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Prevalence and Antibiotic Susceptibility Profile of *Pseudomonas aeruginosa* Isolated from Different Clinical Samples in District Peshawar Pakistan

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Currently, antibacterial agents are widely used to treat bacterial infections. However, bacterial populations have adopted resistance mechanism against these antibacterial agents worldwide. *Pseudomonas aeruginosa,* which is a Gram-negative commensal of human microflora is one of the most common causative agents of nosocomial infections. Different types of clinical samples including pus, blood, urine, fluids and different swabs were collected from patients in district Peshawar, Pakistan. The pathogens were isolated by using sterile bacteriological media, including blood agar, MacConky agar, CLED agar and chocolate agar. All the clinical isolates of *P. aeruginosa* were tested for their sensitivity against antimicrobials including: amikacin (30 ug), ceftazidime (30 mcg), cefoxitin (30 ug), ceftriaxone (30 ug), cefipime (30 ug), ciprofloxacin (5 ug), co-amixoclave (30 ug), gentamicin (10 ug), imipenum (10 ug), meropenum (10 ug), sulzone (91 ug), fosfomycin (50 ug) and pipracillin-tazobactum (110 ug) of standard strengths respectively. We have observed the sensitivity profile of *P. aeruginosa* against Pipracillin-tazobatum is 90.4%, sulzon is 86.6%, amikacin is 82.8%, imipenum is 82.8%, meropenum is 80%, ceftazidime is 71.4%, fosfomycin is 69..5%, cefipime is 67.6%, cifrofloxacin is 71.6%, gentamycin is 63.8%, cefoxitin is 56.1%, co-amixoclave is 54.2% and ceftriaxone is 2.8% respectively. The results obtained herein indicate that a combination of pencilline+beta-lactamase inhibitors, imipenem and amikacin were the drugs of choice that can be used against multidrug resistant strains of *P. aeruginosa.*

Key words: *Pseudomonas aeruginosa*, Prevalence, Antibiotic susceptibility.

Antibacterial agents are widely used to treat infections caused by bacteria. However, bacterial populations have adopted resistance mechanism against these antibacterial agents worldwide. *Pseudomonas aeruginosa* is one of the most common causative agents of nosocomial infections. It is a gram-negative commensal of human microflora in healthy people and is frequently isolated as an opportunistic pathogen in recurrent infections of hospital admitted patients. It is a cost effective bacteria both in community and hospitals (Franco *et al*., 2009; Poole *et al*., 2011).

Mechanisms of resistance are due the mutations in the coding genes e.g. those encoding β-lactamase (Zhao and Hu, 2010) and amino-glycoside modifying enzymes (Poole, 2005) via horizontal gene transfer and mutation of chromosomal genes (target site, efflux mutations) are the target of the fluoroquinolones particularly ciprofloxacin (Strateva and Yordanov, 2009).

Biofilm formation in *P. aeruginosa* contributes the resistance to antibiotics in pulmonary infections with cystic fibrosis (Davies and Bilton, 2009). In chronic cystic fibrosis of lungs infections, hyper mutable strains of *P. aeruginosa*

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were reported (Oliver and Mena, 2010). *P. aeruginosa* is the most common pathogen responsible for nosocomial infection. The acquired drug resistance is contributed by several factors like increased efflux pump, changing pencilline binding proteins (PBPS) and reduced the permeability of the membrane (Khakhkhar *et al*., 2012). *P. aeruginosa* are common pathogen associated with burn infections. They are difficult to treat due to its high frequency of resistance against antimicrobial agents (Ullah *et al*., 2009).

The current study was designed to find out the prevalence and antibiotic susceptibility profile of *P. aeruginosa* isolated from clinical samples in district Peshawar, Pakistan.

MATERIALS AND METHODS

Sample processing

Between June, 2012 and July, 2013, 1040 samples including blood, pus, urine, body fluids and pus swabs were collected aseptically from the patients and brought to the microbiology section of united medical laboratory in Peshawar.

Isolation and identification of pathogen

The pathogens were isolated by using the following standard protocols using sterile bacteriological media, including blood agar, MacConky agar, CLED agar and chocolate agar. All samples were inoculated aseptically and incubated at 37°C for 24 h. Identification of the organisms was done on the basis of Gram staining and biochemical tests including, Oxidase, Indole, Urease, Tripple Sugar Iron and Motility (Ejaz *et al*., 2006; Beyene *et al*., 2011).

Antibiotic Susceptibility Profile

All the isolates of *P. aeruginosa* were subjected for antibiotics susceptibility *in vitro* by using standard Kirby-Bauer disc diffusion method on Muller Hinton agar according to the Clinical Laboratory Standard Institute, (CLSI). A total of thirteen antibiotics were used for the study which were included, amikacin (30 ug), ceftazidime (30 mcg), cefoxitin (30 ug), ceftriaxone (30 ug), cefipime (30 ug), ciprofloxacin (5 ug), co-amixoclave (30 ug), gentamicin (10 ug), imipenum (10 ug), meropenum (10 ug), sulzone (105 ug), fosfomycin (50 ug) and pipracillin-tazobactum (110 ug) of standard strengths. The plates were incubated at 37ºC for 18 h. After incubation, culture plates were examined for zones of inhibition and reported the organism sensitive, intermediate, resistant according to national committee for control laboratory standards (Wayne, 2002; Bauer *et al*., 1996).

