

An evaluation of Susceptibility profile and antibiogram of Nosocomial *Pseudomonas aeruginosa*

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ABSTRACT

Background: *Pseudomonas aeruginosa* is considered to be a foremost cause of hospital acquired infections. The initiation of antimicrobial treatment is more frequently empirical; thus it is imperative to have knowledge of the susceptibility pattern of microbes in order to choose upon the most appropriate antimicrobial drug.

Aim: To upgrade rational empirical antibacterial treatment recommendations.

Methods: In total, 160 clinical isolates of *Pseudomonas aeruginosa* were collected from in-hospital patients at a major tertiary care hospital in Lahore from different wards during the period March 2020 to January 2021. Antimicrobial resistance in the current study was done by using Kirby-Bauer method. Wound swab and urine samples showed a high prevalence of *Pseudomonas aeruginosa* isolates.

Results: The results of this study were in accordance to National Committee for Clinical Laboratory Standards (2017) guidelines. *Pseudomonas aeruginosa* isolates taken from different clinical samples showed decreased susceptibility and increased resistance to various antibiotics. *Pseudomonas aeruginosa* exhibited resistance to ceftazidime (60%), imipenem (53%), ciprofloxacin (55%), gentamicin (55%), meropenem (51%), cefoperazone/sulbactam (58%), amikacin (45%), piperacillin/tazobactam (40%) and aztreonam (50%). Department-wise isolation of *Pseudomonas aeruginosa* was surgery 65(40.6%), medicine 44(27.5%), orthopaedics 19(11.8%), ICU 19(11.9%), ENT 7(4.3%) and gynaecology 6(3.7%) ($p \leq 0.001$). Sample-wise isolation of *Pseudomonas aeruginosa* was wound 89(55.6%) and urine 71(44.4%). P value ≤ 0.05 was considered to be statistically significant. In conclusion, majority of the isolates exhibited increased level of resistance to antibiotics. This emphasizes the importance of antibiotic susceptibility testing and optimization of treatment by combining drugs.

Conclusion: This is a global phenomenon. Limiting over-usage of antibiotics is mandatory and implementing newer policies to counteract this problem is necessary.

Keywords: Nosocomial infection, multidrug-resistant, antibiotics, infection, antimicrobial therapy

INTRODUCTION

Pseudomonas aeruginosa is a leading cause of nosocomial infections globally¹. It is a major cause of morbidity and mortality. The underlying problem worsens when these pathogens gain antimicrobial resistance². Emergence of this antibiotic resistant microbe significantly affects the outcomes of treatment thus challenging health care provision and cost effectivity. It is highly unfortunate that resistance of *Pseudomonas aeruginosa* to antibiotics is increasing³. *Pseudomonas aeruginosa* gains resistance to multiple drugs readily. This has led to the emergence of pan-drug resistant *Pseudomonas aeruginosa* that is even resistant to anti-pseudomonal penicillin in addition to cephalosporin, monobactams, aminoglycosides, polymyxins and fluoroquinolones⁴. This study aims to upgrade the recommended rational empirical antibacterial therapy. *Pseudomonas aeruginosa* isolates in the current study were considered to show multidrug resistance if the isolate was found to be resistant to at least three out of these nine drugs: Ceftazidime, Piperacillin/ Tazobactam, Amikacin, Aztreonam, Ciprofloxacin, Gentamicin, Meropenem, Imipenem and Cefoperazone/sulbactam. These antibacterial agents are the primary antimicrobial

drugs employed for treating infections caused by *Pseudomonas aeruginosa*.

MATERIALS AND METHODS

Samples were randomly collected from in-hospital patients in Jinnah Hospital Lahore from different wards during the period March 2020 to January 2021.

Bacterial isolate: A total of 160 isolates of *Pseudomonas aeruginosa* were isolated from various clinical samples from different wards at Jinnah Hospital Lahore. Identification of *Pseudomonas aeruginosa* colonies was done by studying the morphology and by gram staining. This was followed by the production of pigment pyocyanin, various biochemical tests and finally by employing API20NE.

