Antimicrobial susceptibility of clinical isolates of *Pseudomonas* aeruginosa for Mansoura area.

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ABSTRACT

The aim of the present study was to determine both susceptibility and resistance pattern among clinical isolates of *Pseudomonas aeruginosa* in order to make continuous monitoring to the action of antipseudomonal drug in Egypt and to initiate the treatment with the appropriate antibiotic to avoid failure of treatment due to resistance. A total of 104 clinical isolates were collected and identified as *Pseudomonas aeruginosa* from Mansoura University Hospitals. The isolates were tested for antimicrobial susceptibility against 10 different antibiotics by using disk diffusion method. Resistance rates for imipenem, piperacillin, amikacin, gentamicin and ciprofloxacin were found 19.23%, 20.19%, 24%, 26.92%, and 30.76%; respectively. Resistance rates to chloramphenicol, co-trimoxazole, ceftazidime and ceftriaxone were variable and reached up to 83.65%. Exceptionally all isolates were resistant to carbencillin (100%). Thirty-six (34.61%) of isolates were multi-drug resistant. Imipenem, piperacillin and amikacin were the most effective antimicrobial agents against *Ps. aeruginosa* clinical isolates. It is recommended by the health authority to limit the further increase of antimicrobial resistance among *Ps. aeruginosa* by declining the rational treatment regimen.

Key words: Susceptibility, Disk method, *Pseudomonas aeruginosa*, Antimicrobial, Multidrug resistant.

INTRODUCTION

Continuous survey for susceptibility pattern of *Pseudomonas aeruginosa* in Egypt geographical area is important to grade the antibiotic therapy and for developing resistance control. The present study aimed to determine the antibiotic susceptibility profile in level variable by Hospitals of Mansoura University.

Pseudomonas aeruginosa is а notorious opportunistic pathogen and is isolated mostly from patient with urinary tract infections, wound infections, and severe burns. Ps.aeruginosa has been considered a nosocomial pathogen in a number of studies. Ps.aeruginosa infections are also known to be a serious problem in hospitalized patients with cystic fibrosis, cancer and burns. Half of these infections are fatal. Those infections are problematic due to the bacterial resistance to antibiotic and disinfectants (Lambert, 2002).

Proliferation and transmission of antimicrobial-resistant bacteria is accelerated by close contact between individuals in institutions such as hospitals, day care centers and military barracks (Goldmann, 1999). The mobility of the world's populations also facilitates the rapid dissemination of microorganisms harboring antibiotic resistance genes (Goldmann, 1999). The spread of antibiotic resistance within a hospital, country or globally is usually achieved by clones with a high transmission ability (Livermore, 2003).

The evolution and spread of antibiotic resistance in bacteria is a complex process involving a variety of different mechanisms. Some bacterial species show a high intrinsic resistance to specific antibiotics, while susceptible bacteria may acquire resistance by alterations in their own genome via mutations or the transfer of resistance genes located on mobile DNA elements (Normark and Normark, 2002).

Pseudomonas aeruginosa show intrinsic and acquired resistance to many structurally unrelated antibiotics, and extensive use of antibiotics often leads to multidrug resistant *Pseudomonas aeruginosa* strains (Toscano *et al.*, 1991; Ciofu *et al.*, 1994).

MATERIAL and METHODS Bacterial isolates

One hundred and four non duplicate *Ps. aeruginosa* isolates were collected from Mansoura University hospitals, from different clinical samples (wound swab, burn swab, ear swab, endotracheal swab and urine), identified and verified by using the standard biochemical reactions according to Collee *et al.* (1996). All isolates were collected under approved ethical procedures. **Antimicrobial Susceptibility testing**

The susceptibility was determined by disk diffusion method according to CLSI (2006) on Muller-Hinton agar (Oxoid UK). The susceptibility against the clinical isolates was testing performed against 10 antimicrobial agents belonging to 5 different groups: Ceftriaxone (CRO, 30µg). Piperacillin (PRL, 100µg), Carbenicillin (CAR, 100µg), Ceftazidime (CAZ, 30µg), Imipenem (IPM, 10µg), Ciprofloxacin (CIP,

5µg), Amikacin (AK, 30µg), Gentamicin (CN, 10µg), Sulfamethoxazole /Trimethoprime (SXT, 23.27/1.25µg), Chloramphenicol (C, 30µg). The antibiotics disks were obtained from Oxoid, Hampshire, England. The results were interpreted according to the criteria indicated in CLSI (2006) guideline.

