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Prevalence of *Pseudomonas aeruginosa* and *Acinetobacter baumanni* Isolated from Clinical Samples in a Tertiary Care Hospital Islamabad

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Abstract: *A. baumanni*i and *P. aeruginosa* are Gram-negative coccobacilli and important nosocomial pathogens in healthcare settings, causing significant morbidity and mortality worldwide. Both are intrinsically resistant to many drugs and have the ability to develop resistance to virtually all antimicrobial agents. This study was conducted to determine the prevalence of *Pseudomonas aeruginosa* and *Acinetobacter baumanni*i isolated from various clinical samples. It was a three-year retrospective cross-sectional study. A total of 810 clinical specimens were evaluated using conventional methods for identification, and antimicrobial susceptibility was determined using the Kirby-Bauer disc-diffusion method. The isolates were obtained from suspected bloodstream infections, wound infections, urinary tract infections, and surgical site nosocomial infections. Sociodemographic and other variables of interest were collected using a structured checklist from patient record data. The data was analyzed using SPSS version 26, with a p-value of <0.05 considered statistically significant. A total of 810 clinical samples were evaluated from the years 2021 to 2023. Of these, 440 (54%) were males and 370 (46%) were females, with ages ranging from 1 to 90 and mean and median ages of 39.3 years and 37 years, respectively. Among the samples, *A. baumanni*i accounted for 364 (60.6%) and *P. aeruginosa* accounted for 446 (39.4%). Blood was the main source of isolates (18%), followed by urine (16%) and CSF (10%). In the analysis, we found that *P. aeruginosa* had the highest prevalence in all three years, ranging from 38% to 29%. In conclusion, *A. baumanni*i is an emerging pathogen with a wide range of resistances, making it difficult to control. Effective antibiotics include imipenem, meropenem, piperacillin/tazobactam, ciprofloxacin, gentamicin, tobramycin, colistin, and polymyxin B, while fluoroquinolones, aminoglycosides, cephalosporins, and carbapenems are used.

Keywords: *Pseudomonas aeuroginosa*, *Acinetobacter baumanni* , Antibiotics Resistance, Prevalence

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1. Introduction

Most of the bacterial species in the genus Pseudomonas are saprophytic. This makes it one of the most complex bacterial groups. Opportunistic infections in humans have been associated with around 25 different species. Some notable and clinically relevant species include *Pseudomalinosa*, *Pseudomonas aeruginosa*, *Pseudomonas fluorescens*, *Pseudomonas putida*, Pseudomonas stutzeri, Pseudomonas mallei, Pseudomonas *pseudomallei*, *Pseudomonas maltophila*, and *Pseudomonas putrefaciens*. Clinical specimens of hospitalized patients most frequently reveal the presence of *Pseudomonas aeruginosa*. In healthy people, it is a typical component of the gut microbiome, but in hospitalized patients, particularly those with burns, respiratory diseases, catheters, or impaired immune systems, it can cause infections (Bibi 2020).

*Acinetobacter baumanni*i is a type of non-motile, non-fermenting coccobacilli that is characterized by being gram-negative, catalase-positive, and oxidase-negative (Iswarya 2020). It is common for immunocompromised individuals, particularly those who have been in the hospital for an extended period (more than 90 days), to contract *Acinetobacter baumanni*i, an opportunistic infection (Alrahmany et al. 2021). It has been shown to invade skin and is commonly seen in aquatic environments. Infected individuals' respiratory and oropharyngeal secretions have also been discovered to have it in a highly isolated form (Blanchard and Waters 2022). The medical world is deeply concerned about its wide range of antibiotic resistance, which has resulted in its designation as a "red alert" human pathogen in recent years. The genus Acinetobacter is often considered to have a broad distribution in nature due to the high frequency with which it is detected in soil and surface water samples (Hubeny et al. 2022). Although not all species of Acinetobacter are found in their natural habitat, *Acinetobacter baumanni*i is known to target moist tissues, such as mucous membranes and exposed skin areas after injuries or accidents. Infections caused by *Acinetobacter baumanni*i manifest in the skin and other soft tissues as "peau d'orange," or orange skin. As the infection advances, the skin takes on a sandpaper-like appearance, and eventually, clear vesicles appear. Areas of skin rupture show hemorrhagic bullae, and bacteremia is detected after an evident necrotizing process. Failure to treat this illness can result in septicemia and even death (Vrancianu et al. 2021). *Acinetobacter baumanni*i and *Pseudomonas aeruginosa* are the most common causes of nosocomial infections, which cause many illnesses and deaths worldwide. Healthcare personnel' hands are a primary vector for bacterial transmission, including multidrug-resistant *Acinetobacter baumanni*i and *Pseudomonas aeruginosa* (Motbainor, Bereded, and Mulu 2020). The greatest distinguishing feature of these bacterial species is their antibiotic resistance, which they rapidly develop via a variety of mechanisms including decreased outer membrane permeability, efflux pump systems, enzymatic inactivation, and biofilm formation. These bacteria can live in healthcare settings. This is why they are especially effective against quinolones, aminoglycosides, and -lactams, which are quite uncommon. Antibiotic use, medicine prescriptions without susceptibility testing, self-medication, and extended hospitalizations have all been associated to multidrug resistance (Morris and Cerceo 2020). Hospital-acquired infections caused by drug-resistant *Acinetobacter baumanii* and *Pseudomonas aeruginosa* species are becoming more common and pose a substantial public health danger (Mekonnen et al. 2021). These bacteria can cause a variety of illnesses, such as pneumonia, bacteremia, meningitis, urinary tract infections, and wound infections (Anju et al. 2023). Infections caused by drug-resistant Acinetobacter and Pseudomonas spp. have been linked to longer hospital admissions and mortality (Nelson et al. 2022).

