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Original Research Article

Determine the Prevalence of Pathogens and Their Antibiotics Sensitivity for Patients Suffering from Urinary Tract Infections in Babylon, Iraq

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Abstract: ***Background*:** Urinary tract infections (UTIs) are considered one of the most common infectious diseases, affecting most people of all age. ***Objectives*:** This study was conducted to determine the types of common bacteria causing UTIs and to measure the sensitivity of antibiotics to different bacteria. ***Materials and Methods*:** The current study included 208 of young group patients (aged 18 - 50 years) clinically suspected of having a UTIs. Out of 208 samples, the culture results showed that only 100 samples were positive for the presence of significant bacteria. Samples (midstream urine) were collected in a sterile tube, and significant results were shown for the isolated bacteria on selective media, identified by the VITEK-2 system. Then the Kirby-Bauer method was used to perform antibiotic susceptibility testing according to the Clinical and Laboratory Standards Institute (CLSI) guidelines. ***Results*:** The most common bacteria identified included; *Escherichia coli* (68.3%), *Staphylococcus aureus* (7.8%), *Enterococcus faecalis* (6.7%), *Klebsiella pneumoniae* (5.8%), (3.8%) for both *Pseudomonas aeruginosa* and *Staphylococcus saprophyticus*, While *Streptococcus agalactiae* accounted for (1.9%), *Proteus mirabillus and Serratia marcescens* (0.9%) for all of them. Moreover, there was mixed bacterial growth. Results revealed *E. coli* was sensitive to Imipenem, Ciprofloxacin and resistant to Tobramycin and Amikacin. Meanwhile, *K. pneumoniae* was completely sensitive to Imipenem and Ciprofloxacin with utterly resistant to Piperacillin. *P. aeruginosae* was sensitive to Imipenem, Meropenem and resistant to Azithromycin, Tobramycin, Ceftriaxone and levofloxacin. Whereas, *S.aureus* was sensitive to Ciprofloxacin and Levofloxacin and resistant to Trimethoprim and Azithromycin, *E. faecalis* was sensitive to Vancomycin with resistant to Azithromycin and Ciprofloxacin. ***Conclusions*:** The study concluded that females are more susceptible to UTIs than to males. In addition, females with UTIs tend to be younger than males. *E. coli* is the most common isolate among bacteria that cause UTIs. Study revealed that Imipenem remains the most potent treatment for inhibiting most types of bacteria causing UTIs, in addition, Meropenem, Ciprofloxacin, and Levofloxacin can be considered a potential treatment option.

Keywords: UTI, Antibiotic susceptibility, Uropathogens, Imipenem.

INTRODUCTION

Human UTIs are common, affecting both the upper and lower urinary tracts and severely impairing the quality of life for many people. It is still a problematic health concern in several nations [1]. It is the most common infectious illness among the older population [2].

Both community-associated UTIs (CAUTIs) and healthcare-associated UTIs (HAUTIs) are forms of UTIs [2].

Although there are many other causes of UTIs, uropathogenic bacteria are the most often isolated species globally [3]. Gram-negative bacteria, members of the Enterobacteriaceae family, are a significant cause of UTIs [4]. These infections are difficult to treat due to the high annual infection incidence and imprecise UTI diagnosis [5], UTIs are classified into two categories: "uncomplicated" UTIs, which occur in healthy individuals, non-pregnant women over the age of 14, with no abnormalities in the kidneys or urinary system;

and "complicated" UTIs, which include males and all other women [6, 7].

Even though E. coli makes up a large amount of the normal intestinal flora, it is known to be a very active opportunistic pathogen that causes a sizable percentage of UTIs [8, 9]. In addition to *Staphylococcus aureus*, *Klebsiella pneumonia*, *Enterococcus faecalis*, and *Candida albicans*, a wide range of other species can also infect the urinary system, including *Staphylococcus saprophyticus*, *Enterobacter sp*, *Proteus mirabilis*, and *Pseudomonas aeruginosa* [10–13].

The prevalence of antibiotic resistance in UTIs is quickly increasing, leading to a significant increase in morbidity. Consequently, doctors exercise caution while choosing oral antibiotics [14]. Developing effective antibiotic alternatives is becoming more crucial for safeguarding and advancing global public health [11-13].

