CARBAPENEM RESISTANT ACINETOBACTER-A MAJOR PATHOGEN IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Objective: To determine the frequency of carbapenem resistant Acinetobacter in the Military Hospital Rawalpindi, Pakistan.

Study Design: Descriptive study.

Place and Duration of Study: Department of Microbiology, Army Medical College, from Oct, 2012 to Feb, 2013.

Material and Methods: Clinical specimens like naso-bronchial lavage, blood, pus, sputum and catheter tips were inoculated on blood agar and Mac Conkey agar while the urine samples were inoculated on Cystine Lactose Electrolyte Deficient (CLED) agar. Acinetobacter spp. isolated, were later subjected to antimicrobial susceptibility testing using the modified Kirby-Bauer disc diffusion method on Mueller Hinton agar as per Clinical Laboratory and Standards Institute (CLSI) guidelines.

Results: Out of a total of 85 Acinetobacter spp. 62 isolates were found to be carbapenem resistant. They were also found to be 100% resistant to ciprofloxacin and ceftriaxone thus becoming multidrug resistant followed by tazobactam piperacillin (98%) and trimethoprim sulphamethoxazole (92%). Minimum resistance was seen against tigecycline being 21%.

Conclusion: It is concluded from our study that there is a high frequency (72.94%) of resistance to carbapenems in Acinetobacter spp. in our setup which is associated with increased morbidity and mortality due to limited treatment options.

Keywords: Acinetobacter, Carbapenems, Carbapenem resistant Acinetobacter.

INTRODUCTION

Acinetobacter is a ubiquitous gram negative non-fermenting cocobacillus belonging to the family Moraxellaceae[1]. The first Acinetobacter was isolated in 1911 from a soil sample by MW Beijerinkas reported by Kuo et al in the year 2004[2]. Initially they were thought to be non-virulent saprophytes[3]. The injudicious use of antibiotics lead to the development of the first clinical isolate of carbapenem resistant. A baumannii isolated in 1991[2]. Associated with strong epidemic potential it is among the leading opportunistic pathogens which are worsening the conditions associated with highly increased morbidity and mortality in the current health care system, particularly in the intensive care settings[3,4]. In hospitals especially in the ICUs, prolonged stay, surgical procedures, parenteral nutrition, urinary and intravascular catheters are the major risk factors for the acquisition of infection with Acinetobacter spp. whereas the community-acquired Acinetobacter infections are associated more with renal failure, chronic lung disease, diabetes, alcoholism and smoking[5].

Acinetobacter can infect any system of the body especially in the immunosuppressed persons and the commonly seen infections include ventilator-associated pneumonias and bacteremia[6]. One of the major concerns regarding Acinetobacter is its intrinsic resistance to many antimicrobial agents[7]. In the past gentamicin, fluoroquinolones and cephalosporins showed good results against Acinetobacter spp. but nowadays the resistance against these antibiotics has increased thus making it multi-drug resistant and further decreasing the treatment options[8].

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Although structurally different from penicillins and cephalosporins, carbapenems are beta lactam antibiotics with excellent bactericidal activity. They are broad spectrum antimicrobial agents effective against gram positive and gram negative organisms and also the anaerobes. The development of resistance to carbapenems therefore may lead to increased morbidity and mortality[5]. The increase in resistance against carbapenems is associated with the development of the enzyme beta lacatamase[9-11]. Carbapenemase production, penicillin-binding protein modification, and/or porin loss have been reported as mechanisms of resistance, with carbapenemase production increasingly viewed as the major mechanism[12].

Resistance against carbapenems in itself is sufficient to define an isolate of Acinetobacter as highly resistant[13]. The objective of this study was to establish the frequency of Carbapenem resistant Acinetobacter spp. in the Military Hospital, Rawalpindi.

MATERIAL AND METHODS

This descriptive study was carried out in the Department of Microbiology, Army Medical College, National University of Sciences and Technology Islamabad, affiliated with an 1100 bedded tertiary care hospital, from October 2012 to February 2013.

All clinical isolates of carbapenem resistant Acinetobacter from any routine sample of admitted patients. Duplicate samples taken from the same patient during the same episode of illness and carbapenem susceptible isolates were not included.

Received clinical specimens like blood, pus, ascitic fluid, tracheal aspirate, naso-bronchial lavage, sputum and catheter tips were cultured on Blood and MacConkey agar, while the urine samples on Cystine Lactose Electrolyte Deficient (CLED) agar code and company of these.

