

Prevalence of metallo- β -lactamase-producing (MBL) *Acinetobacter* species in a tertiary care hospital

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Received: March 2013, Accepted: December 2013.

ABSTRACT

Background and Objectives: *Acinetobacter* causes a wide variety of illness in debilitated and hospitalized patients. Carbapenem resistance in *Acinetobacter* is an emerging problem and is a cause of concern as many nosocomial infections with *Acinetobacter* are resistant to most other antibiotics. The present study was aimed to study metallo- β -lactamase (MBL) production in *Acinetobacter* species.

Material and Methods: During one year prospective study, all isolates of *Acinetobacter* obtained from various clinical samples like respiratory, pus, blood and others were included. Antimicrobial susceptibility testing was done by standard Kirby Bauer disk diffusion method. Metallo β -lactamase (MBL) detection was done by imipenem-EDTA combined disk method.

Results: Among 1017 isolates, 964 were *A. baumannii*, 48 were *A. lwoffii* and 5 were *A. hemolyticus*. Out of these, majority of the isolates were obtained from respiratory samples, followed by pus. *A. baumannii* showed high level of resistance to cephalosporins, cotrimoxazole and piperacillin. *A. lwoffii* and *A. hemolyticus* showed lesser resistance to all antibiotics. Imipenem resistance was observed in 389 (40.3 %) isolates of *A. baumannii* and MBL activity was seen in 80.3% of isolates. MBL positive isolates of *A. baumannii* showed higher resistance as compared to MBL negative isolates.

Conclusion: This study demonstrated that multidrug resistant strains of *Acinetobacter* are common in tertiary care hospitals. Unwarranted and unrestricted usage of antibiotics is associated with emergence of resistance in nosocomial pathogens. Regular monitoring and documentation of carbapenem resistant is crucial in developing strategies to control infection due to these bacteria.

Keywords: Carbapenem resistance, metallo β -lactamase, *A. baumannii*

INTRODUCTION

Acinetobacter causes a wide variety of illness in debilitated and hospitalized patients. These bacteria survive for long period in hospital environment and thereby the opportunities for cross infection between patients are enhanced (1). *Acinetobacter* species play a significant role in the colonization and infection of patients admitted in hospitals. It has been implicated

in variety of nosocomial infections. *Acinetobacter baumannii* is intrinsically less susceptible to antibiotics than *Enterobacteriaceae*; moreover, it has propensity to acquire resistance. Because of frequent resistance to aminoglycosides, fluoroquinolones, ureidopenicillins and third -generation cephalosporins, carbapenems are important agents for managing *Acinetobacter* infections (2).

The resistance of *Acinetobacter baumannii* to carbapenem is now a major worldwide issue (3-6). The carbapenems are β -lactam antimicrobial agent with an exceptionally broad spectrum of activity. Carbapenem resistance in *Acinetobacter* is an emerging problem and is a cause of concern as many nosocomial *Acinetobacter* are resistant to

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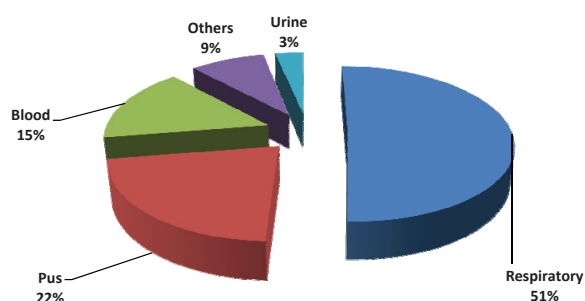


Fig. 1. Distribution of *Acinetobacter* spp. in various samples.

most other antibiotics. Carbapenem resistance in *Acinetobacter* is attributed to various causes such as reduced expression of outer membrane proteins and carbapenemases β -lactamases (7). Some carbapenem resistant isolates produce either metallo-beta-lactamases (Ambler class B β -lactamases) or more commonly OXA type enzymes (Ambler class D β -lactamases or oxacillinases) having weak activity against carbapenems (8). Metallo-beta-lactamase (MBL) producing *Acinetobacter baumannii* has become a growing therapeutic concern worldwide. The rapid detection of MBL positive isolates is necessary to control infection and to prevent their dissemination. The aim of this study was to determine the prevalence of MBL among carbapenem resistant strains of *Acinetobacter* species in our hospital.

