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Multidrug-Resistant *Acinetobacter*

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K e y w o r d s

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A B S T R A C T

Acinetobacter species has been increasingly reported as the cause of nosocomial infections and possess a serious threat to the health care system because of its multi-drug resistance. The present study was a prospective study carried out in the Department of Microbiology from August 2013 to November 2015 in a tertiary care hospital to isolate and speciate *Acinetobacter* species from clinical samples and to determine their antibiogram. 150 clinical isolates of Acinetobacter species were processed for species identification and antimicrobial susceptibility of these isolates was performed by Kirby-Bauer disc diffusion method. Out of 150 *Acinetobacter* isolates, 94(62.67%) were extensively drug resistant, 18 (12%) were multi-drug resistant and 15(10) of the isolates were pan-drug resistant. Proper application of infection control measures and antibiotic stewardship is necessary in order to combat this problem

Introduction

Acinetobacter species has emerged as one of the most troublesome pathogens in the in the healthcare setting both globally and locally.

Its remarkable ability to develop or acquire multiple antibiotic resistance and propensity to survive for prolonged periods under a wide range of environmental conditions, make it a frequent cause of hospital outbreaks and an endemic healthcare associated pathogen.

It commonly targets the most vulnerable hospitalised and critically ill patients with breaches in skin integrity who require airway protection, causing pneumonia, urinary tract infection, wound infection and bacteremia (Tak-chiu, 2011).

Control of hospital acquired infection caused by multi resistant gram negative bacteria by using broad spectrum antibiotics in hospital has subsequently resulted into increased into infection by gram negative bacteria like *Pseudomonas aeruginosa, Stenotrophomonas maltophilia and Acinetobacter species* (Bergogne-Berezin *et al.,* 1996). Among these pathogens *Acinetobacter* species play a significant role in colonisation and infection of patients admitted in hospital. Where they have been implicated in a variety of nosocomial infections like bacteraemia, urinary tract infection, secondary meningitis, ventilator associated pneumonia and wound infections (Bergogne-Berezin, 1987; French *et al.,* 1980). Among the various risk factors

for colonisation and infection exposure to Carbapenems, broad spectrum antibiotics is most common factor for acquisition of drug resistance in *Acinetobacter* (Gaur *et al.*, 2007).

Now a day *Acinetobacter* are revealing resistance to most commonly used antibiotics. They are now becoming Multi-resistant (MDR). *Acinetobacter* resistant to any three classes from third generation Cephalosporins, Penicillins, Aminoglycosides and Fluroquinolones are considered as Multi-drug resistant (MDR) while in addition to this if Carbapenem resistant isolates is also noted considered as Extensively drug resistant (XDR) (Manchanda, 2010).

Acinetobacter resistant to all antimicrobial drug used including Colistin and Tigecycline considered to be Pan drug resistant (PDR). Emergence of drug resistance limits therapeutic options for effective treatment.

Materials and Methods

The present study was conducted in the department of Microbiology. Wound swab, pus samples were collected from the patients of post-operative wound infections.

Sample collection

Pus/ Exudate was collected from deeper part of the wound with the help of two sterile swab sticks in sterile test tube and transported immediately to laboratory.

Identification of *Acinetobacter* species was done by using standard laboratory methods.

Antibiotic susceptibility testing

All the bacterial isolates were subjected to antibiotic susceptibility testing by Kirby Bauer disc diffusion technique (Winn, 2006) (Figure 1).

Since CLSI guidelines for Colistin disc diffusion are not available for *Acinetobacter* species so we used *Pseudomonas aeruginosa* CLSI 2013 guidelines (CLSI 2013). Standard strains of *E. coli* (ATCC- 25922), *Pseudomonas aeruginosa* (ATCC-27853) were used as controls.

Antibiotics used

Commercially available antibiotic disks obtained from Hi- Media Laboratories Ltd, were used. Antibiotics used were Piperacillin (100µg), Ampicillin (30 µg), Ceftazidime (30µg), Ceftriaxone (30µg), Cefepime (30µg), Ampicillin-sulbactum (10/10 µg), Imipenem (10µg), Amikacin (30µg), Ciprofloxacin (5µg) and Colistin (10µg)

Results and Discussion

Total 150 isolates were tested for antibiotic resistant. According to their resistance pattern they are labelled as MDR, PDR and XDR as per definitions. Antibiotic sensitivity pattern of 150 Acinetobacter isolates is as shown in table 1.

As per the definition of MDR *Acinetobacter* 12% were MDR,62.67 % strains were also resistant to Carbapenem along with MDR so considered as XDR and 10 % strain were resistant to all antibiotic including Tigecycline and Colistin considered as PDR (Figure 2).

Acinetobacter exhibited varying degree of resistance to antibiotics. In spite of being on antibiotic prophylaxis if the patient is developing wound infection, the probability of the isolating *Acinetobacter* species with drug resistance is more. In *Acinetobacter* wound infection, it is likely to become Multi-drug resistant (MDR) or Extensive drug

resistant (XDR) or Pan-drug resistant (PDR) upon prior antibiotic use (Table 2).