Later the isolates were identified by a negative gram reaction, positive catalase test, a negative oxidase test and by being nonmotile. The identification of the organisms was carried out using conventional sugar fermentation tests / routine biochemical tests. API20NE (Biomerieux) was used to confirm A. baumannii. Using the modified Kirby-Bauer disc diffusion method bacterial suspensions of all the Acinetobacter isolates equivalent to 0.5 McFarland turbidity standard were prepared and inoculated on Mueller Hinton agar plates. Antibiotic discs of ampicillin (10μg), ceftriaxone (30μg), trimethoprim sulphamethoxazole (1.25μg/23.75μg), amikacin (30μg), meropenem (10μg), gentamicin (10μg), ciprofloxacin (5μg), tigecycline (15μg) and sulbactam cefoperazone (75μg) (Oxoid, UK) were applied followed by incubation at 35°C for 24 hrs. The results were interpreted following Clinical and Laboratory Standards Institute (CLSI) guidelines[14]. The isolates were considered resistant to carbapenems (meropenem / imipenem) if the zones of inhibition around the discs were found to be ≤13mm and susceptible if ≥16 mm. Extended spectrum beta lactamase (ESBL) production was checked while carrying out the antimicrobial susceptibility of the isolates using Jarlier et al method (Jarlier et al, 1998)[15]. American Type Culture Collection A. baumannii (ATCC) 19606 was used as the quality control strain. Data had been analyzed using Statistical Package for Social Sciences version 19. Qualitative variables for example

Table: Referral units of the received carbapenem resistant acinetobacter isolates (n=62)

S/No	Referral Unit	No: of isolates
1	Intensive care unit	46
2	Medical unit	9
3	Surgical unit	5
4	Nephrology unit	2

clinical specimens and antimicrobial susceptibility were expressed as frequency and percentages.

RESULTS

A total of eighty five Acinetobacter spp. were isolated during the study period. Out of these 85 isolates, 62 (72.94%) were found to be (3.22%) from both the sputum and catheter tip as shown in fig-1. As per the referral units maximum number of the samples were received from the medical intensive care unit [(ICU) (46)], out of which 35 were from the nasobronchial lavage, (table). The antimicrobial sensitivity of the carbapenem resistant

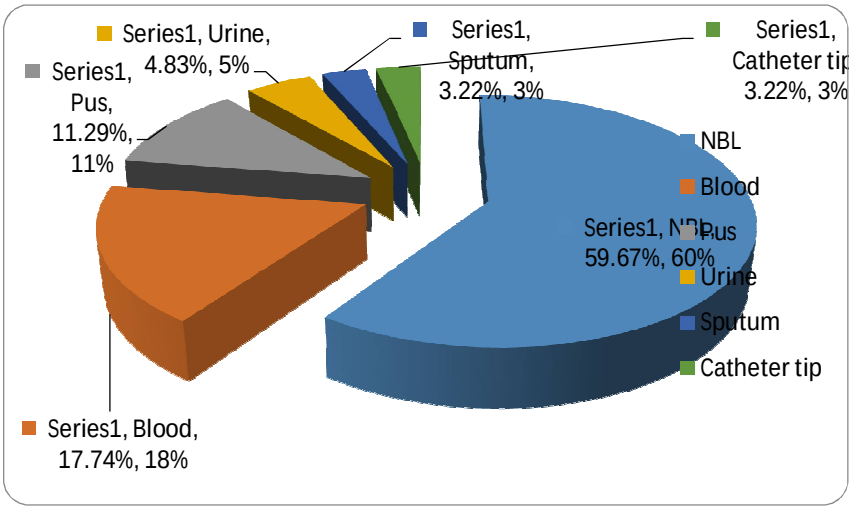

Figure-1: Percentage wise distribution of carbapenem resistant acinetobacter in different clinical samples (n=62).

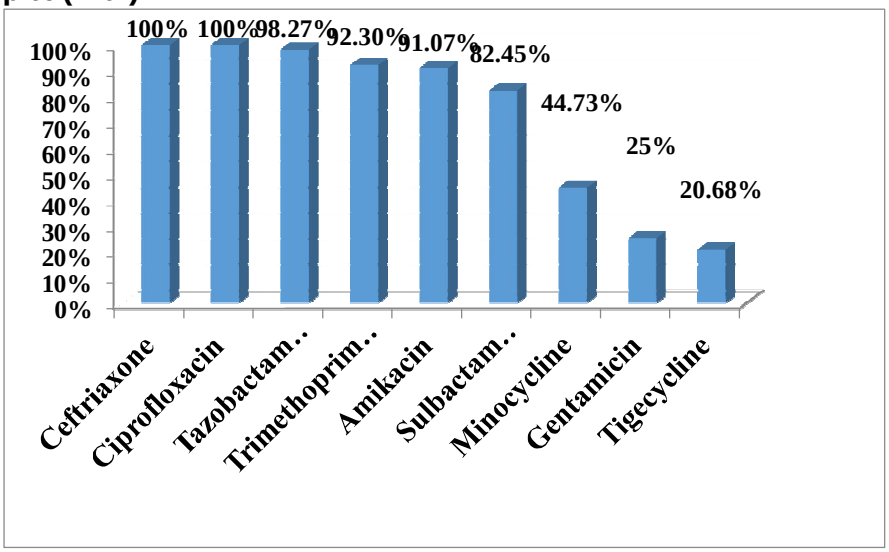

Figure-2: Resistogram of carbapenem resistant acinetobacter isolates (n=62).

