

# ***Study of antibiotic sensitivity of Pseudomonas aeruginosa isolated from women with urinary tract infection***

## ***Abstract***

UTIs, or urinary tract infections, are among the most prevalent infections in the world. It is estimated that 7% to 10% of UTIs are caused by *Pseudomonas aeruginosa*. Antibiotic resistance levels in *P. aeruginosa* isolates from UTIs are often greater than in *E. coli* isolates. So, the current study aimed to test the antibiotic sensitivity to *Pseudomonas aeruginosa* isolated from patients with urinary tract infection. Clinical samples (95) were collected from Kirkuk Hospital in Kirkuk city for the period from May to Augustus 2024 from women who were admitted and hospitalized after consulting the specialist doctor and referring him to the laboratory. The method of collecting samples included the following: Urine samples were taken from women with UTI ranging in age from (5-59 years) of women, after which they were transferred directly to the laboratory to be cultured on the culture media. Of the total samples investigated, 64 (or 67.4%) showed positive findings for *P. aeruginosa* growth when cultured on the blood agar, cetrimide agar, and MacConkey agar. the isolation rate of *P. aeruginosa* from women with urinary tract infection was 18.8%. for antibiotic susceptibility test, *P. aeruginosa* showed 81.8%, 93.8% and 94.9% sensitive toward Gentamycin, Imipenem and Amikacin. On the other hand, *P. aeruginosa* was completely sensitive (100%) toward Tobromycin. Its concluded that *P. aeruginosa* is one of the main causes of urinary tract infection in women.

### **Keywords:**

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### **Introduction**

One of the most prevalent bacterial diseases that plague people at any point in their lives is urinary tract infection (UTI) [1,2]. In the US each year, UTIs cause more than 8 million doctor visits, 1.5 million ER visits, and 300,000 hospital hospitalizations [3, 4]. Moreover, urinary tract infections—which make up 20 to 50% of all noncomial infections—are among the most

prevalent ailments among hospitalized patients. It is estimated that *Pseudomonas aeruginosa* causes 7% to 10% of UTIs in hospital settings [5–6]. A common Gram-negative bacterium that is thought to be the source of healthcare-associated infections is *pseudomonas aeruginosa* [7]. In older patients, *P. aeruginosa* urinary tract infections are linked to increased morbidity and mortality. Antibiotic resistance levels in *P. aeruginosa* isolates from UTIs are often greater than in *E. coli* isolates [8–9]. One of the most significant bacteria creating challenging clinical issues is *P. aeruginosa* [10]. *Pseudomonas aeruginosa* has resistance against aminoglycosides, quinolones, and  $\beta$ -lactam antibiotics, among other drugs [11]. The three main types of resistance that *P. aeruginosa* uses to fend off antibiotic attacks are intrinsic, acquired, and adaptive resistance. *P. aeruginosa*'s intrinsic resistance is comprised of low outer membrane permeability, the formation of efflux pumps that force antibiotics out of the cell, and the manufacture of enzymes that inactivate antibiotics. Resistance in *P. aeruginosa* can be acquired through horizontal gene transfer or mutational changes [12]. Because of its capacity to form biofilms and its innate, acquired, and adaptive resistance mechanisms, *P. aeruginosa* poses a serious threat in the clinical setting [13–14]. It demonstrates resistance to numerous antibiotics, including as fluoroquinolones,  $\beta$ -lactams, and aminoglycosides, by expressing efflux pumps, producing enzymes that inactivate drugs, and having a low outer membrane permeability [15]. The treatment landscape is further complicated by acquired resistance resulting from mutations and horizontal gene transfer, as well as adaptive resistance demonstrated by the creation of biofilms and the appearance of persister cells [16]. So, The purpose of the current study was to evaluate *Pseudomonas aeruginosa* antibiotic sensitivity that was isolated from urinary tract infection patients.