2. Material & Methodology

A cross-sectional retrospective study was conducted to assess the prevalence of *A. baumanni* and *P. aeruginosa* from clinical samples that were tested for bacterial presence dated from January 2021 to December 2023, from tertiary care teaching hospital in Islamabad, Dr. Akbar Niazi Teaching Hospital and CDA Hospital). The clinical information extracted from the microbiology laboratories included type of sample analyzed, name of pathogens isolated and the names of antibiotics used for susceptibility testing and the susceptibility results as recorded in the laboratory report.

The data analyzed during the current study are available from the corresponding author on reasonable request. Ethical clearance for this study was provided by management of hospital, department of medical Laboratory ethical review committee. As this was a retrospective study informed consent was not collected from study participants but a permission was obtained from the hospitals. Participants' study data were recorded in codes and were kept private and confidentially.

Several types of clinical specimens, including urine, blood, sputum, pus, tracheal aspirate, wound swab, ear discharge, pleural fluid, and cerebrospinal fluid (CSF) were cultured. Laboratories sampled for the current study employed similar standard microbiological culturing techniques. Antimicrobial susceptibility test was performed by

both laboratories using the Kirby-Bauer disk diffusion method in Muller Hinton agar and interpreted using CLSI (Kamran et al. 2022).

The data was organized and stored in an Excel file, and statistical analysis was conducted using SPSS 24.0. Proportions of predominant isolates, sociodemographic characteristics, and antibiotic resistance profiles were presented using tables and figures. $P < 0.05$ was considered significantly associated among variables.

3. Results

A total of 810 patient's data that were isolated from various clinical specimens was recorded from the year 2021 to 2023 for antimicrobial susceptibility test of *A. baumanni* and *P. aeruginosa.* Out of which 440 (54%) were males and 370 (46%) were females, whereas the age ranged from 1 to 90 with the mean and median ages of 39.3 years and 37 years, respectively. *A. baumanni* was the predominant isolate 446 (55%) than *P. aeruginosa* which was 364(45%) (Table 1).

Table:1. Socio-Demographic and Clinical Characteristics of Study Participants in ANTH and CDA Hospital, 2021-2023.

Variables		**Frequency**	**Percentage (%)**
Gender N = 810	Male	440	54
	Female	370	46
Age N = 810	<1	118	14
	1-14	145	18
	15-24	127	16
	25-44	172	21
	45-65	152	19
	>65	96	12
Types of Specimens N = 810	Blood	148	18
	Urine	131	16
	CSF	80	10
	Tracheal Aspirate	87	11
	Pus	84	10
	Sputum	79	10
	Wound	77	9
	Pleural Fluid	63	8
	Ear Discharge	61	8
Isolated Bacteria N = 810	*P. aeruginosa*	446	55
	A. baumanni	364	45

Figure 1. In this study, 446 *A. baumanni* and 364 *P. aeruginosa* were analyzed to investigate the trends of antimicrobial resistance. In the analysis, we found that *P. aeruginosa* had the most prevalence in all the three years ranging from 38 to 29%, while *A. baumanni* ranges was 37%, in 2021 and *Pseudomonas aeruginosa* was 38% in 2021 *Acinetobacter* was 45%, while *Pseudomonas* was 55%.