resistant to carbapenems. The maximum number of the carbapenem resistant acinetobacter spp were isolated from nasobronchial lavage (59.67%) followed by blood (17.74%), pus (11.29%), urine (4.83%) and Acinetobacter isolates showed maximum resistance to ciprofloxacin and ceftriaxone being (100%) followed by tazobactam-piperacillin (98%), trimethoprim sulphamethoxazole (92%), amikacin (90%), sulbactam-cefoperazone (82%),

minocycline (45%), gentamicin (26%) and minimum resistance was seen against tigecycline being 21% as is show in fig-2.

Apart from being carbapenem resistant all the isolates were found to be multidrug resistant (MDR) as well, showing 100% resistance to ciprofloxacin and ceftriaxone simultaneously, of course maximum being isolated from the ICUs. None of the isolates were detected to be an ESBL producer.

DISCUSSION

Notorious for their ability to acquire antibiotic resistance, Acinetobacter spp also have the ability to cause nosocomial outbreaks. Carbapenems are beta-lactam antibiotics, with a wide range of activity against gram positive organisms, gram negative organisms and anaerobes as well[16]. They were considered to be the most potent agents for the treatment of infections caused by gram-negative bacilli. Presently, the highly increased level of resistance in Acinetobacter against carbapenems has limited this option to a major extent. Till the early 1970s, nosocomial Acinetobacter infections were successfully treated with antibiotics like gentamicin, minocycline, nalidixic acid and ampicillin[3].

The main problem faced nowadays is the beta-lactamase producing Acinetobacter spp. including serine and metallobeta-lactamase which are resistant to carbapenems[12,17]. The incidence of hospital acquired infections with Acinetobacter spp has increased, the main reason being the injudicious use of antibiotics. In a study conducted by Mahmood et al in the year 2002 almost all the isolates of Acinetobacter spp were reported to be sensitive to carbapenems[16]. In contrast, our study revealed a high percentage of Acinetobacter spp resistant to carbapenems (72.94%). Apart from the increased frequency of resistance to carbapenems the specimen-wise distribution in our study revealed that the maximum number of the carbapenem resistant Acinetobacter spp. were isolated from the samples obtained from the respiratory tract, which was also observed in a study conducted by Ghorbanalizadegan et al , in the year 2007[17]. Concordant with this, our study also showed that the maximum number of the samples growing Acinetobacter spp were obtained from the patients admitted in the intensive care units.

A study conducted by Rahbar et al, in the year 2008 in Iran reported,that the resistance of Acinetobacter against meropenem was 38.5%[10] which in our study was found to be almost the double of that value i.e. 72.94% which shows an increased level of resistance in the Acinetobacter species against carbapenems as seen in the previous studies.

In another study conducted in Kuwait by Al-Sweih et al in the year 2011[18] the incidence of meropenem resistant Acinetobacter was reported to be 50% concordant with a study from Turkey in 2008 by Gur et al[19].

In a recent study by Higgins et al in Brazil from 2004 to 2008, 36 carbapenem-resistant A. baumannii isolates recovered from different patients hospitalized in eight medical centers were screened for carbapenem resistance in which all the organisms were found to be resistant to both imipenem and meropenem[20].

The antimicrobial sensitivity in our study showed 100% resistance to ceftriaxone and ciprofloxacin making all the carbapenem resistant Acinetobacter spp multi drug resistant which was concordant with a study conducted by Vala et al in the year 2013, from Tehran, Iran showing 93% of the carbapenem resistant isolates to be MDR[21].

From an organism of very little significance Acinetobacter has made tremendous progress in the last three decades not only to become the leading pathogen of the intensive care units globally, but also poses a serious threat in terms of its challenge to the therapeutic options usually considered a clinician's last armamenterium.

CONCLUSION

The frequency of carbapenem resistant Acinetobacter spp is dangerously high (72.94%), which in the coming few years may come up with the remaining quarter of Acinetobacter spp. Becoming resistant to carbapenems, thus leaving us without any options to treat them.

CONFLICT OF INTEREST

There is no conflict of interest of any author in this study.

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