This study aimed to identify the most common UTI causing bacteria and to identify the most culture sensitive antibiotics.

METHODOLOGY

This cross-sectional study was conducted in Al-Qasim General Hospital, Babil Governorate of Iraq from the beginning of September 2023 to the end of February 2024. The study included 208 samples from patients suffering from signs and symptoms of UTI with an age range of 18 - 50 years. Only 100 patients showed positive results for the presence of significant bacteriuria. A midstream urine sample was obtained from each participant and cultured on blood agar, MacConkey agar and mannitol salt agar (Figure 3). All isolates were identified using the VITEK2 system, and then sensitivity testing was performed using the Disk diffusion method. The Institutional Approval Committee approved the study, and verbal consent was obtained from each adult patient participating.

Statistical Analysis:

The statistical software SPSS version 26 and Microsoft Office Excel 2010 were used to analyze the current study's data. An independent sample t-test was used to examine the variance in means between the two groups, assuming the variable follows a normal distribution. The chi-square test was used to investigate the relationship between two categorical variables. Risk was measured by estimating the odds ratio and calculating the 95% confidence interval. The threshold for statistical significance was set at a P value of less than 0.05, while the upper level of significance was set at 0.01 or less.

Ethical Approval

The required ethical clearance was received from the hospital's ethics committee, along with the permission of the patients. Prior to sample collection, all patients were thoroughly informed and given the option to grant their permission for the purposes of testing and publishing of findings.

RESULTS

Frequency Distribution of Patients with UTIs According to Gender

The present study included 100 patients with UTIs. Female patients with UTI comprised 77%, more than male patients, 23%. Figure 1

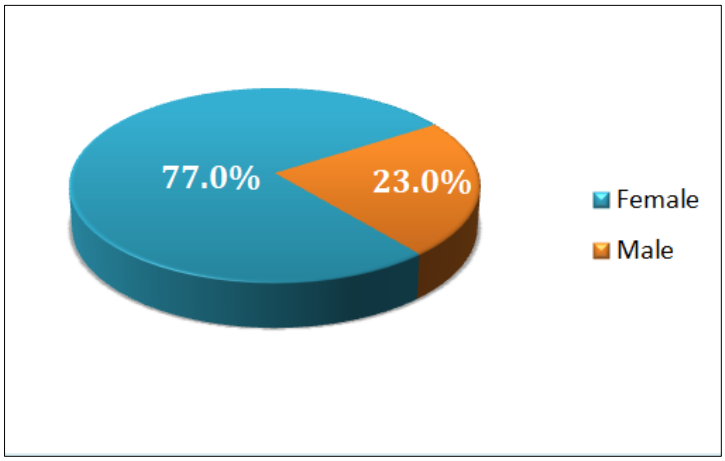

Figure 1: A pie chart illustrating the distribution of patients with UTI based on age

Frequency Distribution of Patients with UTIs According to Age

The frequency distribution of patients with UTI based on age is shown in Figure 2, and comparison of mean age and frequency distribution according to age between male and female patients with UTI is shown in Table 1.

In the present study, the average age of male patients with UTI was 37.47±5.50 years, whereas the average age of female patients was 32.92±8.01. The afflicted age group ranged from 18 to 50 years. On the other hand, the present study observed that most patients were 30-39 years old. The present study showed a

notable disparity in the average age and age distribution between females and males affected by UTI.