## **Materials & methods**

### **Specimen Collection**

95 clinical samples from women who had been admitted and hospitalized after consulting a specialist physician and having him referred to the laboratory were gathered from Kirkuk Hospital in Kirkuk City between May and August of 2024. The following was a part of the sample collection procedure: Women suffering from UTIs, ages 5 to 59, had their urine samples collected. The samples were then sent straight to the lab to be cultured on culture media.

## **Bacterial Identification**

Bacteria were diagnosed based on the following aspects:

### **Morphological diagnosis and media characteristics**

The *P. aeruginosa* colonies growing on blood agar and cefrimide agar were identified based on their culture characteristics, and they were then incubated for 24 hours at 37 °C.

### **Direct examination**

By using a microscope to examine the morphological characteristics of bacterial cells—specifically, how they contacted the gram stain, which indicates the kind of interaction as well as the shape and arrangement of the germ cells—bacterial colonies were found.

### **Biochemical reaction and motility test**

Numerous biochemical tests, such as the H<sub>2</sub>S production, methyl red, citrate, urease, voges-proskauer, catalase, oxidase, and indole test, were carried out in order to identify and diagnose bacteria.

### **Identification of bacteria isolates via VITEK2**

Advanced colorimetric technology is represented by VITEK 2, the next generation of the gold standard in microbial identification. Procedure: All of the following actions were carried out in compliance with the guidelines provided by the manufacturer, Biomerieux.

### **Antibiotic susceptibility test (AST)**

The Kirby-Bauer disc diffusion method employing Muller Hinton (MH) agar was used for the AST for all isolates in compliance with the guidelines set out by the Clinical Laboratory Standards Institute (CLSI, 2020)[17].

## **Results and discussion**

### **Samples distribution**

Table 1 lists the 95 urine samples that were taken from patients who had UTIs for the current investigation. Based on the best cultured media—such as blood agar, Cefrimide agar, and

MacConkey agar—64 (67.4%) of the total samples showed good results for bacterial growth. Thirty-one (32.6%) out of the total samples showed negative results for bacterial growth.

**Table (1): Distributed of study samples according to UTI**

	No. (%) <b>+ve culture</b>	No. (%) <b>-ve culture</b>	Total No.(%)
Women	64(67.4%)	31(32.6%)	95 (100.0%)

According to the current inquiry, 31 patients (32.6%) had no bacterial infection, while 64 patients (67.4%) tested positive for bacterial infections (table 1). However, in other trials, the UTI percentages were, respectively, 75.42 percent and 61 percent [18]. The absence of growth in urine samples could be attributed to the impact of antibiotics offered to patients throughout their hospital stay and the use of broad range antimicrobials in their treatment. Furthermore, the rate of bacterial isolations was significantly decreased by applying the outer sterilizer solutions [19]. Additional causes of anaerobic bacteria, mold, and other bacteria that cannot be separated using the normal procedures utilized in this study and may require specialized techniques for their isolation and development could be revealed by examining the urine samples [19].

### Identification

On blood agar, Cetrimide, and MacConkey agar, the morphology, diameter, and forms of the bacterial isolates were ascertained. Additionally, the results of the biochemical identification were confirmed by means of the System small Vitek-2 equipment and microscopic and biochemical exams, which comprised the particular tests for each kind. The Vitek-2 results were in line with the findings of the biochemical testing. Table (2) shows that the isolation rate of *P. aeruginosa* from women with urinary tract infection was 18.8%, while the isolation rate of other species from women with urinary tract infection was 81.2%.

**Table (2): isolates percentages of gram negative bacteria**

Gram negative bacteria	No.	%
<i>P. aeruginosa</i>	12	18.8
Other types	52	81.2

Total	64	100
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### *Pseudomonas aeruginosa*

The shape and diameter of *P. aeruginosa* isolates on Cetrimide agars are depicted in Figure (1) for the primary isolate, pink-red rods under a microscope, and gram-negative bacteria. Gram stain reaction and other microscopic features were used to diagnose bacterial isolates belonging to the genus. *Pseudomonas aeruginosa* biochemical tests As illustrated in figure (2), it tested negative for urease, indole, and Kligler iron k/k, but positive for citrate, catalase, oxidase, and motility.

