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RESEARCH ARTICLE

EPIDEMIOLOGICAL STUDY OF *ACINETOBACTER BAUMANNII* AND ITS RESISTANCE PATTERN IN CLINICAL ISOLATES FROM A PRIVATE HOSPITAL IN KOLKATA, EASTERN INDIA

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ABSTRACT

Introduction: This retrospective study aims to determine the prevalence and resistance pattern of *Acinetobacter baumannii* in clinical isolates from a private hospital in Kolkata, Eastern India.
Methods: The study was conducted over a 6 months period from October 2017 to March 2018 where all the clinical isolates of *A. baumannii* and their resistance pattern obtained by an automated identification system (Vitek 2 Compact).
Results: Out of 2149 clinical samples, a total of 21 strains of *A. baumannii* were isolated. Majority of isolates (62%) were obtained from ICU followed by IPD (28.5%) and OPD (9.5%). Majority of samples were from tracheal aspirates (43%) and urine (28.5%). Patients of more than 61 years of age are affected most. Out of twenty one strains four were found as Multi Drug Resistant (MDR) and fourteen were Extensive Drug Resistant (XDR) strains. Predominant MDR strains were from tracheal aspirates. 90% strains were resistant to Ciprofloxacin and Levofloxacin. The strains were sensitive to only Colistin and Polymixin-B.
Conclusion: It may be concluded from the study that XDR strains are more predominant than MDR strains of *A. baumannii* among the clinical isolates of this private hospital. This epidemiological study will help to conduct better infection control policy and resistance pattern guide for the treatment of patients in this region.

Key words: *Acinetobacter baumannii,* Prevalence, Resistance pattern.

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INTRODUCTION

Acinetobacter is a non-fermenting gram negative coccobacillus with a high capacity to colonize the human body and the environmental reservoirs (Obediat *et al.,* 2014). *Acinetobacter baumannii* colonizes healthy humans transiently at a low density on the warm and moist skin of axilla, groin, between toes, throat, nares and intestinal tract but it generally does not cause infection (Young *et al.,* 2007). It can infect a patient's respiratory tract, blood, soft tissues, urinary tract and central nervous system, causing a variety of infections, including pneumonia, endocarditis, meningitis, wound infections and urinary tract infections (Maragakis and Perl, 2008). Although *A. baumannii* has classically been recognized as a hospital-acquired pathogen, community-acquired *Acinetobacter* infections have been reported in the literature, mostly occurring in countries with tropical climates (Lowman *et al.,* 2008). It has been identified as an ESKAPE pathogen (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumonia, Acinetobacter baumannii, Pseudomonas aeruginosa* and *Enterobacter species)*; a group of pathogens with a high rate of antibiotic resistance that are responsible for the majority of nosocomial infections (Drummond, 2013). Reports have suggested that the community acquired *Acinetobacter* pathogens are relatively susceptible to antibiotics and the more resistant subtypes have been found almost exclusively in hospitals and intensive care units (Dijkshoorn *et al.,* 2007). It is very resistant and aggressive organism that infects patients with weakened defence like ICU patients and those with invasive devices (Perez *et al.,* 2007). The rising incidence of Multi Drug Resistant (MDR) *A. baumannii* may be attributed to lack of infection-control measures and high selective pressure of commonly used antimicrobial agents (Mutnick *et al.,* 2004). The most active agents against the MDR *A. baumannii* are polymixins – polymixin B and polymixin E(Colistin) and tigecycline (Urban *et al.,* 2003). The present study includes two major objectives. The first is to describe the prevalence of *A. baumannii* in a public hospital in Eastern India. The second objective is to describe the degree of antibiotic resistance in the clinical samples.

