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Isolation and identification of *Pseudomonas aeruginosa* from clinical specimens

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

(وَعَلَّمَكَ مَا لَمْ تَكُن تَعْلَمُ وَكَانَ
فَضْلُ اللَّهِ عَلَيْكَ عَظِيمًا)

صدق الله العلي العظيم

سورة النساء الآية (113)

Dedication

I dedicate my true gratitude to the light of my eyes and the reason for my progress and my effort in life... My father

To the devoted woman who illuminated my life with her kindness and endless love... My Mother

To those who supported me and stayed by my side... My sister and brothers

To the friends of high and beautiful wishes...

I dedicate the fruit of this humble effort...

Kawther

Fatima

Isolation and identification of *Pseudomonas aeruginosa* from clinical specimens

Abstract:

The study included isolation and identification of *Pseudomonas aeruginosa* from clinical specimens as (60) were collected during a period from January to March (2024).The isolates were identified depending on phenotypic criteria, microscopic examination and confirmed these tests by Vitek2 system, Out of 60 specimens, Positive bacterial culture was (71.7%) while negative bacterial culture was (28.3%). *Pseudomonas aeruginosa* recorded (37%) relative to other bacterial causes (63%). The samples from diabetic foot ulcers were (68.7%) isolates in male more than female at more than fifty years old, urine (18.7%) in female than male at twenty decade. Finally in blood (12.6%) ratio in male without infection in female at twenty decade.

Aim of the study:

The purpose of the present study was to characterize *P. aeruginosa* prevalence among UTIs, blood sepsis and wound infections of diabetic patients.

Keywords: *P. aeruginosa*, Urinary Tract Infection, Diabetic foot ulcer, Vitek2

Introduction:

Pseudomonas aeruginosa (*P. aeruginosa*) is one of the most clinically important Gram-negative bacteria. It is an opportunistic pathogen responsible for about 10 to 20% of nosocomial infections worldwide (Edward *et al.*, 2023) which cause many diseases on a large scale, including hospital infections, Septicemia, urinary tract, respiratory, wound infections and soft tissue infections. It is common in individuals with an immunocompromised state such as cystic fibrosis, bronchiectasis, neutropenia, burns, cancer, AIDS, organ transplantation, uncontrolled diabetes, and admission to the intensive care unit, as well as individuals with organs. Invasive catheters, such as indwelling catheters or endotracheal tubes, are also at risk due to the organism's unique ability to form biofilms that are difficult to detect (Mulcahy *et al.*, 2014), as they are commonly

found in the environment, especially in freshwater such as tanks in urban communities, hot tubs, jacuzzis, and bathtubs. Swimming can cause a wide range of community-acquired infections such as folliculitis, puncture wounds leading to osteomyelitis, pneumonia, otitis externa, and others (Kerr and Snelling, 2009).

Among gram-negative bacteria, *Pseudomonas aeruginosa* is the predominant causative agent for diabetic foot ulcer infections in low-resource countries. *P. aeruginosa* possesses a variety of virulence factors, including biofilm formation (Yakout and Abdelwahab 2022)

In UTIs, *P. aeruginosa* are associated with high levels of morbidity and mortality in elderly hospital patients and has been highlighted as an antibiotic resistant pathogen of the highest concern by the World Health Organization (Newman *et al.*, 2022), Because it resistant to many antibiotics, it pose a strong challenge to antibiotic treatment. So it has many virulence factors which classified into cell-associated factors, including adhesins and lipopolysaccharide, as well as secreted factors, such as exotoxin A, proteases, exoenzymes, phospholipases C, pyocyanin, alginate, and DNase (Strateva and Mitov, 2011).

Material and method:

Collection of samples

Samples were collected from patients referred to the General Teaching Hilla Hospital undergoing urinary tract infections, diabetic foot ulcer and blood from patients with 60 patients with different ages.

Bacterial isolation and identification:

The samples were collected under sterile conditions include sterile test tubes and swabs then transported immediately to the Microbiology laboratory to do cultural tests on the media for *P. aeruginosa* isolation such as Nutrient agar, MacConkey agar and chromo agar media, and incubated at 37°C for 24-48 hours, after the microscopically, morphologically also were identified the bacterial Colonies with pigments on Nutrient agar, this phenomenon considered as indicator

to diagnose of *P. aeruginosa* (McFadden, 2000). Finally Vitek2 technique has done for confirmation the tests according to the product Company (BioMeriux).

