Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.53555/rxsa0808

ANTIBIOTIC RESISTANCE PROFILES OF MULTIDRUG-RESISTANT PSEUDOMONAS AERUGINOSA IN CLINICAL ISOLATES: A PROSPECTIVE STUDY FROM JINNAH POSTGRADUATE MEDICAL CENTRE, PAKISTAN

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ABSTRACT

Objective: Pseudomonas aeruginosa is a major pathogen causing hospital-acquired infections worldwide, with multidrug-resistant (MDR) strains posing a significant public health challenge. This study aimed to investigate the antibiotic resistance profiles of P. aeruginosa isolates from diverse clinical samples to guide effective treatment strategies.

Methodology: This prospective study was conducted at the Clinical Pathology Laboratory, Jinnah Postgraduate Medical Centre (JPMC), Karachi, Pakistan, from April 2020 to December 2020. P. aeruginosa isolates from various clinical specimens were analyzed along with demographic details. Bacterial identification and antimicrobial susceptibility testing were performed using standard microbiological techniques and the Vitek 2 system. Data analysis was carried out using R Studio.

Results: A total of 1066 P. aeruginosa isolates were identified, with the highest frequency (33%) in individuals under 20 years of age. The majority of isolates came from pus (47%) and urine (23%) samples. High resistance rates were observed for amikacin (77%), ceftizoxime (43%), and norfloxacin (40%). In contrast, cefoperazone-sulbactam exhibited the lowest resistance at 15%.

Conclusion: The findings emphasize the need for antibiotic susceptibility testing in managing P. aeruginosa infections. Although amikacin and tobramycin showed high sensitivity, increasing

resistance trends for gentamicin and amikacin warrant careful antibiotic selection and monitoring. Treatment should be tailored based on local resistance data, as resistance patterns vary for antibiotics such as piperacillin-tazobactam and imipenem. Regular surveillance of resistance trends is crucial to ensure effective treatment strategies.

Keywords: Multidrug-Resistant Pseudomonas Aeruginosa, Antibiotic Resistance, Clinical Isolates, prospective Analysis

INTRODUCTION

Pseudomonas aeruginosa is a notable pathogen recognized for its intrinsic and acquired resistance to multiple antibiotics, which poses significant challenges in clinical settings. This Gram-negative bacterium is responsible for a range of infections, particularly in immunocompromised patients, such as those with cystic fibrosis, burn wounds, and other underlying health conditions [1]. The ability of P. aeruginosa to thrive in diverse environments, coupled with its complex resistance mechanisms, makes it a critical focus in the study of antibiotic resistance [2]. The resistance of P. aeruginosa to antibiotics is attributed to several factors, including the presence of efflux pumps, alterations in porin channels, and the production of β-lactamases [3]. Carbapenem resistance in P. aeruginosa due to the production of metallo-beta-lactamases is an increasing global concern, leading to a reduction in available therapeutic options [4]. These mechanisms not only diminish the efficacy of common antibiotic treatments but also facilitate the emergence of multidrug-resistant (MDR) strains, complicating therapeutic options [5]. The rise of MDR P. aeruginosa is alarming, as it contributes to increased morbidity and mortality rates, prolonged hospital stays, and higher healthcare costs [6]. Recent studies highlight the urgent need for novel antimicrobial strategies and the importance of antimicrobial stewardship to combat the growing threat of antibiotic resistance in P. aeruginosa [7].

In developing countries, where factors such as limited access to healthcare, inadequate regulatory frameworks, and the over-the-counter availability of antibiotics contribute to the problem. In many of these regions, antibiotics are often used indiscriminately for self-medication or as a first-line treatment for infections without proper medical supervision. This misuse accelerates the development of resistant strains of bacteria, including Pseudomonas aeruginosa, which is frequently isolated in hospital settings and linked to high morbidity and mortality rates [8]. The lack of robust antimicrobial stewardship programs further exacerbates this issue, as healthcare facilities often struggle to implement effective infection control measures. Additionally, public health infrastructures may be under-resourced, limiting surveillance efforts that could identify and monitor resistance patterns. The situation is compounded by the rising burden of non-communicable diseases, which shifts healthcare resources away from tackling infectious diseases. Without coordinated global and local efforts to address antibiotic resistance, developing countries face the dual threat of increasing infection rates and diminishing treatment options, leading to a public health crisis that could have far-reaching consequences [9].

In Pakistan, the situation is more critical due to a combination of factors, including over-the-counter availability of antibiotics, lack of strict regulations on antibiotic use, and insufficient infection control measures in healthcare settings. P. aeruginosa is a common pathogen in hospitals across the country, often leading to nosocomial infections, particularly in intensive care units (ICUs). The lack of proper antimicrobial stewardship programs, coupled with poor hygiene and sanitation standards, exacerbates the spread of MDR strains. Recent studies from tertiary care hospitals in Pakistan indicate alarmingly high rates of resistance to carbapenems, one of the last-resort classes of antibiotics, underscoring the urgent need for comprehensive policies to control antibiotic misuse and prevent the further rise of MDR pathogens (Cheesman, Ilanko et al. 2017) [10].

METHODOLOGY

This Prospective study was conducted at the Clinical Pathology Laboratory, Jinnah Postgraduate Medical Centre (JPMC), Karachi, Pakistan, using data from April 2020 to December 2020 on *Pseudomonas aeruginosa*.

Approval for the study was obtained from the Research and Ethics Committee of JPMC. Data on *P. aeruginosa* isolates from various clinical samples including collected between April 2020 to December 2020 were retrieved from the computerized system and microbiology laboratory records. The dataset included the patients' sociodemographic characteristics as well as the antibiotic resistance patterns of *P. aeruginosa*.