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MATERIALS AND METHODS

This retrospective study was carried out over a period of 6 months from October 2017 to March 2018 in the Department of Microbiology, in a private hospital located in Kolkata, Eastern India. Samples were collected and processed during the course of routine diagnostic work of the clinical samples from patients in ICU, Wards (IPD) and Out Patient Department (OPD) of the hospital. The identification of pathogens using routine microbiological techniques were analyzed and *A. baumannii* isolates were picked up for further studies. The specimens studied included urine, respiratory samples (sputum, endo-tracheal aspirate and bronchoalveolar lavage), blood, pus and body fluids (pleural fluid, cerebrospinal fluid etc). The specimens received in the laboratory were inoculated on 5% Blood agar and Mac Conkey Agar, and incubated overnight aerobically at 37^0C. All blood samples were collected aseptically and introduced into BacT / ALERT FA PLUS bottles and incubated in BacT / ALERT 3D system (Biomerieux). The BacT / ALERT Microbial Detection System utilizes a colorimetric sensor and reflected light to monitor the presence and production of carbon dioxide (CO_2) dissolved in the culture medium. CO_2 is produced when the organisms metabolize the substrates in the culture medium. With the production of CO_2, the colour of the gas-permeable sensor installed in the bottom of each culture bottle changes from blue-green to yellow.

The lighter colour results in an increase of reflectance units monitored by the system. Bottle reflectance is monitored and recorded by the instrument every 10 minutes. The positive blood culture samples were subcultured in blood agar and MacConkey agar respectively. The pure colonies obtained from the subculture were subjected to Gram staining followed by oxidase test. *A. baumannii* isolates were initially identified by colonial morphology, Gram staining, growth at 37^0C and a negative oxidase test. The pure growth of NLF oxidase negative Gram negative bacilli from all samples were utilized for identification using Biomerieux GN – ID cards in VITEK 2 machine. The VITEK- 2 (Biomerieux) compact systems is a fully automated growth based technology that performs bacterial / yeast identification by biochemical analysis using colorimetric method. The Gram negative bacterium identification card uses 47 biochemical tests for identification of the organism. The susceptibility tests were done by using Biomerieux AST – N280 cards in VITEK- 2 machine. The antibiotics against which susceptibility were checked by MIC include amikacin (30mcg), cefepime (30mcg), cefoperazone/sulbactam (75/30mcg), ciprofloxacin (5mcg), levofloxacin (5mcg), colistin (10mcg), polymixin-B (300 Units), gentamicin (10mcg), neitilmicin (10mcg), imipenem (10mcg), meropenem (10mcg), doripenem (10mcg), piperacillin/ tazobactam (100/10mcg) and trimethoprim/ sulfamethoxazole (25mcg).

RESULTS

In our study 2149 clinical samples (urine-1506, sputum-177, blood-247 and others; pus, wound swab, tracheal aspirates-225) were collected over a period of 6 months from October 2017 to March 2018 from ICU, In-patient wards and OPD of a private hospital. Gram negative isolates were 230 (10.7%), of which 21 isolates were *A. baumannii* (9.1%). Table1 shows that the majority of *A. baumannii* was found from tracheal aspirates (43%), followed by urine (28.5%), sputum (19%), and wound (9.5%).

Table 1. Prevalence of *A. baumannii* in clinical isolates (Total number of *A. baumannii* [n] = 21)

Sample	Number	Percentage (%)
Tracheal aspirates	9	43%
Urine	6	28.5%
Sputum	4	19.0%
Wound/Pus	2	9.5%

Among 21 *A. baumannii* isolates, 13 were obtained from ICU patients and the rest from from IPD and OPD (Table 2). So majority of the infection comes from ICU patients.

Table 2. Prevalence of *A. baumannii* in different wards

Ward	Isolates	Percentage (%)
ICU	13	62%
IPD	6	28.5%
OPD	2	9.5%

In this study majority of *A. baumannii* were isolated from >61 age group (57.14%) followed by (41-60) age group (33.33%).

Table 3.Age wise distribution of *A. baumannii* (n = 21)

Age in Years	No. of Isolates	Percentage (%)
0 to 15	0	0
16 to 40	2	(9.53%)
41 to 60	7	(33.33%)
> 61	12	(57.14%)
T O T A L	21	(100%)

In this study most of *A. baumannii* were highly resistant to the tested antibiotics. The resistance pattern shows 90% resistance to levofloxacin and ciprofloxacin followed by cefotaxime and cefepime (85.7%), ceftriaxone (71.4%), piperacillin/ tazobactam (71.4%), imipenem, meropenem and doripenem (66.6%), amikacin, gentamicin and neitilmicin (61.9%).The most effective antibiotic was colistin and polymixin-B which shows 100% sensitive (Table 4).