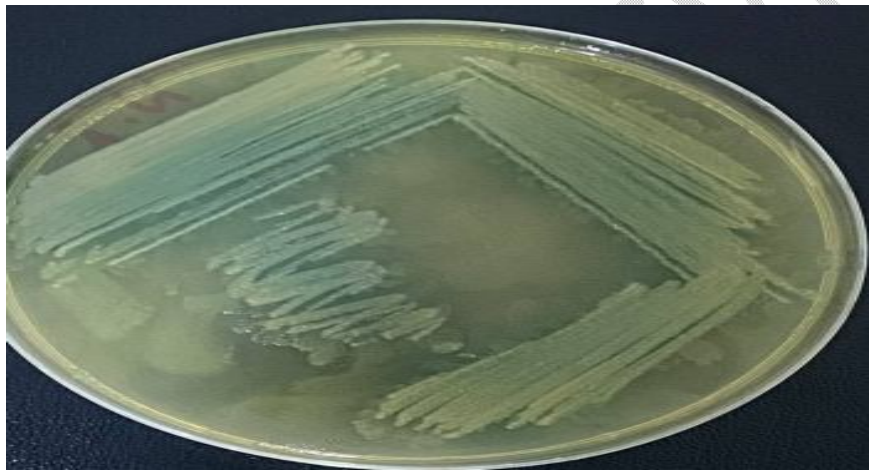


Figure (1): *P. aeruginosa* colonies on Cetrimide agar

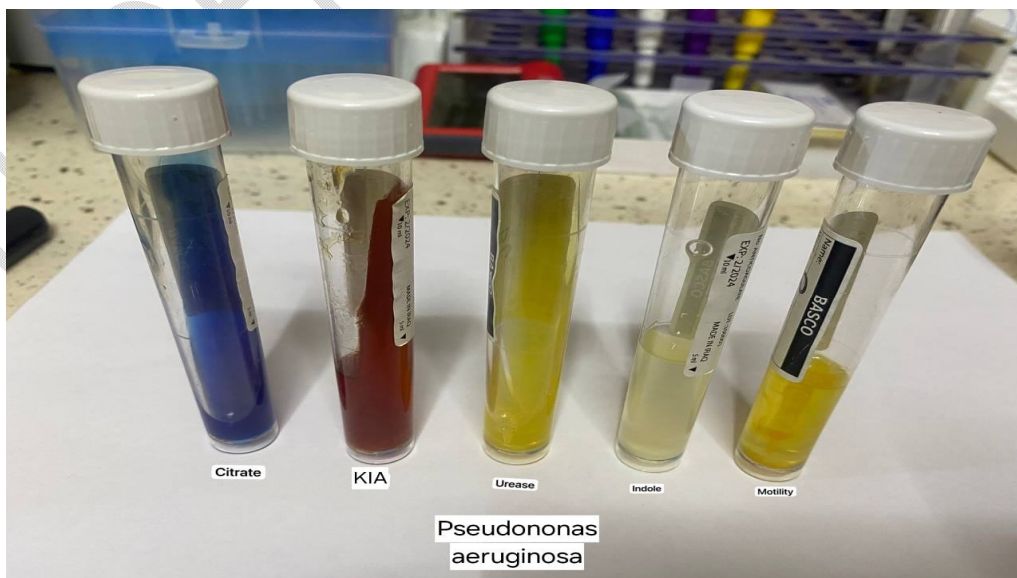


Figure (2): the biochemical tests for *P. aeruginosa* isolates.

### Antibiotic susceptibility test

*P. aeruginosa* showed 81.8%, 93.8% and 94.9% sensitive toward Gentamycin, Imipenem and Amikacin. On the other hand, *P. aeruginosa* was completely sensitive (100%) toward Tobromycin respectively.