Bacterial identification method and antibiotic susceptibility testing

Microbiological specimens such as swab, pus, blood, urine, sputum and others clinical samples received and processed at the Clinical Pathology Lab Microbiology laboratory JPMC. Specimens were microscopically observed after staining with Gram stain and cultured by plating on MacConkey agar (Oxoid, UK) and incubated at 37°C. The isolates were identified by the standard microbiological methods and the automated Vitek 2 system (Bio-Mérieux, France). Antimicrobial susceptibility testing was performed using Kirby-Bauers disc diffusion method on Muller-Hinton agar with the following antibiotic panel by using Oxoid antibiotic discs: Gentamicin (10 μg), ciprofloxacin (5 μg), piperacillin/tazobactam (100/10 μg), imipenem (10 μg), meropenem (10 μg), amikacin (10 μg), ceftazidime (CAZ) (30 μg), and colistin (10 μg). For CTAZ, a minimum inhibitory concentration was determined by the Epsilometer (E) test, as recommended by Clinical Laboratory Standards Institute. The strains that show resistance to at least one drug in three or more classes of antibiotics were categorized as MDR organisms. Quality control was performed using *P. aeruginosa* ATCC 27853.

Statistical analysis

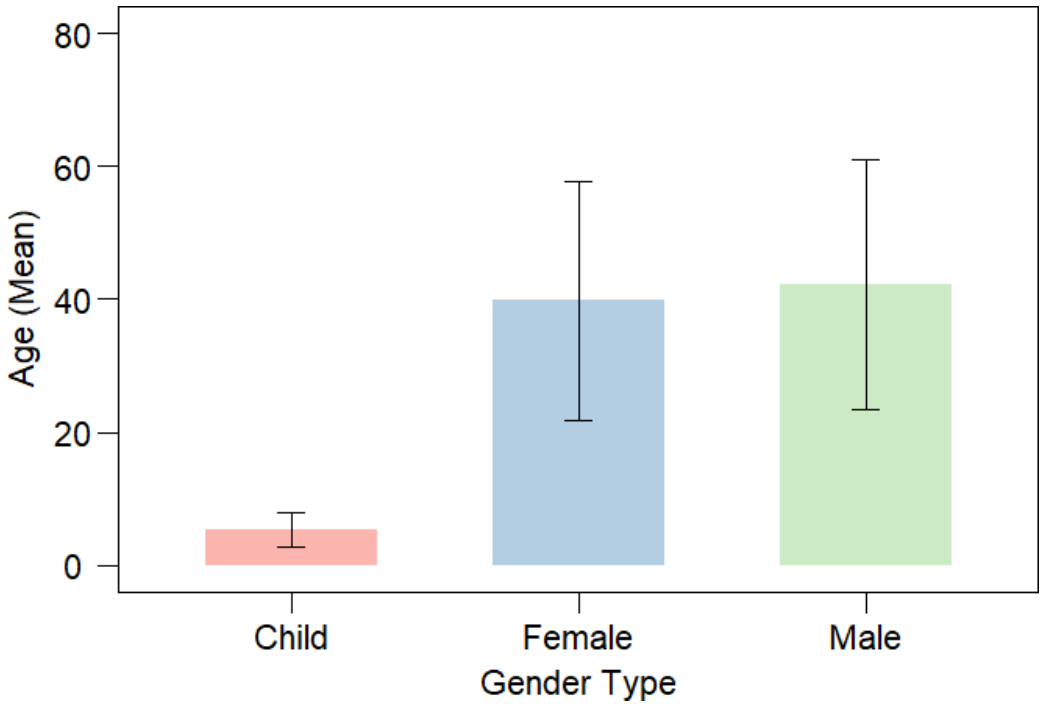
The data was systematically analyzed using R studio **(2024.04.0).** Frequencies of gender and age groups were checked both in total and month wise. One sample Chi-square test was done to find the significance of mode values among the months and antibiotics. Sample wise isolates were classified for descriptive and subjected to the Pearson Chi-square test. Month-wise antibiotic susceptibility pattern was arranged and the trends plotted for the period April 2020 to December 2020. $P < 0.05$ was indicative of deviation from expected frequencies.

RESULTS

A total of 1,066 pseudomonas *aeruginosa* positive patients were included in the study. The median age of the patients was 32 years (range: 17–52 years), with a notable variation across the months. The median age ranged from a low of 20 years in August to a high of 41 years in December. A statistically significant difference in age distribution was observed across the months, as indicated by a p-value of <0.001.

The mean age for male patients was 42.2 years with a standard deviation of 18.9, while for female patients; the mean age was 39.8 years with a standard deviation of 17.9. Among pediatric patients (children under 10 years), the mean age was 5.36 years with a standard deviation of 2.66 [Figure 1A].

Figure 1. Distribution of P. aeruginosa **A**) Gender and age mean, ((**B**) Age Distribution, (**C**) Culture Sample distribution, (**D**) Month distribution of the samples. (**E**) Department distribution of the population



