

The Identification of *Pseudomonas Aeruginosa* in Diverse Clinical Specimens and the Pattern of Antibiotic Resistance in Humans

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ABSTRACT

Objective: This study aims to identify the pollution foci of *Pseudomonas aeruginosa* (*P. aeruginosa*) in hospital environments, utilizing a random sampling approach to collect diagnostic data from various sources in Al-Karama Teaching Hospital and Al-Hey Hospital. **Methods:** Eighty samples were collected, evenly distributed with 40 samples from each hospital. Biochemical and agronomic tests were conducted to identify *P. aeruginosa* isolates. Antibiotic sensitivity was assessed for ten isolates using the Kirby-Bauer disc diffusion method across ten antibiotic classes. **Results:** The tests identified ten isolates of *P. aeruginosa*. The antibiotic sensitivity evaluation revealed that nearly all isolates exhibited considerable resistance to the antibiotics tested, demonstrating a broad spectrum of resistance to multiple antibiotic classes. **Novelty:** This study highlights the prevalence of *P. aeruginosa* in hospital settings and its significant resistance to commonly used antibiotics, emphasizing the critical need for enhanced infection control measures and antibiotic stewardship in healthcare facilities.

INTRODUCTION

According to the World Health Organization (WHO), *Pseudomonas aeruginosa* has been flagged as a species of “greatest public health concern.” Although *P. aeruginosa* rarely causes serious infections in healthy individuals, it is associated with various invasive infections in patients with long-term hospitalization, critically ill intubated patients, those treated with broad-spectrum antimicrobial therapy or cancer chemotherapy, and patients with ventilator-associated pneumonia [1].

Resistance of *P. aeruginosa* to antipseudomonal drugs continues to increase over the years in several regions of the world. Antimicrobial resistance in the intensive care unit (ICU) contributes significantly to the incidence of *P. aeruginosa* infections within a few days of antimicrobial therapy commencement.

Furthermore, co-infection with other microorganisms prior to the isolation, total parenteral nutrition usage, comorbid cerebrovascular disease, history of cerebrovascular disease, admission to the ICU, malignancy, compromised immune system, and obstructive coronary disease are known risk factors for *P. aeruginosa* infection [2].

Mechanical ventilation, acute respiratory failure, and infection site in the respiratory tract and central.

It has been acknowledged that *P. aeruginosa* is a bacterium that poses a major and grave risk to public health in general and human health in particular in light of World Health Organization findings. While *P. aeruginosa* infections do not always result in serious infections in healthy individuals, they are known to be invasive in hospital environments and are associated with extended hospital stays, particularly in patients with severe illnesses and those who take numerous antibiotics. Those utilizing mechanical respirators and those receiving chemotherapy [1].

Due to the careless use of antibiotics, it has been shown that *P. aeruginosa* resistance has grown over time in various parts of the world. It has been noted that when patients are given antibiotics incorrectly, which leads to mutations resistant to these treatments, the resistance of the bacteria in the critical care unit grows within a few days. *P. aeruginosa* infection is also caused by a variety of other factors, including complete reliance on parenteral nutrition, the prevalence of immune disorders like malignant tumors, admission to the intensive care unit, weakened immunity, inherited illnesses, and obstructive coronary artery diseases. These risk variables are all involved in bacterial infections [2].

The existence of numerous resistance mechanisms in *P. aeruginosa*, particularly at infection sites including the respiratory system, cardiac catheters, and surgical equipment, has been reported to limit the mechanism of treatment with antibiotics employed against the bacterium. In 2014, Kollef et al. Some of the most common bacterial co-infections are *Haemophilus influenzae*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Mycoplasma pneumoniae*, *Streptococcus pneumoniae*, and *Pseudomonas aeruginosa* [3], [4]. The primary source of *P. aeruginosa*'s pathogenicity is the variety of cellular virulence factors that it is capable of producing. All of these substances have a harmful impact on mammalian cells, and they may be summed up as follows: alkaline protease, exotoxin U (exoU), secretion protein III, exoenzyme A (exoA), exotoxin S (exoS), elastase, and protease IV. each of which has a toxic effect on mammalian cells [5], [6].

P. aeruginosa may become resistant to antibiotics in a number of ways, the primary one being the synthesis of enzymes. Since metallo-beta-lactamase (MBL) can break down beta-lactam antibiotics with the exception of aztreonam, it may be produced quickly, exhibits significant carbapenemase activity, is resistant to beta-lactamase inhibitors, and is very dangerous to human health [7], [8], [9].

Aim of the study is (1) *Pseudomonas aeruginosa* isolation and identification from clinical specimens, (2) examine how resistant *Pseudomonas aeruginosa* is to various medications.

RESEARCH METHOD

A. Sample Collection

The present investigation comprised the gathering of sixty samples of diverse clinical from Alhey Hospital and Karma Hospital. In order to explore the *P. aeruginosa* bacterial contamination and develop diagnostic, prophylactic, and therapeutic measures, random sampling was carried out. The samples were taken from 20 ear swabs, 25 pus samples, 15 stool samples, and 20 urine samples, divided by ear. Additionally, the samples were obtained from various locations and also came from dogs. The planning technique was used to grow the samples, and the plates were incubated on blood agar and MacConkey for 24 hours at 37°C.

B. Detection of the Isolates

Based on the API 20 E confirmatory test, biochemical and morphological tests, and comparing the results with the findings reported by Holt et al., 1994 [10].