Results and Discussion:

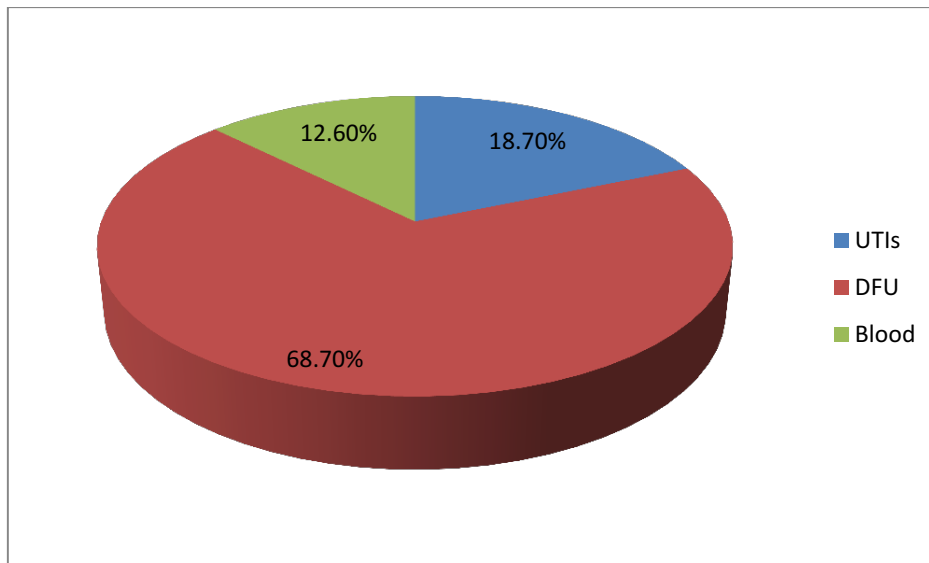
A total number of 60 specimens were checked for isolation and identification of *P. aeruginosa*. The results revealed that (37%) of *P. aeruginosa* among bacterial isolates as shown in Table (1) after identification by morphological, microscopical and confirmed tests by VITEK2 system.

Table (1): The percentage of *P. aeruginosa* among patients

No. of clinical specimens	Results		No. of isolates (%)
60	Positive bacterial culture	<i>P. aeruginosa</i>	16 (37%)
		Other bacterial causes	27(63%)
		Total	43(71.7%)
	Negative bacterial culture		17 (28.3%)

The results revealed high rate of DFU infection by *P. aeruginosa* 11\16 (68.7%), while UTIs 3\16 (18.7%) and blood specimens was 2\16(12.6%) as showmen in Figure 1. Our results were near to another studies as Maharjan, (2022) who found *P. aeruginosa* in urine as (29.41%) and in blood infections (11.76%).Another observation was reported in the study of Gupta & Shrestha, (2019) that found the rate in wound swab (55.6%) and urine 10 (11.1%). The high rate of isolation from the DFU specimens in our study may be due to its ability to cause wound infections in most people. A study by Mohamed *et al.*, (2022) who identified *P. aeruginosa* in DFU as (27.02%) which lower than our study.

Figure 1: The percentage of *P. aeruginosa* among diseases



According to gender, the results showed high infection in DFU patients by *P. aeruginosa* in male (81.8%) compared to female (18.2%) as shown in Table 2. It can be attributed to women's interest in personal hygiene and foot cleanliness in the event of an injury. The infections were distributed among age groups more than fifty years old which due to the improper blood sugar control, suppressed immunity, inadequate blood supply and neuropathy, burdens of life and exercise habits.

Table 2: Gender wise distribution of clinical isolates of *Pseudomonas aeruginosa*

Samples	No. of samples	Female	Male	Female%	Male%
DFU	11	2	9	18.2%	81.8%
UTIs	3	2	1	66.6%	33.4%
Blood	2	0	2	0%	100%

A similar observation by Ahmed *et al.*, (2020) who found the infection in the age group (54 – 63) by (80%). In the finding of retrospective group study among Saudi population reported by Al-Rubeaan *et al.*, (2015) who demonstrated that 45 years age or older is a risk factor for developing DFU. In agreement with most studies found that male gender was predominant in DF patients Semedo *et al.*, (2016) and Magiorakos *et al.*, (2012).

In UTIs patients, the female recorded high percentage (66.6%) while male was (33.4%) as showed in (Table 2) that due to a different predisposing factors such as anatomical characteristics, hormonal status, pregnancy and lifestyle habits which they lead to much more common in female (Storme *et al.*, 2019). UTIs in males are uncommon due to the longer length of the urethra, antibacterial properties of prostatic fluid and less colonized periurethral, so the problems of infection appear after the 50 age (Shaheen *et al.*, 2019; Al.Dulaimi *et al.*, 2022). It's quite similar to the study which showed that female infected more than male at ratio (80.4 %) in female and (19.6%) in male (Al.Dulaimi *et al.*, 2022). The infection was distributed among twenty decade old, this result was agreement with another studies by Aya, (2019), and Al-Naqshbandi *et al.*, (2019) were determined a high ratio of positive patients between (26-30) and (21-30) years old.

Finally, in blood both specimens were male (100%) and were in twenty decade old, this result was agree with Elsaid *et al.*, (2022) who found blood infection in male (90%) and female (10%) and disagree with the study of Bibi *et al.*, (2015) which showed (0%) of *Pseudomonas aeruginosa* in blood sepsis. Abdelrahman and Ahmed, (2021) reported that *P. aeruginosa* prevalence depends on other various factors like nature of geographical locations, degree of contamination, immune status of patients, virulence of strains and degree of implementation of measures of infection control in hospitals.

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