In present study, *Acinetobacter* species were resistant to Ampilcillin n=144 (94%) followed by Ciprofloxacin n=140 (93.33%).

In Cephalosporins maximum number of isolates were resistant to Cefepime n=132 (86.67%) followed by Ceftriaxone n=127 (84.67%) followed by Ceftazidime n= 125 (82.67%).Amikacin resistance was n=125 (82%).

In present study resistant to Carbapenem is high, viz.to Imipenem was 78% and Meropenem was 76.67%. Colistin was found to be most susceptible antibiotic as 127 (84.67%) were susceptible. followed by Thirty six isolates (24%) were sensitive to Ampi-sulbactum. Result of present study correlate with many other studies.

Gaur *et al.*, (2008) study from north India showed similar results. In their study antibiotic susceptibility testing was done by disc diffusion method. *Acinetobacter* isolates revealed 80% resistance to third generation Cephalosporins and in Quinolones, 81% were resistant to Ciprofloxacin. Among Aminoglycosides, 74% were resistant to Amikacin (Gaur *et al.*, 2008)

A study from south India showed high resistance to Ampicillin (86.8 %) which is similar to present study. In their study Ceftazidime (74.5 %), Amikacin (51.6 %), Gentamicin (60.6%), and Ciprofloxacin (69.6 %) (Sivaranjani, 2013).

Table.1 Antibiotic sensitivity pattern of *Acinetobacter* isolates by disc diffusion and agar dilution

Sr no	Antibiotic(µg)	Disc diffusion (n=150)			
		Sensitive		Resistant	
		No of strains	Per-centage (%)	No of strains	Per-centage (%)
1	Ampicillin	6	4	144	94
2	Ceftrixone	23	15.33	127	84.67
3	Ceftazidime	25	16.67	125	83.33
4	Cefepime	18	12	132	88
5	Ampi-sulbactum	36	24	114	76
6	Ciprofloxacin	10	6.67	140	93.67
7	Imipenem	26	17.33	124	82.67
8	Colistin	124	82.67	26	17.33
9	Amikacin	25	16.67	125	83.67

Table.2 MDR, XDR and PDR distribution of *Acinetobacter* isolates

	MDR	**XDR**	**PDR**
Total no of strain	18	94	15
Percentage	12	62.67	10

Fig.1 Antibiotic screening test by Kirby Bauer method

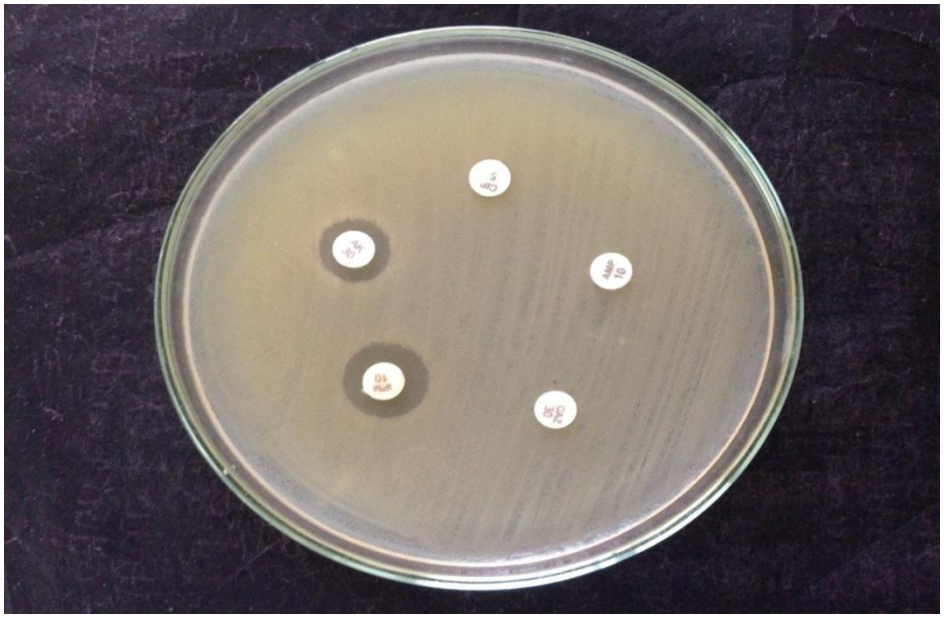
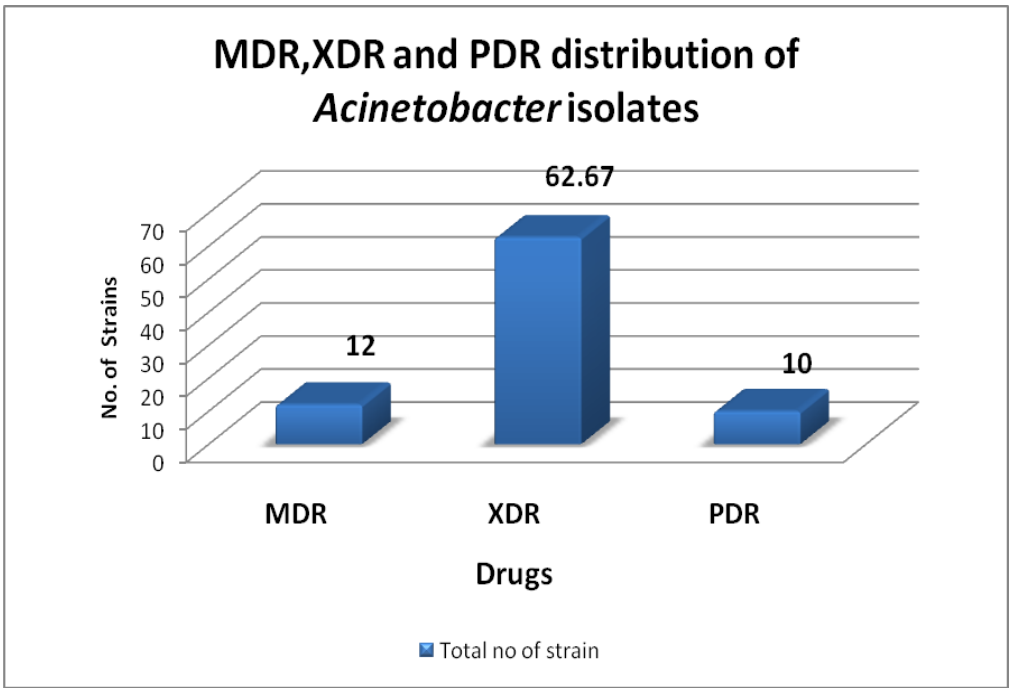