C. Antibiotic Ten Test (Qualitative Disk Method)

The sensitivity of isolates of *Pseudomonas aeruginosa* was assessed using twelve antibiotic disks, which included amikacin (AK), amoxicillin-clavulanic acid (AMC), ceftriaxone (C), trimethoprim-sulfamethoxazole (TMP), cefotaximase (CTX), N-acetyl cysteine (NA), Meropenem (MEM), Azithromycin (ATM), gentamicin (CN), and rifampicin (RA) [11].

RESULTS AND DISCUSSION

A. Isolation and Characterization of *Pseudomonas aeruginosa*

As table 1 shows, eighty specimens were obtained from patients at Wasit City hospitals. Ten local samples were analyzed based on microscopic and cultural characteristics. Utilizing confirmatory testing with API 20 E and biochemical assays, genus and species were identified.

Table 1. Types of sample, number and Percentage of *Pseudomonas aeruginosa* isolated from clinical samples of human.

Sample	Type	The number	Percentage
Clinical	Urine	20	25 %
Clinical	pus	25	31.35 %
Clinical	stool	15	18.55 %
Clinical	Ear	20	25 %

B. Cultural Characteristics

The primary goal of the sample collection was to isolate the *P. aeruginosa* bacteria. The isolates were separated based on the phenotypic features of the developing colonies, such as the dark color of the colonies and the presence of a clear halo around the center of the blood agar, which indicated the bacteria's ability to break down blood. The results of the biochemical tests indicated that the oxidase test was positive. The isolates were all

distinguished by their inability to produce hydrogen sulfide gas and by the fact that neither sucrose nor lactose nor their corresponding sugars were fermented. Enterobacteriaceae identification was done using the API 20 E confirmatory to highlight the biochemical results. The findings demonstrate that *P.aeruginosa* was the tested isolate. as seen in Figure 1 and Table 2.

Table 2. Api 20E technique of *P.aeruginosa*.

No.	Active ingredients	Symbol test	Results
1.	Ortho NitroPhenyl-Bd-Galactopyranside	ONPG	-
2.	L-arginine	ADH	-
3.	L-Lysine	LDC	+
4.	L-Ornithin	ODC	+
5.	Trisodium citrate	CIT	+
6.	Sodium thiosulfate	H ₂ S	+
7.	Urea	URE	-
8.	L-tryptophane	TDA	+
9.	L-tryptophane (indole production)	IND	-
10.	Sodium pyruvate	VP	-
11.	Gelatin (bovine origin)	GEL	+
12.	D-Glucose	GLU	-
13.	D-Mannitol	MAN	+
14.	Inositol	INO	-
15.	D-Sorbitol	SOR	+
16.	L-Rhamnose	RHA	-
17.	D-Saccharose (sucrose)	SAC	+
18.	D-Melibiose	MEL	+
19.	Amygdaline	AMY	-
20.	L-Arabinose	ARA	-



Figure 1. Api 20 E technique for *P.aeruginosa*.

C. Antimicrobial susceptibility

Azithromycin, gentamicin, rifampicin, amikacin, amoxicillin-clavulanic acid, ceftriaxone, trimethoprim-sulfamethoxazole, cefotaximase, N-acetyl cysteine, Meropenem, and Acetaminophen are among the antibiotics that *P.aeruginosa* isolates can shown 100% resistance to. There was a 100% prevalence of MDR. The current investigation found that the ratios of *P. aeruginosa* resistant isolates to antibodies RA and CTX, where the bacteria resisted all isolates, increased considerably. The proportion of resistant isolates to antibodies C and MEM was lower than that of resistant isolates to antibodies AMC and TMP. It can decipher the percentage of highly expressed resistance to these antibodies displayed by *P.aeruginosa* isolates, which is essential in the current investigation to use the random to these antibiotics. In addition to resistance to evolution, the usage of therapeutic dosages caused this bacterium to arise, which in turn led to the formation of isolation mutation [12], [8].

CONCLUSION

Fundamental Finding : The study highlights that *P. aeruginosa* can be isolated from mucosal membranes in all clinical conditions, underscoring its prevalence and adaptability. Additionally, the research identified the role of medication delivery in contributing to antibiotic resistance in these bacteria. The frequent occurrence of resistance across samples suggests that hospital environments, including staff and equipment, may play a significant role in spreading this pathogen. The API 20 E test emerged as a vital diagnostic method for early detection of infections, particularly for immunocompromised individuals, despite its potential contamination risks. **Implication :** These findings emphasize the urgent need for enhanced infection control measures in healthcare settings to curb the spread of *P. aeruginosa*. Hospital workers and equipment should undergo stringent sterilization protocols to minimize contamination. The role of diagnostic tools like the API 20 E test in early detection reinforces their importance in clinical settings. However, their susceptibility to contamination necessitates the adoption of improved practices to ensure accuracy and patient safety. **Limitation :** This study is limited by its focus on specific clinical settings and diagnostic tools, which may not fully represent all potential sources or conditions contributing to *P. aeruginosa* spread. Moreover, it does not extensively explore the molecular mechanisms underlying resistance or the impact of different treatment regimens. The absence of broader sampling and advanced detection methods also limits the generalizability of the results. **Future Research :** Further studies should employ advanced molecular techniques to detect *P. aeruginosa* early and identify appropriate antibiotics efficiently. Expanded research is needed to explore susceptibility patterns in lab animals and multidrug-resistant strains, especially in bloodstream and urinary tract infections. Developing innovative treatments and strategies to minimize antibiotic resistance is crucial for addressing this growing threat in clinical settings.

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