Antibiotic susceptibility testing: Susceptibility pattern of isolates of *Pseudomonas aeruginosa* against different antibacterial drugs was done using Kirby-Bauer disc diffusion method. *Pseudomonas aeruginosa* ATCC 27853 strain was employed as quality control. The antimicrobial drugs against which susceptibility was studied in the current study were amikacin (30µg), ciprofloxacin (5µg), gentamicin (10µg), cefoperazone/sulbactam (75-10µg), imipenem (10µg), piperacillin/tazobactam (100µg),

aztreonam (10µg), meropenem (10µg) and ceftazidime (30µg)¹⁶.

RESULTS

Isolates of *Pseudomonas aeruginosa* showed 55% resistance to Gentamicin, 55% resistance to Ciprofloxacin, 60% to Ceftazidime, 53% to Imipenem, 40% to Piperacillin/Tazobactam, 50% to Aztreonam, 45% resistance to Amikacin, 51% to Meropenem and 58% to Cefoperazone/Sulbactam (Figure 1). Isolates were obtained from wound swab of the order of 55.6% and urine samples 44.4% (Table 1). Specimens were collected from a number of different wards of the hospital. Out of total 160 isolates of *Pseudomonas aeruginosa*, department-wise distribution of samples was surgery (65), medicine (44), orthopaedics (19), ICU (19), ENT (7) and gynaecology (6) (Table 2).

Table 1: The distribution of *Pseudomonas aeruginosa* from various clinical specimens

Site/Source	Number of isolates	% of isolates
Wound	89	55.6
Urine	71	44.4
Total	160	100

Table 2. Distribution of specimens based on wards.

Department	Urine	Wound	Total
Surgery	30	35	65(40.6%)
Medicine	26	18	44(27.5%)
Orthopaedic	4	15	19(11.8%)
ICU	4	15	19(11.9%)
ENT	3	4	7(4.3%)
Gynaecology	4	2	6(3.7%)
Total	71	89	160(100%)

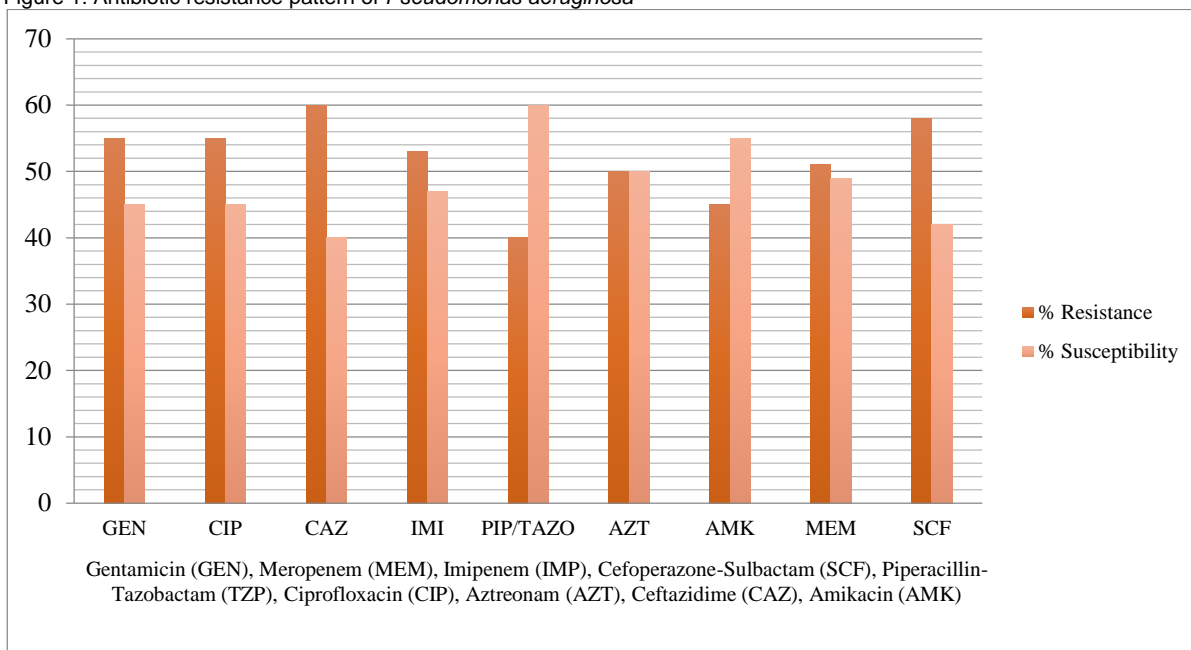
Table 3: Antibiotic resistance of *Pseudomonas aeruginosa* based on wards.