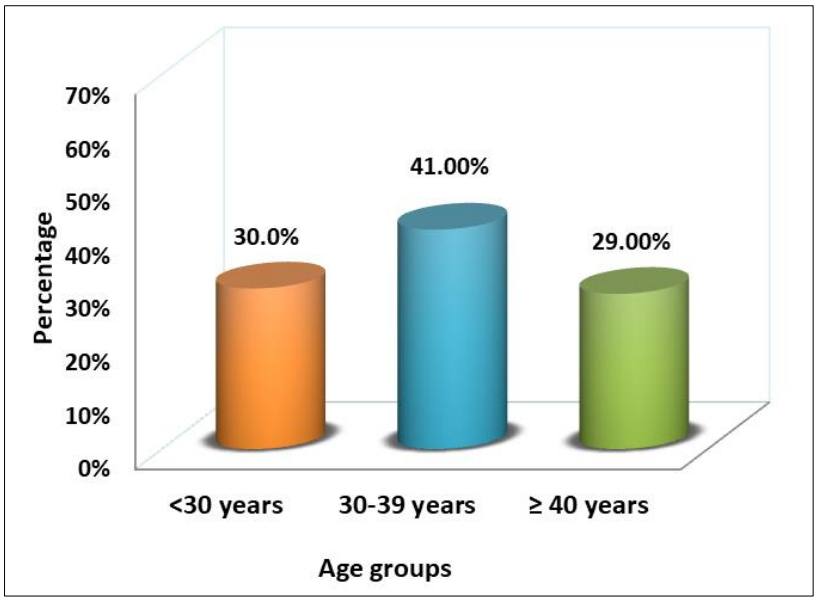

Figure 2: Histogram showing the frequency distribution of patients with *UTI* according to age

Table 1: Comparison of mean age and age group between male and female patients with UTI

Age (years)	**Gender**		***p*-value**
	Male *n* (%)	**Female *n* (%)**	
< 30 year, *n* (%)	1 (4.3 %)	29 (37.7 %)	0.009 C S
30-39 year, *n* (%)	13 (56.5 %)	28 (36.4 %)	
> 40 year, *n* (%)	9 (39.2 %)	20 (26.0 %)	
Mean ±SD	37.47 ±5.50	32.92 ±8.01	0.012 I S
Range	27-46	18-50	

n: number of cases; **SD**: standard deviation; C: Chi-square test; **NS**: not significant at $p > 0.05$; **I**: independent samples *t*-test;

Isolation and Distribution of Bacterial from Patient with UTIs

Out of 208 samples, the culture results showed that only 100 samples were positive for the presence of significant bacteria. While the rest of the samples showed contaminated or non-bacterial growth. Bacterial isolated associated UTI were shown in Table 2.

Table 2: Bacterial infection associated UTI

Characteristic	**Patients with UTI**	**P value**
Bacterial species		
Escherichia coli, *n* (%)	71 (68.3 %)	p < 0.001 HS
Staphylococcus aureus, *n* (%)	8 (7.8%)	
Klebsiella Pneumoniae, *n* (%)	6 (5.8 %)	
Enterococcus faecalis, *n* (%)	7 (6.7%)	
Staphylococcus saprophyticus, *n* (%)	4 (3.8%)	
Pseudomonas aeruginosa, *n* (%)	4 (3.8%)	
Streptococcus agalactiae, *n* (%)	2 (1.9%)	
Proteus mirabilis, *n* (%)	1 (0.9 %)	
Serratia marcescens, *n (%)*	1 (0.9 %)	

n: number of cases;