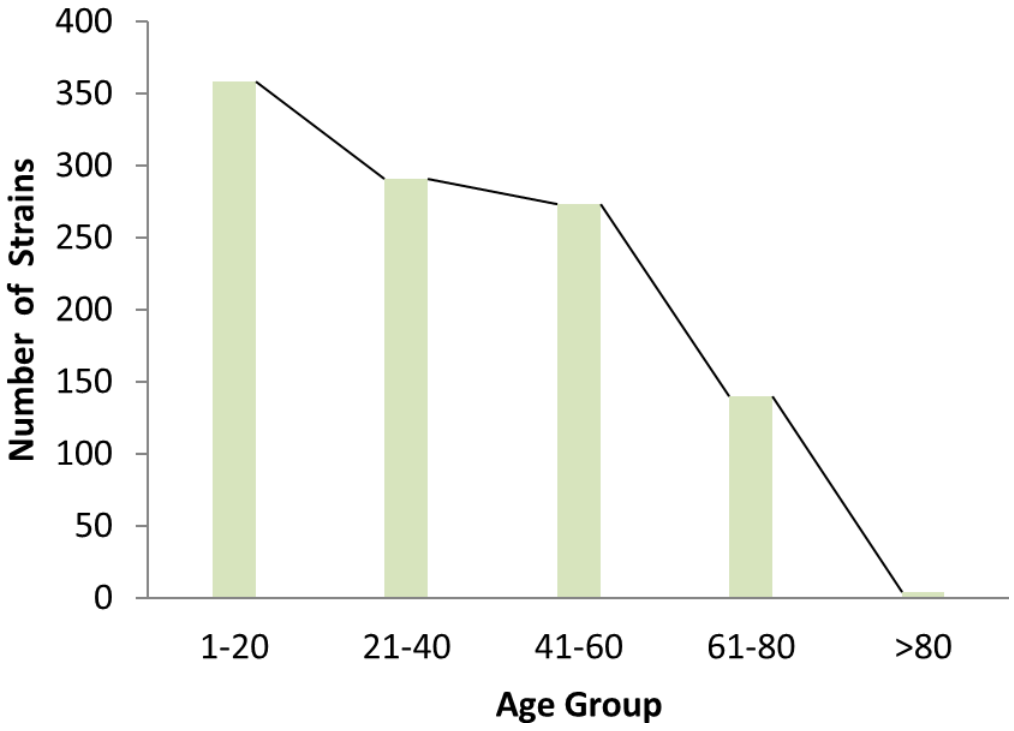
When analyzing gender distribution, 17% of the patients were children, with significant variability in the proportion of children month by month, ranging from 6.1% in December to 36% in August (p-value <0.001). Female patients made up 36% of the total cohort, with monthly variations from 5.3% in February to 47% in September. Male patients comprised 47% of the cohort overall, with the highest proportion observed in December (62%) and the lowest in August (31%).
These findings suggest significant differences in the demographic composition of patients across the months, both in terms of age and gender, further confirmed by the highly significant p-values for both characteristics (<0.001) [Table 1].

Table 1: Sociodemographic Characteristics of Patients with Pseudomonas Isolates

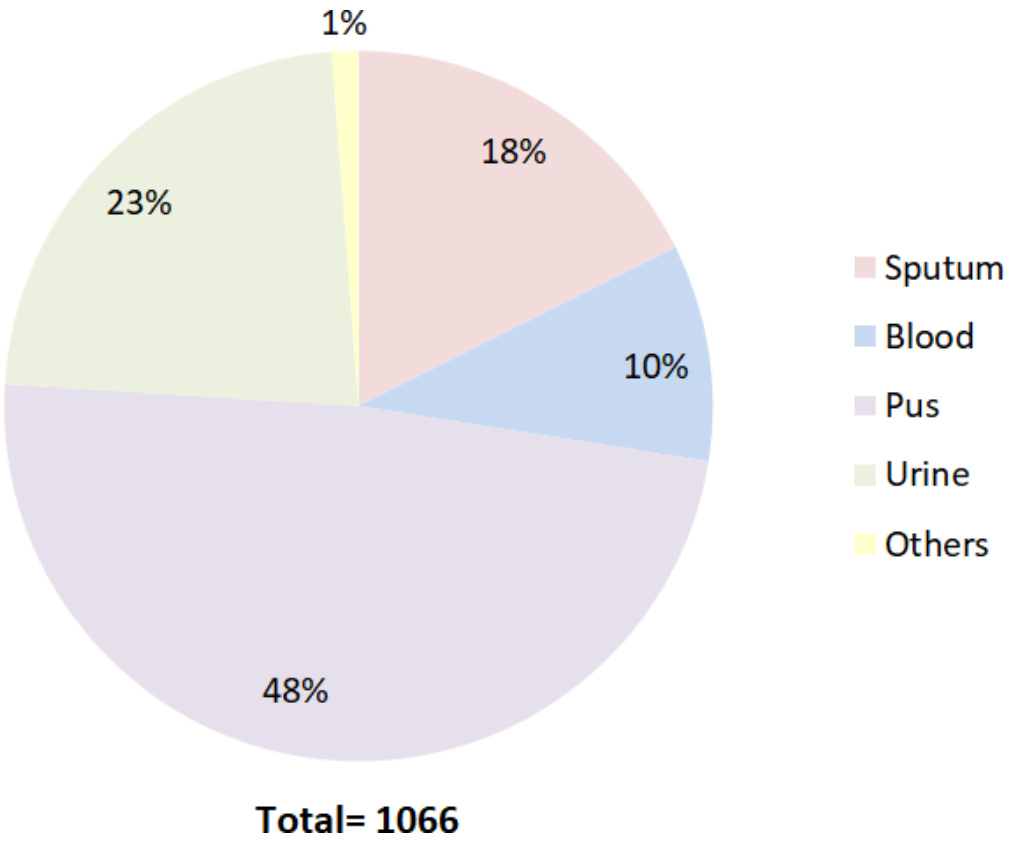
Characteristic	Overall	April	May	June	July	August	September	October	November	December	p-value[2]
	N = 1,066	N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	
Age[1]	32 (17, 52)	30 (18, 57)	30 (7, 54)	32 (20, 51)	35 (9, 65)	20 (6, 38)	37 (23, 55)	34 (20, 45)	38 (24, 48)	41 (25, 55)	<0.001
Gender											<0.001
Child	185 (17%)	13 (17%)	43 (33%)	11 (8.3%)	29 (27%)	58 (36%)	13 (7.8%)	8 (8.6%)	4 (4.0%)	6 (6.1%)	
Female	384 (36%)	4 (5.3%)	41 (31%)	59 (45%)	36 (34%)	53 (33%)	79 (47%)	43 (46%)	38 (38%)	31 (32%)	
Male	497 (47%)	58 (77%)	48 (36%)	62 (47%)	42 (39%)	51 (31%)	75 (45%)	42 (45%)	58 (58%)	61 (62%)	

[1] **Median (Q1, Q3); n (%)**
[2] **Kruskal-Wallis rank sum test; Pearson's Chi-squared test**

Among different age groups, *P. aeruginosa* was more frequently isolated from under 20 years (33%) , 21-40 years (27%) , 31-60 years (25%) , 61-80 years (13%) and 80+ (0.38%) ($P < 0.001$)[1B].