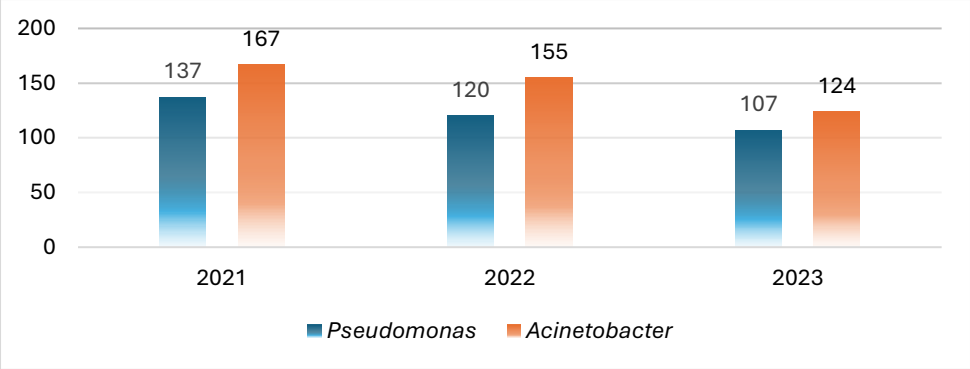

Figure 1: Frequency Distribution of Isolates

Figure 2 Among the two labs, 9 different types of clinical specimen were processed, but the highest level of the isolates was found in the blood 97 (35%), followed by urine 81 (29%), CSF 57 (20%), and Tracheal Aspirate 46 (16%) specimen. *P. aeruginosa* was the predominant isolate in urinary tract infection, sepsis and respiratory infections.

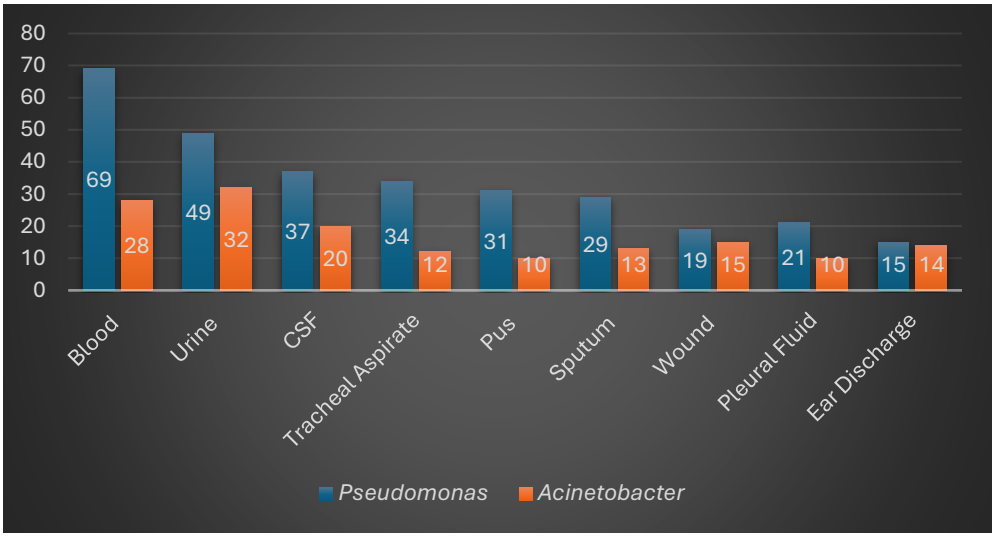

Figure 2: Sample wise distribution of Isolates.

Most of the strains were resistant to fluoroquinolones, aminoglycosides, cephalosporins and carbapenem group of drugs. The highest level of resistance was observed against Gentamicin, which is an aminoglycoside with 51 strains to be resistant, followed by ciprofloxacin, a fluoroquinolone group of drug (49; 30.2%), ceftazidime (45; 27.7%) cefoperazone-sulbactam (41;25.3%), amikacin (40;24.6%), meropenem (31; 19.1%), cefepime (31; 19.1%), imipenem (30; 18.5%), piperacillin tazobactam (29; 17.9%) and ertapenem (27; 16.6%). Most of the strains showed sensitivity towards Colistin and Polymyxin B. Forty-six strains (28%) of Pseudomonas spp were multidrug resistant (MDR) i. e, strains resistant to three or more than three different group of drugs. And interestingly, all the 46 strains were *Pseudomonas aeruginosa*. (Table 2)