RESULTS

Susceptibility patterns of *Ps. aeruginosa* to different antibiotics

Antimicrobial susceptibility pattern of Pseudomonas aeruginosa were varied markedly with the antibiotic tested. Highest resistance rates were found to carbencillin (100%) and the least resistance were found to imipenem (19.23%). The resistance pattern of Ps. aeruginosa to various antibiotics tested was in the following descending order: carbencillin (100%), ceftriaxone (83.65%), ceftazidime (69.23%), sulfamethoxazole/trimethoprime (56.73%), chloramphenicol (38.46%), ciprofloxacin (30.76%), gentamicin (26.92%), amikacin (24%), piperacillin (20.19%) and imipenem (19.23%) as shown in Table (1) and Figure (1-2). Multi-drug resistance (resistance to 3 or more different families of antibiotics tested) was found in 36 isolates (34.61%) of Ps. aeruginosa tested (Table 2).

Table (1): Susceptibility patterns of Ps	seudomonas aeruginosa to different antibiotics.
Antibiotic Disk	Pseudomonas spp (n=104)

Anubiouc Disk	Pseudomonas spp (n=104)						
	R			Ι		S	
	Ν	%	Ν	%	Ν	%	
PRL	21	20.19	0	0	83	79.8	
IPM	20	19.23	2	1.93	82	78.84	
CAZ	72	69.23	18	17.3	14	13.46	
CRO	87	83.65	3	2.88	14	13.46	
CAR	104	100	0	0	0	0	
AK	25	24	3	2.88	76	73	
CN	28	26.92	4	3.84	72	69.23	
CIP	32	30.76	7	6.73	65	62.5	
C30	40	38.46	21	20.19	43	41.34	
SXT	59	56.73	32	30.76	13	12.5	

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Table (2): Numerical	antimicrobial	resistance	patterns of	Pseudomonas	aeruginosa	clinical isolates.

No of groups of antibiotics	No of <i>Ps. aeruginosa</i> isolates	% of Ps. aeruginosa isolates
Resistance < 3 groups	68	65.38%
Resistance = 3 groups	1	0.96%
Resistance $= 4$ groups	16	15.38%
Resistance $= 5$ groups	19	18.26%



Figure (1): Susceptibility of *Pseudomonas aeruginosa* toβ-Lactam (Subclass Penicillins), used Piperacillin (PRL) and Carbencillin (CAR), (Subclass Cephalosporins), used Ceftazidime (CAZ) and Ceftriaxone (CRO), (Subclass Carbapenem), used Imipenem (IPM).



Figure (2): Susceptibility of *Pseudomonas aeruginosa* to Quinolone used Ciprofloxacin (CIP), Aminoglycosides used Amikacin (AK) and Gentamicin (CN), Chloramphenicol (C₃₀), Suphamethoxazole trimethoprime (SXT).

DISCUSSION

Among the 104 Pseudomonas aeruginosa isolates collected, thirty six

(34.61%) were showed multidrug resistance while other study revealed that (20.69%) reported in Nepal by Chander *et al.* (2013),

while other study revealed that (19.6%) reported in Malaysia by Pathmanathan *et al.* (2009). From these MDR isolates only one (2.77%) isolate was resistant to 3 groups of antimicrobial agents, sixteen (44.44%) isolates were resistant to 4 groups of antimicrobial agents and nineteen (52.77%) isolates were resistant to 5 groups of antimicrobial agents (Table 2).

In the present study about 19.23% of *Ps.aeruginosa* isolates were resistant to imipenem (IPM) which was less than 39.34% and 41.4% reported in Egypt by Zafer *et al.* (2014) and Afifi *et al.* (2013), respectively. One study in the world was in complete agreement with the present study reported by Bonfiglio *et al.* (19.3%) in 1998, and other studies gave comparable results in the range between 14% and 26.1% reported by (Bouza *et al.* 1999; Fatima *et al.* 2012; and Akhtar, 2010); respectively.

Our investigation also showed about 20.19% of *Ps.aeruginosa* isolates were resistant to piperacillin (PRL) which was diverted with that reported by Al-Tawfiq (2007) and Shenoy *et al.* (2002) where 11.5% and 54.66% of the isolates, respectively were resistant to piperacillin.

In the present study about 69.23% of *Ps.aeruginosa* isolates were resistant to ceftazidime (CAZ) which in agreement with Zafer *et al.* 2014 in Egypt which reported the resistance 60.6% and disagreement with that reported by Afifi *et al.* (2013) in Egypt where 19.5% of the isolates were resistant.

