam (100µg)/10 µg), Ampicillin-sulbactam (10 µg)/10 µg), Imipenem (10 µg), Meropenem (10 µg), Gentamicin (10 µg), Amikacin (30 µg), Cotrimoxazole (25 µg), Ciprofloxacin (5 µg), Norfloxacin (30 µg), PolymixinB (300 units) and Colistin. (10 µg). All dehydrated media and antimicrobial discs were procured from Hi Media Labs, Mumbai, India.

STATISTICAL ANALYSIS

Statistical analysis was done by descriptive statistics using simple ratio and percentages method. Microsoft office 2007 was used to generate Tables.

RESULTS

A total of 48 *A.baumannii* isolates were obtained from total 545 samples collected from ICU patients (Table-1).

Maximum number of *A.baumannii* were isolated from respiratory samples-tracheal aspirate, ET secretions and sputum (68.7%) followed by pus (12.5%), blood (8.3%), Intercostal drain tube and CSF (4.2%), urine (2.08%) (Table-2).

All *A.baumannii* isolates were resistant to ceftazidime and cefepime. Higher level of resistance was also recorded for piperacillin-tazobactam (95.8%), gentamicin and amikacin (93.7%), ciprofloxacin (91.6%), cotrimoxazole (91.6%) ampicillin-sulbactam (75%). Resistance towards imipenem was recorded as 47.9% and meropenem as 58.2%. Minimum resistance was shown towards polymixin B (2.08%) and colistin (4.1%) (Table-3) (Table-4).

DISCUSSION

In the present study, 8.8% (48/545) *A.baumannii* isolates were obtained from different ICU samples. In India, *A.baumannii* is reported to cause about 13.2% of nosocomial infections in ICU patients.¹⁵ In our study, respiratory samples showed *A.baumannii* (68.75%) as compared to non-respiratory samples. This study is in concordance with a study by Jaggi et al who

reported isolation rate of *A.baumannii* in respiratory samples as 59.6%.⁵

Very low isolation rate was reported from urine samples (2.08%). The results are almost similar to Jaggi et al⁵ and Nahar et al⁴ who reported it as 2.9% and 3.1% respectively. This shows that *A.baumannii* shows relatively low prevalence in causing UTI. Siau et al¹⁶ reported that respiratory tract was the most common site from where *A.baumannii* was isolated in ICU patients. Villers et al¹⁷ have also reported a predominance of *A.baumannii* in tracheobronchial secretions from 24.8% to 48.8% and Suri et al¹⁸ as 45.6% in their studies respectively. This study showed *A.baumannii* was extremely resistant to all routinely used antibiotics in the ICU. Many Indian studies have reported high level of resistance in Acinetobacters. Most isolates were from critical care setting and source was most often respiratory samples.¹⁹ In a study by Nahar et al 100% resistance was recorded towards amoxicillin, ceftriaxone, cefuroxime and gentamicin. Higher level of resistance was recorded was amikacin (68.4%) and Imipenem (66.7%) but lower level of resistance was shown in colistin (10.5%).⁴ Rahbar et al also reported high level of resistance towards Piperacillin-tazobactam (90.9%), ceftriaxone (90.9%), ceftazidime (84.1%) and ciprofloxacin (90.9%).²⁰ In another study by Shakibaie et al, they found that many isolates of Acinetobacter were resistant to almost all antibiotics routinely used in the ICU of their hospital.²¹ The high resistance pattern seen in our isolates may be related to selective pressure of extensive usage of third generation cephalosporins. It has also been observed that Acinetobacter can develop resistance when the patient is on treatment. In case of pan drug resistant Acinetobacter infections, alternative antibiotics available are colistin, polymixin B and tigecycline.¹⁰

CONCLUSION

The high proportion of antibiotic use in our ICU's might explain the high resistance observed in *A.baumannii*. Rational use of antibiotics is necessary to prevent microbial resistance catastrophe. The major mode of transmission from patient to patient is the contaminated hands of health care workers (HCW's). Therefore, improving hand hygiene compliance among HCW's and standard precautions may be adequate to control multidrug resistant ICU bugs in endemic settings. The findings of this study will help our clinicians to apply adequate antibiotics for treatment of patients admitted in ICU.

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