RESULTS AND DISCUSSION

A total of 1040 clinical samples were processed for isolation of *Pseudomonas aeruginosa*. Out of 1040 samples, 105 (10.09%) were found positive for *P. aeruginosa* (Fig. 1). Among 105 positive samples for *P. aeruginosa*, 46 (47.91%) were isolated from pus samples, 24 (25%) were from urine samples, 12 (12.5%) were from fluids

Table 1. Sensitivity profile of selected antibiotics against *Pseudomonas aeruginosa* isolated from different clinical samples

S. No	Antibiotics	Sensitivity	Sensitivity %	Resistant	Resistant %	Intermediate	Intermediate %
1	Ceftazidime	75	71.43	28	26.67	2	1.90
2	Sulzone	91	86.67	10	9.52	4	3.81
3	Cefoxitin	59	56.19	35	33.33	3	2.86
4	Amikacin	87	82.86	8	7.62	10	9.52
5	Co-amixoclave	57	54.29	44	41.90	4	3.81
6	Gentamicin	67	63.81	32	30.48	6	5.71
7	Ciprofloxacin	71	67.62	32	30.48	2	1.90
8	Ceftriaxone	3	2.86	102	97.14	0	0.00
9	Fosfomycin	73	69.52	24	22.86	8	7.62
10	Cefipime	71	67.62	32	30.48	2	1.90
11	Imipenum	87	82.86	18	17.14	0	0.00
12	Meropenum	84	80.00	20	19.05	1	0.95
13	Pipracillin-tazobactum	95	90.48	8	7.62	2	1.90

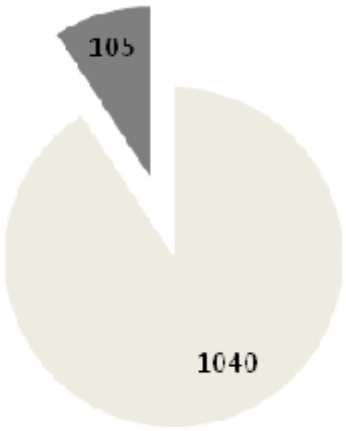

Fig. 1. Pie diagram showing the total cases and positive *Pseudomonas aeroginosa* cases

samples, 8 (8.33%) were from different swabs and 6 (6.25%) were obtained from blood samples.

The antibiogram (Table 1) of *P. aeruginosa* showed that most sensitive drug is pipracillin-tazobactum (TZP) which is 90.47% sensitive then followed by sulzone (SCF) 86.66%, amikacin (AK) 82.85%, mipenum (IPM) 82.85%, meropenum (MEM) 80%, ceftazidime (CAZ) 71.42%, fosfomycin (FOS) 69.52%, cefipime (FEP) 67.61%, ciprofloxacin (CIP) 67.61%, gentamicin (CN) 63.80%, cefoxitin (FOX) 56.19%, co-amixoclave (AMC) 54.28% and ceftriaxone (CRO) 2.85% respectively.

Pseudomonas aeruginosa is one of the most common causative gram negative bacteria of nosocomial infections. In our current study, among 105 positive samples 47.91% were isolated from pus 25% were from urine 12.5% were from fluids samples 8.33% were from different swabs and 6.25% were obtained from blood samples. This report was in agreement with the results of Anjum and Mir (2010) in Rawalpindi, Pakistan, who also reported *P. aeruginosa*, 41% isolated from pus, 32% from urine, 3% from ear swabs, 4% from blood, 3% from catheters, 5% from sputum and 8% from environmental sources. In the study of Olayinka (2004) the frequency *P. aeruginosa* were higher in urine samples, while in our study the prevalence of *P. aeruginosa* was higher in pus samples. This disagreement is due to the differences in geographical location or may be due to the differences in hygienic conditions and species variation.

Among the 96 positive samples for *P. aeruginosa* 54 (56.25 %) were isolated from males while 42 (43.75%) were from females. The prevalence was higher in males than female which was in accordance with the previous report of Fatima *et al*., 2012, who reported a higher prevalence of *P. aeruginosa* (70.80%) in males than females 29.1%. In the present study the susceptibility profiles were tested, pipracillin-tazobatum showed 90.4%, sulzon (cepaferazone+sulbactam) 86.6%, amikacin 82.8%, imipenum 82.8%, meropenum 80%, ceftazidime 71.4%, fosfomycin 69.5%, cefipime 67.6%, cifrofloxacin 71.6%, gentamycin 63.8%, cefoxitin 56.1%, co-amixoclave 54.2% and ceftriaxone 2.8% respectively. The same study was conducted by Anjum and Mir (2010), Pakistan who also reported nearly the same susceptibility pattern for *P. aeruginosa* isolated from different clinical samples. Aman *et al* (2012) reported that *P. aeruginosa* isolates from fresh water showed 94% resistance to chloramphenicol, 88% to colistin sulphate and 84% to cotrimoxazole, which shows that fresh water is a reservoir of resistant pathogens. Zulfiqar *et al* (2005) reported multiple drug resistant *P. aeruginosa* isolated from the burn patients. A high resistance pattern were noted against septran and chloramphenicol as 100% followed by tobramycin 95%, imipenem 77%, ciprofloxacin 54.5%, cefepime 50%, amikacin 30%, aztreonam 6.8% and pipracillin 18.2% respectively. Mansoor *et al* (2009) isolated *P. aeruginosa* from the ear discharge samples which showed a high sensitivity profile against amikacin 96%, ceftazidime 89%, ciprofloxacin 85%, gentamicin 81% and imipenem 76% respectively.

CONCLUSION

Nosocomial infections with multidrug resistance *Pseudomonas aeruginosa* is a problem worldwide which contributing high resistance against different antimicrobial agents. A combination of pencilline+beta-lactamase inhibitors, imipenem and amikacin were the drugs of choice used against multidrug resistant strains of *P. aeruginosa.*

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