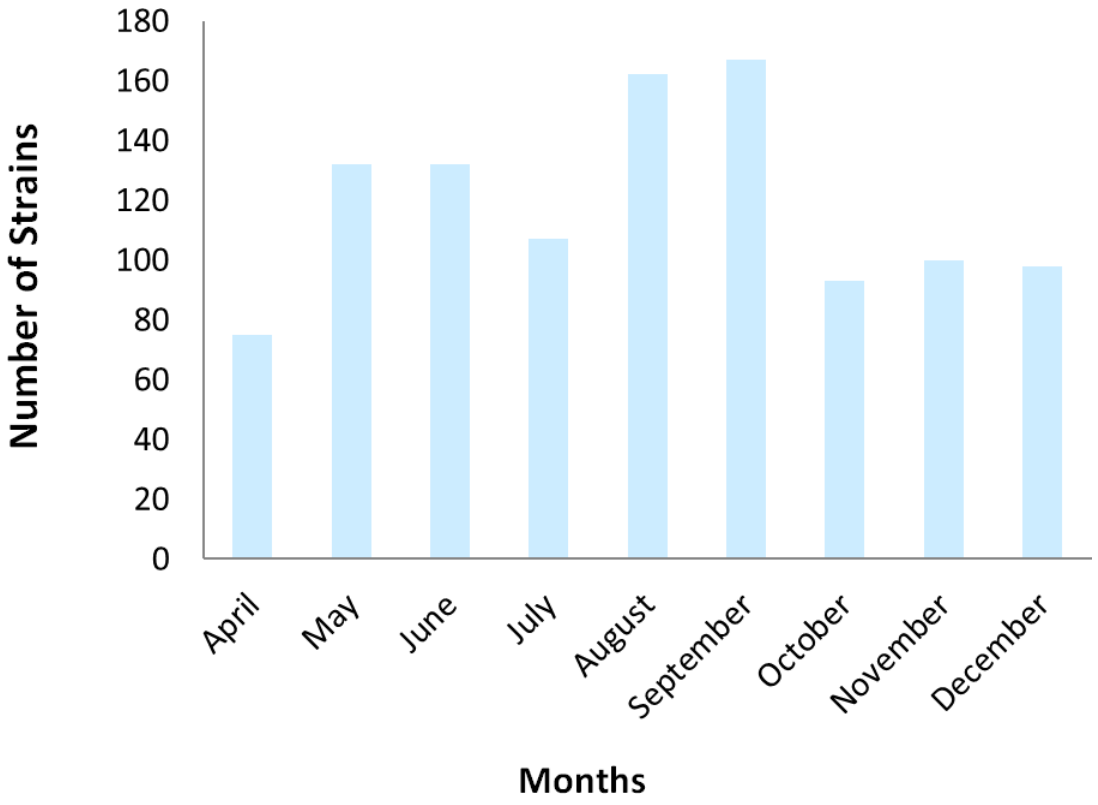
With respect to the source, *P. aeruginosa* was predominantly isolated from Pus (48%), urine (23%), sputum (18%), blood (10%) and other (1%) [Figure 1C].



Temporal Changes in P. Aeruginosa Isolates

The monthly distribution of *Pseudomonas aeruginosa* positive samples revealed notable variations throughout the year. In April, the prevalence of positive cases was relatively low, accounting for 7.04% of the total. This was followed by a significant rise in May and June, where the proportion of positive samples reached 12.38% in both months. In July, the rate slightly decreased to 10.04%, but a sharp increase was observed in August, with 15.20% of samples testing positive. September saw the highest rate of *Pseudomonas aeruginosa* positivity at 15.67%, indicating a potential peak in late summer and early autumn. The trend reversed in the following months, with October showing a decrease to 8.72%, November to 9.38%, and December closing the year with a rate of 9.19%.

This distribution suggests a seasonal pattern, with higher prevalence in the late summer and early autumn months, potentially due to environmental or epidemiological factors influencing bacterial transmission and infection rates. These findings highlight the importance of monitoring *Pseudomonas aeruginosa* trends throughout the year to better understand and manage the risks associated with this pathogen. (Figure 1D).