MATERIALS AND METHODS

The one year prospective study was conducted in the Department of Microbiology. Various clinical samples like respiratory, pus, blood, urine and others were processed according to the standard procedures. The isolates were identified as non fermenting Gram negative bacilli (NFGNB) on the basis of

colony characteristics, Gram's staining, motility test, oxidase and alkaline reaction on Triple Sugar Iron agar. All oxidase negative and nonmotile NFGNB isolates were further identified by various tests like OF-glucose, arginine dihydrolase, growth at 44°C, citrate utilization and haemolysis on blood agar (9). The antimicrobial susceptibility testing was done by Kirby Bauer disc diffusion method using gentamicin (10 μ g), amikacin (30 μ g), netilmicin (30 μ g), cotrimoxazole (1.25/23.75 μ g), ciprofloxacin (5 μ g), ceftazidime (30 μ g), cefepime (30 μ g), piperacillin (100 μ g), piperacillin/tazobactam (100/10 μ g), and imipenem (10 μ g) as per CLSI Guidelines (10). Imipenem resistant isolates were selected for the detection of MBL production by Imipenem-EDTA combined disc test (6).

RESULTS

Among 1017 isolates, 515 (50.6%) isolates were from respiratory samples, 222 (21.8%) from pus, 159 (15.6%) from blood, 88 (8.6%) from other clinical samples and 33 (3.2%) from urine samples (Fig. 1).

Out of total isolates, 964 (94.7%) were identified as *A. baumannii*, 48 (4.7%) *A. lwoffii* and 5 (0.4%) *A. hemolyticus*. *A. baumannii* showed high level of resistance to cephalosporins, cotrimoxazole and piperacillin. Majority of *A. baumannii* (71%) were sensitive to piperacillin-tazobactam. Among aminoglycosides, netilmicin showed lesser resistance (46.9%) than amikacin (64.9%) and gentamicin (88.1%). *A. lwoffii* and *A. hemolyticus* showed lesser resistance to all antibiotics as compared to *A. baumannii*. All isolates of *A. lwoffii* and *A. hemolyticus* were sensitive to imipenem whereas 389 (40.3 %) isolates of *A. baumannii* were found to be

Table 1. Comparison of Antibiotic resistance profile of MBL+ve and MBL-ve *A. baumannii*.

Antibiotic	MBL +ve <i>A. baumannii</i> (313)	MBL -ve <i>A. baumannii</i> (76)
Amikacin	273 (87.22%)	48 (63.16%)
Gentamicin	295 (94.25%)	63 (82.89%)
Netilmicin	209 (66.77%)	41 (53.95%)
Ciprofloxacin	307 (98.08%)	61 (80.26%)
Ceftazidime	309 (98.72%)	61 (80.26%)
Cefepime	304 (97.12%)	60 (78.95%)
Piperacillin	285 (91.05%)	62 (81.58%)
Piperacillin-Tazobactam	179 (57.19%)	32 (42.10%)
Cotrimoxazole	305 (97.44%)	75 (97.40%)