Antibiotic		Surgery	Medicine	Ortho	ICU	ENT	Gynae	Total	p-value
Gentamicin	S	35	14	4	14	5	3	75	0.001
	R	53.8%	31.8%	21%	73.6%	71.4%	50%	46.8%	
	Total	30	30	15	5	2	3	85	
Ciprofloxacin	S	25	15	9	15	4	4	72	0.000
	R	38.4%	34%	47.3%	78.9%	57.1%	66.7%	45%	
	Total	40	29	10	4	3	2	88	
Ceftazidime	S	17	20	12	9	3	3	64	0.062
	R	26.1%	45.4%	63.1%	47.3%	42.8%	50%	40%	
	Total	48	24	7	10	4	3	96	
Imipenem	S	20	29	7	14	3	2	75	0.005
	R	30.7%	65.9%	36.8%	100%	42.8%	33.3%	47%	
	Total	45	15	12	5	4	4	85	
Pip/tazo	S	37	29	10	10	5	5	96	0.048
	R	56.9%	65.9%	52.6%	52.6%	71.4%	83.3%	60%	
	Total	28	15	9	9	2	1	64	
Aztreonam	S	41	21	2	8	5	3	80	0.001
	R	63%	47.7%	10.5%	42.1%	71.4%	50%	50%	
	Total	24	23	17	11	2	3	80	
Amikacin	S	26	20	18	15	6	3	88	0.001
	R	40%	45.4%	94.7%	78.9%	85.7%	50%	55%	
	Total	39	24	1	4	1	3	72	
Meropenem	S	29	14	18	11	3	3	78	0.000
	R	44.6%	31.8%	94.7%	57.8%	42.8%	50%	49%	
	Total	36	30	1	8	4	3	82	
Cefoperazone/Sulbactam	S	25	15	8	12	3	4	67	0.000
	R	38.4%	34%	42.1%	63.1%	42.8%	66.6%	42%	
	Total	40	29	11	7	4	2	93	
Total	S	65	44	19	19	7	6	160	
	R	61.5%	65.9%	57.8%	36.8%	57.1%	33.3%	58%	
	Total	65	44	19	19	7	6	160	
Total	S	65	44	19	19	7	6	160	
	R	100%	100%	100%	100%	100%	100%	100%	
	Total	65	44	19	19	7	6	160	

Table 4. Antibiotic susceptibility pattern of *Pseudomonas aeruginosa* isolates based on site of specimens.

Source	GEN	CIP	CAZ	IMI	PIP/TAZO	AZT	AMK	MEM	SCF
Urine	52	52	62	54	22	29	42	62	62
Wound	69	67	37	55	80	44	39	67	50

Figure 1: Antibiotic resistance pattern of *Pseudomonas aeruginosa*



DISCUSSION

Pseudomonas aeruginosa has emerged as a leading causative agent for nosocomial infections globally and is most certainly a major cause of morbidity and mortality in hospitalized patients. In the current study, 160 isolates from a major government sector hospital in Lahore Pakistan were studied. Unfortunately, multidrug resistance is obvious. Wound and urine isolates were collected. According to Olayinka, usage of indwelling catheters increases risk of infection with *Pseudomonas aeruginosa*⁵. Isolates from wound samples were 55.6% while isolates from urine were 44.4%. Hospitalized patients are usually prescribed multiple antimicrobial drugs leading to colonization of the distal intestinal tract by *Pseudomonas aeruginosa*⁶. The strong intrinsic resistant mechanisms that are possessed by *Pseudomonas aeruginosa* such as β -lactamase enzyme production, major efflux pumps, enzymes that modify aminoglycosides, poor membrane antibiotic permeability plus topoisomerase II and IV alteration make *Pseudomonas aeruginosa* quinolone resistant. Unfortunately, all these mechanisms exist simultaneously giving rise in multidrug resistant strains of *Pseudomonas aeruginosa*. Multidrug resistance in *Pseudomonas aeruginosa* is attributed to all these diverse mechanisms¹².

The lower percentage is attributed to the empirical therapy that most of the patients receive. *Pseudomonas aeruginosa* is a leading cause of septicaemia and pneumonia with recorded death rate of the order of 30%, especially in immunocompromised individuals⁷.