In the present study, the clinical isolates show high resistance against ceftriaxone, where 83.65% of isolates were resistant. This high resistance was in agreement with that reported by Afifi *et al.* (2013), Rashid *et al.* (2007) and Bhandari *et al.* (2012) where 78.1%, 86% and 93.9% of the isolates were resistant.

In the present study, 24% of *Ps.aeruginosa* isolates were resistant to amikacin (AK), which was in agreement with

that reported by Zafer *et al.* 2014 (32.8%), where 7.8 % of the isolates were reported resistant by Afifi *et al.*(2013). High resistance was expressed by the same organisms in studies performed by Viren *et al.*(2008), and Sivanmanliappan & Sevanan (2011) where 48.2% and 33.4% were susceptible; respectively.

In the present study about 26.92% of *Ps.aeruginosa* isolates were resistant to gentamicin (CN) which in agreed with that reported by Afifi *et al.* 2013 (21.1%). This resistance was less than that reported by Zafer *et al.* 2014 and Viren *et al.* (2008) where 50% and 67.86% of the isolates were resistant; respectively. High resistance was expressed by the *Ps.aeruginosa* in study carried by Updhaya *et al.* (2014) where 47.1% of resistance were recorded.

According to Gales *et al.* (2001), ciprofloxacin was reported as one of the most potent drug. As shown in Table (1) and Figure (2), this investigation revealed that 30.76% of the isolates were resistant to ciprofloxacin, this result was consistent with that reported by Zafer *et al.* 2014 (43.4%), Amadi *et al.* 2008 (25.5%), Updhaya *et al.* 2014 (35.5%) and Brown *et al.* 2004 (23.5%), respectively.

In the present study about 38.46% of *Ps.aeruginosa* isolates were resistant to chloramphenicol (C30). This resistance was less than that reported by Amadi *et al.* 2008 (58.8%) and Viren *et al.* 2008 (75%).

In the current study about 56.73% of Ps. aeruginosa isolates were resistant to sulphamethoxazol/trimethoprim(SXT) which is consistent with that reported by Brown et al.(2004) and Chander *et al.*(2013), where 56.9% and 51.72% of the isolates were resistant; respectively. The data revelead that amikacin. imipenem, piperacillin, gentamicin, and ciprofloxacin are considered options the best in treatment of Ps.aeruginosa. Other antimicrobial agent under investigationwere not encouraged to be

considered in the treatment of *Ps.aeruginosa* infections (Chander et al., 2013; Brown et al., 2004 and Fatima et al. 2012). In conclusion, results of the present study revealed the distributions of the resistance to various antipseudomonal drugs among the Ps.aeruginosa isolates. Imipenem and piperacillin are still the best as antipseudomonal drugs. Aminoglycosides (amikacin and gentamicin) and ciprofloxacin have good activity against the Ps.aeruginosa Periodic monitoring of isolates. the antimicrobial has great importance in order to help the medical team to prescribe the optimal antipseudomonal agents and rule out the ineffective ones from the regimen therapy as well as the avoidance of emerging multi-drug of resistant strains of Ps.aeruginosa.

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تعيين حساسية العزلات السريرية لميكروب السودوموناس ايروجينوزا ضد المضادات الحيوية في المنطقة المحيطة لمدينة المنصورة

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تهدف الدراسة الحالية الى تقييم فاعلية المضادات الحيوية ومدى تطور المقاومة بين عزلات السودوموناس اريجنوزا تجاه هذه الادوية . تم تجميع والتعرف على ١٠٤ عزلة من عزلات السودوموناس اريجنوزا من مستشفيات جامعة المنصورة . تم عمل اختبار الحساسية بطريقة الانتشار الاجاري للقرص ضد ١٠ مضادات حيوية مختلفة.

أوضحت النتائج وجود مقاومة عالية من عزلات السودوموناس اريجنوزا تجاه كل من الكاربنسللين والسلفاميثوكسازول/ترايميثوبريم. بينما اظهرت النتائج فاعلية كل من الاميبنيم والبيبر اسللين ضد عزلات السودوموناس اريجنوزا. كما اوصت الدراسة الحالية بضرورة متابعة تطور المقاومة بين عزلات السودوموناس اريجنوزا وذلك لتجنب خطورة عدم فاعلية الادوية ضد الامراض المسببة ببكتريا السودوموناس اريجنوزا.