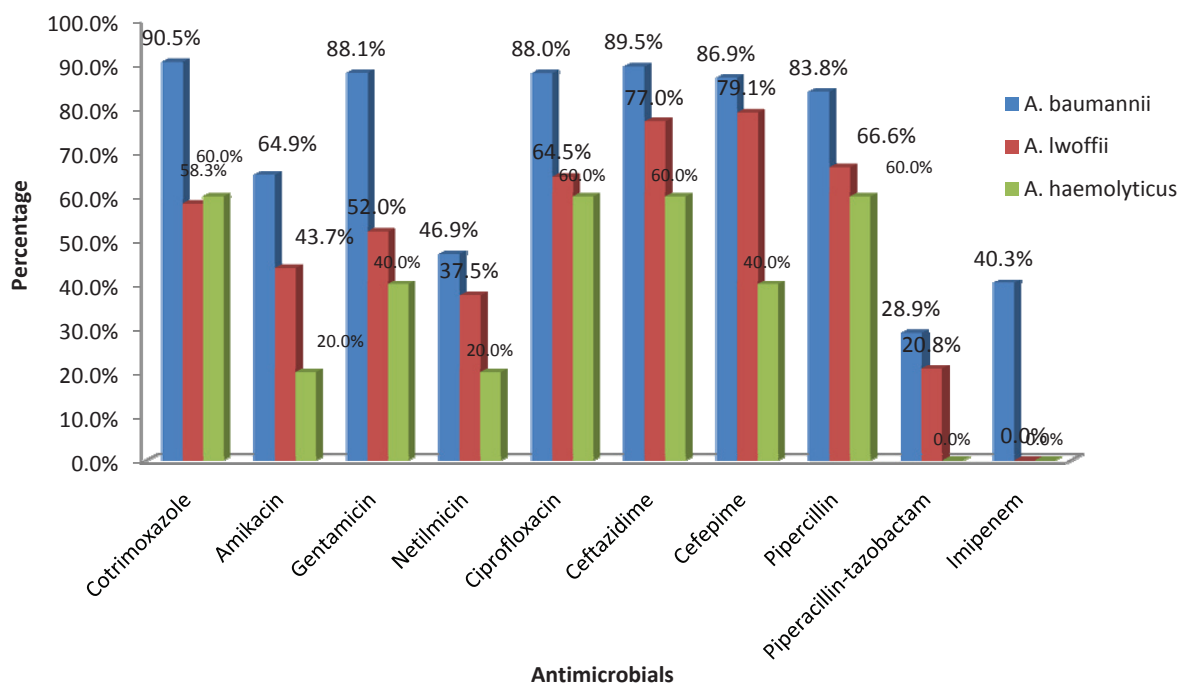


Fig. 2. Antimicrobial resistance profile of *Acinetobacter* spp. (n = 1017).

imipenem resistant (Fig. 2).

MBL activity was seen in 313 (80.3%) of imipenem resistant *A. baumannii* isolates. MBL positive isolates of *A. baumannii* were showing significantly higher resistance to all antimicrobials tested except cotrimoxazole as compared to MBL negative isolates and it was statistically significant ($P < 0.05$) (Table 1).

DISCUSSION

In our study, the most common *Acinetobacter* species identified from various samples was *A. baumannii* followed by *A. lwoffii* and *A. haemolyticus*. Similar results had been reported in literature (11, 12). Most of the nosocomial infections are caused by *A. baumannii*, whereas other species are considered less virulent. *A. baumannii* isolates were resistant to most of the antibiotics used. Resistance to cephalosporins was observed in $> 80\%$ isolates and among aminoglycosides, resistance to amikacin was seen in about 65% and resistance to gentamicin was seen in about 89% of isolates similar to the reports in literature (13-15). Netilmicin showed higher sensitivity as compared to gentamicin and amikacin in *Acinetobacter* spp.

All *Acinetobacter* isolates were sensitive to imipenem in a study by Malini *et al.* (13). Resistance

to imipenem was observed in 40.3% of *A. baumannii* isolates in our study whereas 14.2%, (16) 23% (17) and 57.4% (18) of *Acinetobacter* spp were resistant to imipenem as reported in literature. Among imipenem resistant isolates, 80.3% of *A. baumannii* showed MBL production whereas higher (96.6%) MBL production (17) and lower MBL production 7.5%, 14.8%, 49%, 56%, 74% in *Acinetobacter* spp. (5, 19, 20, 4, 18) as compared to our study was reported by various authors. In our study, statistically significant difference was found in the resistance profile of MBL positive and negative isolates for cephalosporins, aminoglycosides, quinolones, piperacillin, piperacillin-tazobactam which is consistent with the other studies (21, 22). This study demonstrated that multidrug resistant *Acinetobacter* are common in hospitals. Unwarranted and unrestricted usage of antibiotics is associated with emergence of resistance in common nosocomial pathogens like *Acinetobacter* species. Use of third generation cephalosporins has been shown to increase carbapenem resistance in *Acinetobacter* strains (2). Production of MBL has tremendous therapeutic consequences since these organisms also carry multidrug resistance genes and the only viable option remains the potentially toxic polymyxin B and colistin (23).

In conclusion, the present study revealed high

proportion of MBL producing *Acinetobacter* isolates. Respiratory, pus and blood samples collected from patients were found to be the main sources of MBL producing isolates. Early detection and infection control practices are the best defenses against these organisms; therefore, systematic surveillance to detect MBL producers is necessary. It is important to follow antibiotic restriction policies to avoid excessive use of carbapenem and other broad spectrum antibiotics.

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