Incidence of MDR *Pseudomonas aeruginosa* is different and varies from community to community and amongst individuals as well^{8,9}. The clinician is expected to manage the antibiotic choice to be most effective using the resistance patterns and prevalence of disease. Guidelines for antimicrobial therapy helps to avoid any misuse of drugs; thus physicians, members of health service team and consultants should show participation in coming up with the best layout of drug regimen¹⁰. Additionally, surveillance programs should periodically be conducted for evaluation of the susceptibility and sensitivity of various bacteria against antimicrobial agents. Emergence of antibiotic resistance remains latent only to be expressed after a certain period; thus antimicrobial agents are to be prescribed with immense caution so as to avoid the problem of antibiotic drug resistance¹¹. Various in vitro sensitivity testing aids in prescribing cost effective treatment to patients having a low socio-economic status.

CONCLUSION

Time-wise evaluation of resistance and susceptibility patterns in different geographical locale helps in monitoring and maintainance of drug resistance profiles. This eventually guides the formulation for a better treatment regimen. Concludingly, *Pseudomonas aeruginosa* showed resistance to most of the antibiotics. Inappropriate administration of antibiotic drugs given as empirical therapy is a leading cause for antimicrobial resistance. This is a global phenomenon. Limiting over-usage of antibiotics is mandatory and implementing newer policies to counteract this problem is necessary.

Funding: No funding was received for this study.

Declaration: This study is part of the Ph.D thesis of Maria Muddassir.

Conflict of interest: None.

Acknowledgements: Authors acknowledge contribution and help from Department of Microbiology, Allama Iqbal Hospital Lahore and concerned staff members at IMBB department, The University of Lahore.

REFERENCES

1. Strateva T, Yordanov D (2009). Pseudomonas aeruginosa-a phenomenon of bacterial resistance. J. Med. Microbiol., 58: 1133-1144.
2. Lagamayo E (2008). Antimicrobial resistance in major pathogens of hospital-acquired pneumonia in Asian countries. Am. J. infect. Cont., 36: S101-S108.
3. Rubin J, Walker R, Blickenstaff K, Bodeis-Jones S, Zhao S (2008). Antimicrobial resistance and genetic characterization of fluoroquinolone resistance of Pseudomonas aeruginosa isolated from canine infections. Vet. Microbiol., 131: 164-172.
4. Babic M, Hujer A, Bonomo R (2006). What's new in antibiotic resistance? Focus on beta-lactamases. Drug. Resist. Upd., 9: 142-156.
5. Olayinka A, Onile B, Olayinka B (2004). Prevalence of multi-drug resistant (mdr) pseudomonas aeruginosa isolates in surgical units of ahmadu bello university teaching hospital, zaria, nigeria: an indication for effective control measures. Ann. Afr. Med., 3: 13-16.
6. Zuanazzi D, Souto R, Mattos M, Zuanazzi M, Tura B, Sansone C, Colombo A (2010). Prevalence of potential bacterial respiratory pathogens in the oral cavity of hospitalised individuals. Arch. Oral. Biolo., 55: 21-28.
7. Scarff J, Goldberg J (2008). Vaccination against Pseudomonas aeruginosa pneumonia in immunocompromised mice. Clin. Vac. Immunol., 15: 367-375.
8. Wroblewska M, Rudnicka J, Marchel H, Luczak M (2006). Multidrugresistant bacteria isolated from patients hospitalised in Intensive Care Units. Int. J. Antimicrob. Agen., 27: 285-289.
9. Marcel J, Alfa M, Baquero F, Etienne J, Goossens H, Harbarth S, Hryniewicz W, Jarvis W, Kaku M, Leclercq R (2008). Healthcareassociated infections: think globally, act locally. Clin. Microbiol. Infect., 14: 895-907.
10. Masood S (2010). In Vitro Susceptibility Test of Different Clinical Isolates against Ceftriaxone. Oma. Med. J., 25: 199-202.
11. Jombo G, Akpan S, Epoke J, Denen A, Odey F (2010). Multidrug resistant Psudomonas aeruginosa infections complicating surgical wounds and the potential challenges in managing post-operative wound infections: University of Calabar Teaching Hospital experience. Asi. Pac. J. Trop. Med., 3: 479-482.
12. Strateva T, Yordanov D (2009). Pseudomonas aeruginosa–A phenomenon of bacterial resistance. Journal of Medical Microbiology. 58(9): 1133-1148.