Table 4. Antibiotic resistance patterns of *A. baumannii* isolates (n = 21)

Antibiotic Group	Antibiotic Agent	Resistance (%)
enicillin	Piperacillin/Tazobactam (PIT)	15(71.4%)
	Trimethoprim/Sulfamethoxazole (COT)	12 (57.1%)
Cephalosporin	Cefoperazone/Sulbactam (CS)	11 (52.4%)
	Cefotaxime (CTX)	18 (85.7%)
	Cefepime (CPM)	18 (85.7%)
	Ceftriaxone (CX)	15 (71.4%)
Carbapenem	Imipenem (IPM)	14 (66.6%)
	Meropenem (MRP)	14 (66.6%)
	Doripenem (DOR)	14 (66.6%)
Aminoglycosides	Amikacin (AK)	13 (61.9%)
	Gentamicin (GEN)	13 (61.9%)
	Neitilmicin (NT)	13 (61.9%)
Fluoroquinolones	Ciprofloxacin (CIP)	19 (90.4%)
	Levofloxacin (LE)	19 (90.4%)
Polymixin	Colistin	0 (0%)
	Polymixin-B	0 (0%)

Table 5 . Multi Drug (MDR) and Extensive Drug (XDR) Resistant strains among *A. baumannii* (n = 21

Susceptibility pattern	No. of Isolates	Percentage (%)
Sensitive	3	14%
XDR	14	67%
MDR	4	19%

DISCUSSION

In this study the prevalence of *A. baumannii* was 9.1% of the total Gram negative isolates. This corresponds to similar study carried out by H Siau et al (H Siau *et al.,* 1996) where *A. baumannii* isolates were 11% of the total Gram negative isolates. Among the source of isolates, most of *A. baumannii* was isolated from tracheal aspirates followed by urine, sputum and pus which is similar to the result reported by Shanthi and Sekar (2009) where most of the isolates of *A. baumannii* were obtained from the respiratory tract (41.8%) followed by urinary tract (25.5%), wound (20%) and blood (12.7%). According to the age group the most affected are above 61year age group. These results are similar to those of many authors (Ozgur *et al.,* 2014, Punpanich *et al.,* 2012).The old age of patients was recognized as an independent risk factor of the acquisition of *A.baumannii* infection (Turkoglu *et al.,* 2011). In this study *A. baumannii* was mostly isolated from ICU (62%) which is identical with most of the global studies (Munoz *et al.,* 2008, Wiks *et al.,* 2006). They concluded that *A. baumannii* is the ICU Superbug. This statement reveals that a lot of risk factors are associated with *A. baumannii* infection. ICU may act as a potential environmental reservoir for its infection, cross transmission among immunocompromised patients and the patients having multiple wounds. The resistance profile of the *A. baumannii* isolates showed that 19% were MDR which corresponds to the resistance to three groups of antibiotics including aminoglycosides, fluoroquinolones and cephalosporins. 67% were XDR which corresponds to the resistance to most of the antibiotics. In our study *A. baumannii* shows 100% sensitive to colistin which is correlated with other studies also (Mushtaq *et al.,* 2013). Several authors report that colistin may be the only option for treatment of serious *Acinetobacter* infections where this organism is strongly suspected to be resistant to other antibiotics (Shareek *et al.,* 2012).

Conclusion

An increase in the occurrence of multiple antibiotic resistant bacteria population has severe impact on human health. Multi drug resistant human pathogens are serious threat worldwide. Indiscriminate use of broad spectrum antibiotics and their improper disposal in public hospitals are the major cause for this resistance. A strict control of the hospital environment, hand hygiene and optimizing the use of antibiotics is recommended in order to reduce the MDR and XDR frequency. *A. baumannii* being mostly a hospital acquired infection needs a thorough epidemiological investigation to identify its antibiotic resistance pattern in a given area. A good knowledge of their resistance pattern will help to determine the use of proper drug in the public hospitals.

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