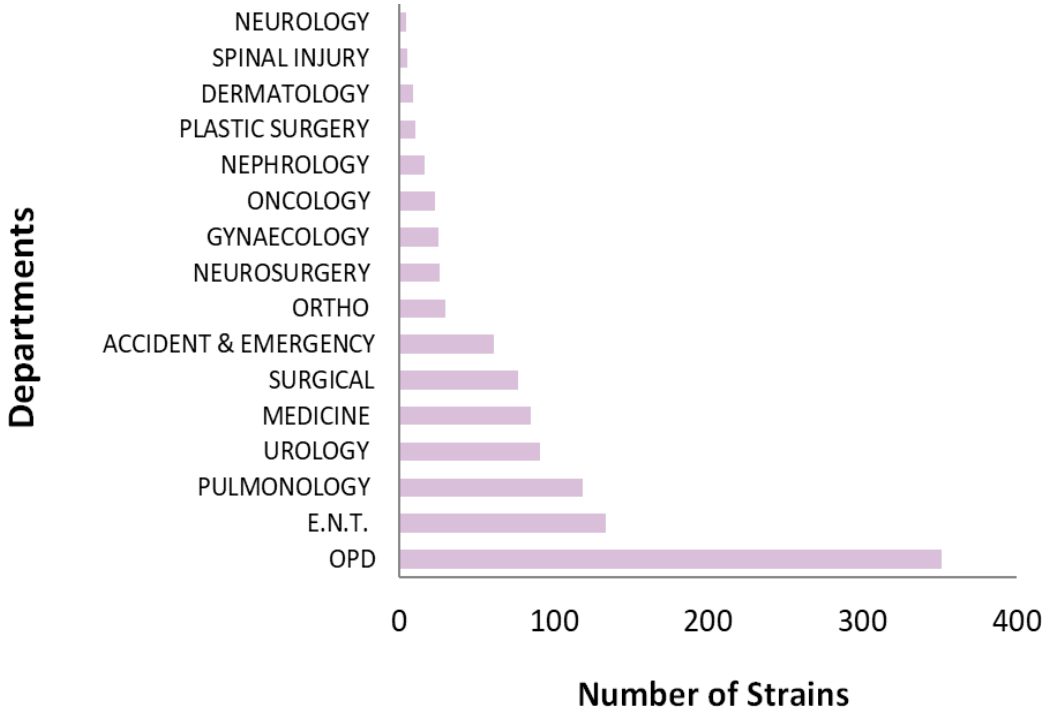


Departmental Distribution of Pseudomonas aeruginosa

The distribution of *Pseudomonas aeruginosa* positive samples across various hospital departments showed distinct patterns. The highest percentage of positive cases was recorded in the Outpatient Department (OPD), accounting for 32.93% of the total, which indicates a significant proportion of cases being managed outside of specialized wards. The Ear, Nose, and Throat (E.N.T.) department followed with 12.57%, reflecting a high incidence of infections in this area. Pulmonology also showed a considerable rate, with 11.16% of samples testing positive, which could be related to respiratory complications commonly associated with *Pseudomonas aeruginosa* infections.

Urology and Surgical departments contributed 8.54% and 7.22%, respectively, to the total number of positive cases, emphasizing the need for vigilance in postoperative and urinary tract-related infections. The Medicine department had a positive rate of 7.97%, while Accident & Emergency accounted for 5.72%, potentially due to the acute nature of infections presented in these settings.

Other departments such as Gynaecology (2.35%), Neurosurgery (2.44%), and Oncology (2.16%) had lower rates but still represent significant areas of concern. Orthopaedics (2.81%) and Plastic Surgery (0.94%) departments showed moderate to low positivity, likely reflecting their patient populations' specific vulnerability to infections. Nephrology (1.50%) and Dermatology (0.84%) had comparatively lower rates, with Neurology (0.38%) and Spinal Injury (0.47%) contributing minimally to the total cases [Figure 1E].



This distribution highlights the diversity in the clinical settings where *Pseudomonas aeruginosa* is encountered, with a particularly high burden observed in OPD, respiratory, and surgical-related departments. These findings stress the importance of department-specific infection control measures to manage the spread of this pathogen effectively.

Antibiotic Resistance and Sensitivity in Pseudomonas aeruginosa

Amikacin showed strong activity against *Pseudomonas aeruginosa*, with a high susceptibility rate of 77% (812/1055), indicating that the majority of isolates remained sensitive to this antibiotic. Intermediate resistance (I) was observed in only 3% of cases (27/1055), while resistance (R) was noted in 20% (216/1055), showing a notable minority of strains resistant to this commonly used aminoglycoside. Resistance remained consistent throughout the months, peaking in May and July at 25%, with the lowest rate recorded in April at 15%. Gentamicin showed lower efficacy compared to amikacin, with only 67% (269/401) of isolates sensitive to it. Intermediate resistance was rare (2%, 8/401), but resistance was more prevalent, observed in 31% (124/401) of isolates, peaking in July at 44%. This indicates a concerning trend of resistance to gentamicin, an important antibiotic for treating *Pseudomonas* infections. Tobramycin demonstrated high efficacy, with 78% (521/667) of isolates sensitive. The intermediate resistance rate was minimal (1%, 4/667), and resistance was observed in 21% (142/667), with July and August showing the highest rates (26% and 22%, respectively).

Table 2: Month wise antibiotic resistance pattern of Pseudomonas Isolates: from 2a-2j

Table 2a: Amikacin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	2 (2.8%)	2 (1.5%)	5 (3.8%)	3 (2.8%)	3 (1.9%)	3 (1.8%)	2 (2.2%)	7 (7.0%)	0 (0%)	27 (3%)
R	11 (15%)	23 (18%)	25 (19%)	25 (23%)	36 (22%)	35 (21%)	16 (17%)	25 (25%)	20 (20%)	216 (20%)
S	58 (82%)	105 (81%)	100 (77%)	79 (74%)	123 (76%)	126 (77%)	75 (81%)	68 (68%)	78 (80%)	812 (77%)

Table 2b: Gentamicin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	0 (0%)	4 (3.8%)	2 (2.0%)	1 (3.1%)	0 (0%)	0 (0%)	0 (0%)	1 (4.0%)	0 (0%)	8 (2%)
R	2 (100%)	22 (21%)	26 (26%)	14 (44%)	17 (37%)	16 (44%)	6 (40%)	10 (40%)	11 (28%)	124 (31%)
S	0 (0%)	80 (75%)	72 (72%)	17 (53%)	29 (63%)	20 (56%)	9 (60%)	13 (52%)	28 (72%)	269 (67%)