Table 2: Antimicrobial resistance pattern of Pseudomonas spp

Antibacterial drug	**% of isolates showing resistance**	**% of isolates showing sensitivity**
Amikacin	24.6%	75.4%
Gentamicin	31.4%	68.6%
Ceftazidime	27.7%	72.3%

Ciprofloxacin	30.2%	69.8%
Imipenem	18.5%	81.5%
Meropenem	19.1%	80.9%
Ertapenem	16.6%	83.4%
Ofloxacin	14.1%	85.9%
Cefaperazone-sulbactam	25.3%	74.7%
Cefepime	19.1%	80.9%
Piperacillin-tazobactam	17.9%	82.1%
Tigecycline	13.5%	86.5%
Polymyxin B	3.7%	96.3%
Colistin	3.1%	96.9%

4. Discussion

Pseudomonas aeruginosa is the nosocomial infection-causing gram-negative, nonfermenting bacteria most commonly detected in clinical specimens. The coccobacilli *Acinetobacter baumanni* lack motility, fermentative activity, oxidase activity, and Gram-negative status. Significant morbidity and mortality have been associated with *Pseudomonas aeruginosa* and *Acinetobacter baumanni*i, the leading nosocomial infection causes. Finding out how often *Acinetobacter* and *Pseudomonas* species were found in various clinical samples and looking at patterns of antibiotic resistance were the main objectives of our investigation.

We studied 364 *A. baumanni* and 446 *P. aeruginosa* in our study to investigate the prevalence of Pseudomonas and Acinetobacter. In the analysis, we found that *A. baumanni* had the highest prevalence in all three years. The prevalence of *P. aeruginosa* in 2021 was 38%, while *Acinetobacter* was 37%, and in 2022, *A. baumanni* was 35%, while *P. aeruginosa* was 33%. The difference regarding the type and frequency of pathogens could be linked to several factors, like environmental conditions, health practices, patient conditions, personal hygiene, the number of patients involved in each study, and laboratory procedures.

This study was performed in a tertiary care hospital. *Pseudomonas aeruginosa* were examined with respect to the specimen. The maximum number of specimens from which *Pseudomonas aeruginosa* was isolated were blood, sputum, pus/wound swab, and tracheal aspirate. In this study, a total of 810 isolated species were *Pseudomonas aeruginosa* and Acinetobacter from various clinical samples. In this study, a higher prevalence of *Pseudomonas* infections was found in female patients than males (51.5 and 43.5%, respectively), similar to a study in the northern part of Nigeria (Hasan, Najati, and Abass 2020; Nelson et al. 2022).

In our study, the highest prevalence of infections due to *P. aeruginosa* was observed in the blood (51%), followed by urinary tract (25%) specimens and sputum (22%). De Francesco et al. and Yayn et al. reported that most of the isolates of *P. aeruginosa* were obtained from the respiratory tract, followed by the urinary tract, wounds, and blood (Mekonnen et al. 2021; Wang et al. 2021). While the highest prevalence of infection due to *A. baumanni* was observed in blood (46%), CSF (33%), followed by the urinary tract (19.4%), different studies also reported the isolation of *A. baumanni* from these clinical specimens (Benk\Ho et al. 2020). However, the current finding was lower than the study conducted in Dessie, which isolated *A. baumanni* from wound specimens at a rate of 51.4%, and in Iran at the rate of 56.7% which is higher than this study (Araya et al. 2023; Kamran et al. 2022).

5. Conclusion

In conclusion, *A. baumanni*i is a significant, opportunistic, and emerging pathogen that can cause devastating nosocomial infections. It can stick to surfaces, build biofilms, resist antimicrobials, and acquire genetic material from unrelated genera, making it a diverse and tough enemy to control and destroy. We found Imipenem, Meropenem, Piperacillin/Tazobactam, Ciprofloxacin, Gentamycin, Tobramycin, Colistin, and Polymyxin B to be the most effective antibiotics. Fluoroquinolones, aminoglycosides, cephalosporins, and carbapenems are first-line antibiotics for *Pseudomonas aeruginosa* due to its growing resistance. Most strains were colony- and polymyxin-B-sensitive. Pseudomonas spp. strains resistant to three or more medication groups included 46 MDR strains.

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