Table (3): Antibiotic susceptibility test of *P. aeruginosa*

Antibiotics	Sensitive %	Intermediate %	Resistant %	P value
AMP	41.1	20.4	38.5	0.0001
VN	53.8	0.0	46.2	
DA	69.2	0.0	30.8	
TMP	72.3	9.1	18.2	
CAZ	69.2	0.0	30.8	
CTX	61.5	0.0	38.5	
CFM	53.8	0.0	46.2	
CN	81.8	18.2	0.0	
IMI	93.8	0.0	6.2	
NA	76.9	0.0	23.1	
CIP	53.8	3.1	43.1	
LEV	53.8	3.1	43.1	
AZT	61.2	8.3	30.5	
AK	94.9	5.1	0.0	
TOB	100.0	0.0	0.0	

AMP= Ampicillin, VN=Vancomycin, DA=, Clindamycin, TMP=Trimethoprim, CAZ=Ceftazidime, CTX=, Cefotaxime, CPM=Cefepime, CN=Gentamicin, IMI=Imipenem, NA=Nalidixic acid, CIP=Ciprofloxacin, LEV= Levofloxacin, AZT = Azithromycin, AK=Amikacin, TOB = Tobramycin.

Certain European nations have been reported to have the highest rates of resistance to aminoglycosides [20]. According to the findings, every *P. aeruginosa* sample tested positive for amikacin (90.9%), and all isolates were very sensitive (100%) to imipenem and tobramycin. The study's obtained results, which are displayed in Table (3), indicate a notable increase in pseudomonal resistance to beta-lactam antibiotics. These findings concurred with research published by [21]. Between cefixime, levofloxacin, ciprofloxacin, and vancomycin, the percentage of resistance to other antibiotics was 46.2%. Modifications in the permeability of the

outer membrane through changes in porin protein channels constitute a component of many resistance mechanisms. *P. aeruginosa* can acquire resistance to this antibiotic through the outer membrane, which offers an efficient intrinsic barrier to accessing the targets, which are located in the cytoplasm, cell wall, or cytoplasmic membrane [22]. The present findings were in contrast to those of Hegazy et al. [23], who reported that 74.4% of *E. coli* bacterial isolates were cefotaxime-resistant. The results of this investigation were less than those of theirs since 38.5% of the bacteria were resistant to ceftazidime. 6.8% of Iranian bacteria were resistant to ceftazidime, compared to 15.5%, 42.2%, and 30% to cefotaxime. The researchers in those investigations hypothesized that the bacteria's resistance may be due to the presence of natural efflux pumps in those organisms [24–25]. The current study's findings indicated that there was 30.8% and 23.1% resistance to aztreonam and nalidixic acid, respectively. Primarily, these resistances serve as a significant marker for the existence of ESBLs. Unquestionably, one of the most significant etiological culprits of many serious and potentially fatal nosocomial infections is ESBL-producing Gram negative rods [26]. In Gram-negative bacilli, the genes producing ESBLs are typically found on sizable, transportable plasmids that are readily able to proliferate [27]. Nalidixic acid is a well-established antibiotic that is still a top choice for treating urinary tract infections (UTIs) in patients since *E. coli* isolates show only a moderate level of resistance to it. The results of this study were consistent with other previous studies, including those carried out in Iran by Tajbakhsh et al. [29] and India by Mittal et al. [28]. After isolating bacteria from patients with urinary tract infections, the results showed that the rate of bacterial resistance to this antibiotic was rather low, reaching 6.6% and 6.25%, respectively.

## **Conclusions**

The results of the current work showed that *P. aeruginosa* is one of the main causes of urinary tract infection in women, and that *P. aeruginosa* has high resistance against many antibiotics, but Tobromycin was the best drug in treating *P. aeruginosa*.

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Details of the AI usage are given below:

- 1.
- 2.
- 3.

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