Table 2c: Tobramycin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	0 (0%)	0 (0%)	0 (0%)	1 (1.2%)	1 (0.8%)	0 (0%)	0 (0%)	2 (2.6%)	0 (0%)	4 (1%)
R	12 (18%)	0 (0%)	4 (13%)	21 (26%)	28 (22%)	29 (23%)	17 (21%)	19 (24%)	12 (18%)	142 (21%)
S	56 (82%)	0 (0%)	28 (88%)	60 (73%)	100 (78%)	99 (77%)	65 (79%)	57 (73%)	56 (82%)	521 (78%)

Table 2d: Piperacillin+Tazobactam

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	1 (1.5%)	4 (9.5%)	4 (3.1%)	4 (3.7%)	6 (3.7%)	6 (3.6%)	4 (4.3%)	10 (10%)	5 (5.1%)	44 (5%)
R	11 (17%)	2 (4.8%)	19 (15%)	20 (19%)	28 (17%)	36 (22%)	13 (14%)	28 (28%)	20 (20%)	177 (18%)
S	53 (82%)	36 (86%)	107 (82%)	83 (78%)	128 (79%)	123 (75%)	76 (82%)	62 (62%)	73 (74%)	741 (77%)

Table 2e: Ciprofloxacin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	1 (1.7%)	3 (2.3%)	1 (0.8%)	1 (1.0%)	4 (2.5%)	1 (0.6%)	2 (2.2%)	4 (4.1%)	1 (1.1%)	18 (2%)

R	20 (33%)	38 (29%)	37 (28%)	32 (31%)	43 (27%)	45 (28%)	26 (28%)	38 (39%)	29 (31%)	308 (30%)
S	39 (65%)	88 (68%)	92 (71%)	71 (68%)	113 (71%)	116 (72%)	65 (70%)	55 (57%)	64 (68%)	703 (68%)

Table 2f: Norfloxacin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0

Table 2g: Meropenem

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	1 (1.3%)	2 (1.5%)	2 (1.5%)	2 (1.9%)	2 (1.2%)	1 (0.6%)	1 (1.1%)	4 (4%)	1 (1%)	16 (1%)
R	4 (5.3%)	5 (3.8%)	7 (5.3%)	7 (6.5%)	10 (6.2%)	9 (5.4%)	6 (6.5%)	10 (10%)	5 (5%)	63 (6%)
S	70 (93%)	125 (95%)	123 (93%)	98 (92%)	150 (93%)	157 (94%)	86 (92%)	86 (86%)	92 (94%)	987 (93%)

Table 2h: Imipenem

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	2 (2.7%)	2 (1.5%)	3 (2.3%)	3 (2.8%)	3 (1.9%)	2 (1.2%)	2 (2.2%)	4 (4%)	1 (1%)	22 (2%)
R	5 (6.7%)	7 (5.3%)	8 (6.1%)	10 (9.3%)	12 (7.4%)	11 (6.6%)	5 (5.4%)	8 (8%)	7 (7%)	73 (7%)
S	68 (91%)	123 (93%)	121 (92%)	94 (88%)	147 (91%)	154 (92%)	86 (92%)	88 (88%)	90 (92%)	971 (91%)

Table 2i: Levofloxacin

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	0 (0%)	1 (0.8%)	2 (1.5%)	2 (1.9%)	1 (0.6%)	1 (0.6%)	1 (1.1%)	2 (2%)	1 (1%)	11 (1%)
R	19 (25%)	38 (29%)	41 (31%)	35 (33%)	49 (30%)	47 (28%)	29 (31%)	40 (40%)	35 (36%)	333 (31%)
S	56 (75%)	93 (70%)	89 (67%)	70 (65%)	112 (69%)	119 (71%)	63 (68%)	58 (58%)	62 (63%)	722 (68%)

Table 2j: Cefepime

Antibiotics	April	May	June	July	August	September	October	November	December	Total
N = 75	N = 132	N = 132	N = 107	N = 162	N = 167	N = 93	N = 100	N = 98	N = 1066	
I	1 (1.3%)	2 (1.5%)	3 (2.3%)	3 (2.8%)	2 (1.2%)	2 (1.2%)	2 (2.2%)	3 (3%)	2 (2%)	20 (2%)
R	6 (8%)	7 (5.3%)	8 (6.1%)	10 (9.3%)	12 (7.4%)	11 (6.6%)	7 (7.5%)	12 (12%)	10 (10%)	83 (8%)
S	68 (91%)	123 (93%)	121 (92%)	94 (88%)	148 (91%)	154 (92%)	84 (90%)	85 (85%)	86 (88%)	983 (92%)

Overall, tobramycin remains a reliable option for treating *Pseudomonas* infections, though vigilance is needed for emerging resistance. Piperacillin-tazobactam showed good efficacy, with 77% (741/962) of isolates sensitive. Intermediate resistance was seen in 5% (44/962), while 18% (177/962) were resistant, with resistance trends fluctuating, peaking in November at 28% and July at 19%. This combination remains crucial in therapy but highlights the importance of continued monitoring for resistance trends. Ciprofloxacin displayed moderate activity, with 68% (703/1029) of isolates sensitive. Intermediate resistance was 2% (18/1029), while 30% (308/1029) of isolates were resistant. Resistance showed a steady presence, with peaks in November (39%) and April (33%), emphasizing a significant level of resistance that may impact its use as a frontline treatment for *Pseudomonas* infections.

Norfloxacin had a smaller sample size, revealing considerable resistance, with 40% (28/70) of isolates resistant, while the remaining 60% (42/70) were sensitive. Ceftazidime, a commonly used beta-lactam, retained significant efficacy, with 75% (552/740) of isolates sensitive. Intermediate resistance was rare (1%, 10/740), while 24% (178/740) were resistant, peaking in July and November at 24% and 28%, respectively. These results indicate that ceftazidime remains effective, though emerging resistance requires attention. Cefepime showed a lower sensitivity rate, with 66% (455/688) of isolates sensitive. Intermediate resistance was noted in 4% (29/688), while 30% (204/688) of isolates were resistant, peaking in July and November at 33%.