Fig.2 MDR, XDR and PDR distribution of *Acinetobacter* isolates



Another study of Fatma *et al.*, (2014) showed high resistance of *Acinetobacter* species to different classes of antibiotics. She also reported high resistant to different generation of Cephalosporins viz. 88.3% of isolates were resistant to Ceftazidime and Ceftriaxone, 90% to Cefotaxime, 91.7% to Cefepime. Resistance to other classes of drug was 96.7% Ampicillin/sulbactam, 76.7 to Amikacin, 86.7 to Ampicillin and 75% Ciprofloxacin. This correlates with present study (Al-Zahraa, 2014).

In Rahul Kamble study (2015), 98.84%, *Acinetobacter* species were resistant to Ceftriaxone, 98.26% to Ceftazidime, 94.35% to Cefepime, 79.7% to Ciprofloxacin and 72% to Amikacin (Rahul Kamble study, 2015).

In Sana Islahi study (2014), >80% of *Acinetobacter* isolates were resistant to Amikacin, Gentamycin, Ceftriaxone, Ciprofloxacin and Tetracycline (Sana Islahi study, 2014). These results are similar to present study. Whereas resistant to colistin was 19.56%.

In Shrestha *et al.,* (2008) study, resistance to Amikacin and Gentamycin was low (51%) as Aminoglycosides used as reserved drug for *Acinetobacter* infection and mostly in combination with Cephalosprins (Shrestha *et al.,* 2008).

Acinetobacter resistant to any three classes from third generation Cephalosporins, Penicillins, Aminoglycosides and Fluroquinolones are considered as Multi-drug resistant (MDR) while in addition to this if Carbapenem resistant isolates is also noted considered as Extensively drug resistant (XDR). *Acinetobacter* resistant to all antimicrobial drugs used including Colistin and Tigecycline considered to be Pan drug resistant (PDR) (Mnachanda *et al.,* 2010). In present study, 12% isolates were MDR, 62.67 % isolates were XDR and 10 % were PDR. Carbapenem are the drug of choice for MDR *Acinetobacter* infection so their use is increased now days, so recently we came across with XDR *Acinetobacter* in hospitals. Our observations are consistent with Rahul Kamble (2015) study from Mumbai (Rahul Kamble, 2015). He noticed 14% isolates were MDR and 58% Isolates were XDR. No PDR isolates were found in their study.

Inchai Juthamas *et al.,* (2015) observed MDR were <20%, XDR 65.3% and 3.6 % PDR *Acinetobacter*. Japoni-Nejad A. *et al.,* (2013) noticed 11% PDR and 89% XDR *Acinetobacter*. In their study more number of XDR could be due to small study group and samples mainly from seriously ill patients where Carbapenem use is frequent (Inchai Juthamas *et al.,* 2015; Japoni-Nejad *et al.,* 2013).

Acinetobacter species is emerging as an opportunistic pathogen which causes numerous infections in hospitalised patients. As a result of alarming increase in its drug resistance, it seems that nowadays we left with very few options of antibiotic to treat it. Furthermore, the inconsistencies in defining multidrug resistance in this pathogen have caused considerable confusion to both clinicians and researchers. One way to circumvent this is to have proper definitions for terms like MDR, XDR and PDR through surveillance programmes.

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