These findings highlight a concerning resistance level that may limit cefepime's effectiveness in treating *Pseudomonas* infections. Imipenem displayed strong efficacy, with 77% (438/568) of isolates sensitive. Intermediate resistance was low (2%, 12/568), while 21% (118/568) of isolates were resistant, with peaks in May (24%) and November (23%).

DISCUSSION

The resistance mechanisms of *Pseudomonas aeruginosa* are complex, encompassing both intrinsic traits and acquired resistance factors. Key intrinsic mechanisms include efflux pumps and modifications of porin channels, which contribute significantly to the organism's ability to evade the effects of various antibiotics. Moreover, the acquisition of resistance genes frequently occurs through horizontal gene transfer, complicating the management of infections caused by this pathogen 11].

In Pakistan, there is a concerning trend of increasing multidrug-resistant (MDR) and extensively drug-resistant (XDR) *P. aeruginosa* strains. This rise is often attributed to the inappropriate use of antibiotics and inadequate infection control measures in healthcare settings [12]. The situation is further exacerbated by the limited availability of effective antibiotics and the slow pace of new drug development, which together contribute to treatment failures and rising healthcare costs [13]. Therefore, understanding the epidemiology and resistance mechanisms of *P. aeruginosa* in Pakistan is critical to addressing this public health challenge.

Effective antimicrobial stewardship and enhanced infection control practices are vital to mitigate the threat posed by *P. aeruginosa*. The ability of *P. aeruginosa* to colonize medical equipment surfaces and form highly resistant biofilms significantly complicates treatment options. Additionally, its rapid mutation rate facilitates the acquisition of drug resistance, making it a leading cause of nosocomial

infections. The widespread misuse of antibiotics has resulted in an alarming increase in drug-resistant bacterial infections, leading to more severe illnesses and higher mortality rates.
In our analysis of 1,066 *P. aeruginosa*-positive patients, we found a median age of 32 years, with significant monthly variation ranging from 20 years in August to 41 years in December ($p < 0.001$). Male patients had a mean age of 42.2 years, while females had a mean age of 39.8 years. Notably, pediatric patients (under 10 years) had a mean age of 5.36 years. The gender distribution varied, with children accounting for 17%, females for 36%, and males for 42%. Similar to other studies, the prevalence of males was higher than that of females [14,15] . Noteworthy monthly gender variations were observed, such as the low representation of females at 5.3% in February compared to 47% in September.

Among different age groups, *P. aeruginosa* was most frequently isolated in individuals under 20 years (33%), followed by those aged 21–40 years (27%), 31–60 years (25%), and 61–80 years (13%). The bacteria were predominantly isolated from pus (48%), urine (23%), sputum (18%), and blood (10%) similar to previous study [16]. The monthly data revealed a seasonal pattern in *P. aeruginosa* prevalence, with the highest positivity rates in September (15.67%) and the lowest in April (7.04%). Positive cases were most frequently recorded in the Outpatient Department (32.93%), followed by the ENT (12.57%) and Pulmonology (11.16%) departments, while Urology, Surgery, Medicine, and other departments contributed lower but clinically significant proportions.
The analysis of antibiotic susceptibility patterns for *Pseudomonas aeruginosa* isolates collected from April to December 2021 provides critical insights into resistance trends that are essential for informing clinical practices and treatment guidelines. Notably, amikacin exhibited a high sensitivity rate of 77% (812/1055), making it one of the most effective antibiotics against *Pseudomonas aeruginosa* in this study. The resistance rate of 20% (216/1055) indicates that while amikacin remains a reliable therapeutic option, increasing resistance trends cannot be overlooked. The slight uptick in resistance during specific months, particularly in July and November, may be attributed to seasonal variations in infection patterns, changes in antibiotic usage, or the potential emergence of resistant strains within the hospital environment. This underscores the importance of continuous surveillance and the need for an adaptive approach to antibiotic stewardship.

Gentamicin, in contrast, presented a lower sensitivity rate of 67% (269/401) and a notable resistance rate of 31% (124/401). This decline in efficacy suggests that gentamicin may not be the optimal choice for treating *Pseudomonas aeruginosa* infections, especially given the significant peaks in resistance observed in July (44%) and September (40%). Such trends highlight the necessity for clinicians to consider alternative therapies or combination treatments to mitigate the risk of treatment failure. The resistance profile observed here aligns with global reports of increasing gentamicin resistance in hospital-acquired infections, raising concerns about its future utility in clinical settings.
Tobramycin exhibited a sensitivity rate of 78% (521/667), which is comparable to that of amikacin, with a resistance rate of 21%. The relatively high susceptibility of tobramycin suggests it remains a viable option for treating *Pseudomonas aeruginosa* infections, particularly in cases where other treatments may fail. However, similar to amikacin, the emergence of resistance necessitates ongoing monitoring to ensure the antibiotic's effectiveness over time.

Other antibiotics, including piperacillin-tazobactam, ciprofloxacin, and imipenem, were also assessed, revealing varying susceptibility and resistance patterns that reflect the complex and dynamic nature of *Pseudomonas aeruginosa* resistance. The results indicate that while certain antibiotics maintain efficacy, the rising resistance rates necessitate vigilance in clinical practice.

CONCLUSION

This study finding highlights the critical importance of antibiotic susceptibility testing in managing *Pseudomonas aeruginosa* infections. Among the antibiotics analyzed, amikacin and tobramycin

demonstrated the highest sensitivity rates, making them the most effective options for treating these infections within the studied timeframe. However, the notable resistance rates particularly for gentamicin and increasing trends in resistance for amikacin, highlight the urgent need for vigilant antibiotic stewardship and continuous monitoring of resistance patterns.

Piperacillin-tazobactam and imipenem also showed variable susceptibility, further indicating that treatment choices should be tailored based on local resistance profiles. Given the dynamic nature of antibiotic resistance, clinicians must remain adaptable, employing susceptibility data to guide empirical therapies effectively.

Ethical approval: The study was conducted from April 2020 to December 2020 and Ethical approval for the study was obtained from the Clinical Pathology Laboratory, Jinnah Postgraduate Medical Centre (JPMC), Karachi, Pakistan.

Author's Contributions: All authors were involved in conducting the study and have reviewed and approved the final version of the manuscript.

Conflict of Interest: The authors declare that there are no conflicts of interest associated with this study.

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

REFERENCES

1. Malhotra, S., et al. (2019). Cystic fibrosis and Pseudomonas aeruginosa: The host-microbe interface. Clinical Microbiology Reviews, 32(3). https://doi.org/10.1128/CMR.00138-18
2. Pachori, P., et al. (2019). Emergence of antibiotic resistance Pseudomonas aeruginosa in intensive care unit: A critical review. Genes & Diseases, 6(2), 109-119. https://doi.org/10.1016/j.gendis.2019.04.001
3. Langendonk, R. F., et al. (2021). The building blocks of antimicrobial resistance in Pseudomonas aeruginosa: Implications for current resistance-breaking therapies. Frontiers in Cellular and Infection Microbiology, 11, 665759. https://doi.org/10.3389/fcimb.2021.665759
4. El-Far, A., et al. (2021). High rates of aminoglycoside methyltransferases associated with metallo-beta-lactamases in multidrug-resistant and extensively drug-resistant Pseudomonas aeruginosa clinical isolates from a tertiary care hospital in Egypt. Infection and Drug Resistance, 14, 4849-4858. https://doi.org/10.2147/IDR.S332333
5. Terreni, M., Taccani, M., & Pregnolato, M. (2021). New antibiotics for multidrug-resistant bacterial strains: Latest research developments and future perspectives. Molecules, 26(9), 2671. https://doi.org/10.3390/molecules26092671
6. Morris, S., & Cerceo, E. (2020). Trends, epidemiology, and management of multi-drug resistant gram-negative bacterial infections in the hospitalized setting. Antibiotics, 9(4), 196. https://doi.org/10.3390/antibiotics9040196
7. Majumder, M. A., Rahman, S., Cohall, D., Bharatha, A., Singh, K., Haque, M., & Gittens-St Hilaire, M. (2020). Antimicrobial stewardship: Fighting antimicrobial resistance and protecting global public health. Infection and Drug Resistance, 13, 4713-4738. https://doi.org/10.2147/IDR.S242852
8. Ahmad, I., Malak, H. A., & Abulreesh, H. H. (2021). Environmental antimicrobial resistance and its drivers: A potential threat to public health. Journal of Global Antimicrobial Resistance, 27, 101-111. https://doi.org/10.1016/j.jgar.2021.08.005
9. Salam, M. A., Al-Amin, M. Y., Salam, T. M., Pawar, J. S., Akhter, N., Rabaan, A. A., & Alqumber, M. A. (2023). Antimicrobial resistance: A growing serious threat for global public health. Healthcare, 11(13), 1946. https://doi.org/10.3390/healthcare11131946

10. Cheesman, M. J., et al. (2017). Developing new antimicrobial therapies: Are synergistic combinations of plant extracts/compounds with conventional antibiotics the solution? Pharmacognosy Reviews, 11(22), 57. https://doi.org/10.4103/phrev.phrev_21_17
11. Nnadozie, C. F., Kumari, S., & Bux, F. (2017). Status of pathogens, antibiotic resistance genes, and antibiotic residues in wastewater treatment systems. Reviews in Environmental Science and Bio/Technology, 16(3), 491-515. https://doi.org/10.1007/s11157-017-9440-9
12. Magiorakos, A. P., Burns, K., Rodríguez Baño, J., Borg, M., Daikos, G., Dumpis, U., & Weber, J. T. (2017). Infection prevention and control measures and tools for the prevention of entry of carbapenem-resistant Enterobacteriaceae into healthcare settings: Guidance from the European Centre for Disease Prevention and Control. Antimicrobial Resistance & Infection Control, 6, 1-17. https://doi.org/10.1186/s13756-016-0160-1
13. Pisabarro, A. G., & de la Torre, D. P. R. (2020). Impact of access to medicines on public health: Problems associated with access, use, and abuse of antibiotics. Antimicrobial Resistance and Infection Control. https://doi.org/10.1007/s00248-020-01465-9
14. Fenta, A., Dagnew, M., Eshetie, S., & Belachew, T. (2020). Bacterial profile, antibiotic susceptibility pattern and associated risk factors of urinary tract infection among clinically suspected children attending at Felege-Hiwot comprehensive and specialized hospital, Northwest Ethiopia. A prospective study. BMC Infectious Diseases, 20, 1-10. https://doi.org/10.1186/s12879-020-05262-5
15. Almalki, M. A., & Varghese, R. (2020). Prevalence of catheter-associated biofilm-producing bacteria and their antibiotic sensitivity pattern. Journal of King Saud University-Science, 32(2), 1427-1433. https://doi.org/10.1016/j.jksus.2019.11.008
16. Singh, I., Jaryal, S. C., Thakur, K., Sood, A., Grover, P. S., & Bareja, R. (2015). Isolation and characterization of various Pseudomonas species from distinct clinical specimens. Journal of Dental and Medical Sciences, 14, E80-E87.