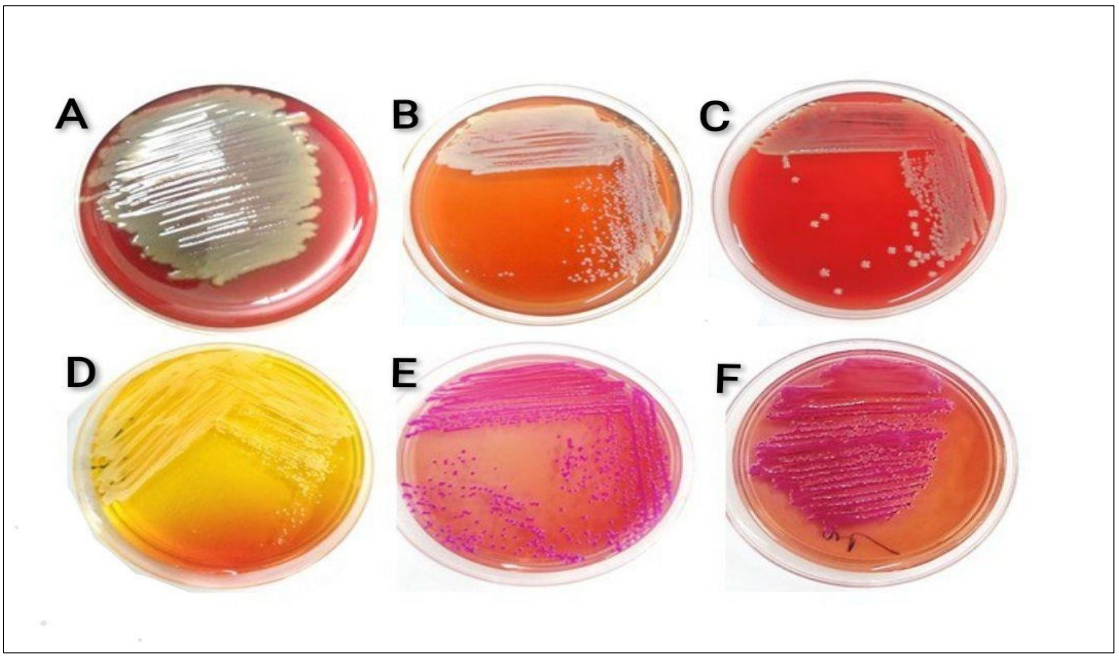

Figure 3: Bacterial growth on different media.

A- *Proteus mirabilis* on blood agar. B- *Staphylococcus aureus* on blood agar. C- *E. coli* on blood agar. D- *Staphylococcus aureus* on mannitol salt agar. E- *E. coli* on MacConkey agar. F- *Klebsiella pneumoniae* MacConkey agar

Antibiogram Testing

Antibiogram testing was performed with selected antibiotics commonly used and recommended by CLSI -2023. The antibiotic susceptibility test was assessed by disc diffusion method in vitro. Figure 4 and Tables 3, 4.

Table 3: Sensitive Testing against different species of gram negative bacteria

Classes	**Antibiotics**	***E. coli (71)***		***K. pneumoniae (6)***		***P. mirabilis (1)***		***P. aeruginosa (4)***		***Serratia marcescens (1)***	
		S	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**	**R**
Aminoglycoside	Amikacin	21 *(29.5%)*	50 *(70.5%)*	2 *(33.3%)*	4 *(66.7)*	1 *(100%)*		2 (50%)	2 (50%)	1 *(100%)*	
	Gentamicin	27 (38%)	44 (52%)	5 *(83.3%)*	1 *(16.7%)*		1 *(100%)*			1 *(100%)*	
	Tobramycin	14 (19.7%)	57 (80.3%)	2 *(33.3%)*	4 *(66.7)*		1 *(100%)*	1 *(25%)*	3 *(75%)*		
Cephalosporin	Ceftriaxone	27 (38%)	44 (52%)	5 *(83.3%)*	1 *(16.7%)*	1 *(100%)*		1 *(25%)*	3 *(75%)*	1 *(100%)*	
	Ceftazidime	35 (49.3%)	36 (50.7%)	4 *(66.7)*	2 *(33.3%)*	1 *(100%)*		2 (50%)	2 (50%)	1 *(100%)*	
Quinolones	Ciprofloxacin	64 (90.1%)	7 (9.9%)	6 *(100%)*		1 *(100%)*		2 (50%)	2 (50%)	1 *(100%)*	
	Levofloxacin	57 (80.2%)	14 (19.8%)	5 *(83.3%)*	1 *(16.7%)*	1 *(100%)*		1 *(25%)*	3 *(75%)*	1 *(100%)*	
Penicillin	Piperacillin	21 (29.5%)	50 (70.5%)		6 *(100%)*		1 *(100%)*	2 *(50%)*	2 *(50%)*		1 *(100%)*
	Ampicillin	25 (35.2%)	46 (64.8%)	1 *(16.7%)*	5 *(83.3%)*		1 *(100%)*				1 *(100%)*
	Amoxicillin	28 (39.4%)	43 (60.6%)	2 *(33.3%)*	4 *(66.7)*		1 *(100%)*				1 *(100%)*
Carbapenems	Imipenem	70 (98.5%)	1 (1.5%)	6 *(100%)*		1 *(100%)*		4 *(100%)*		1 *(100%)*	
	Meropenem	65 (91.5%)	6 (9.5%)	5 *(83.3%)*	1 *(16.7%)*	1 *(100%)*		3 *(75%)*	1 *(25%)*	1 *(100%)*	
Sulfonamides	Trimethoprim	21 (29.5%)	50 (70.5%)	5*(83.3 %)*	1 *(16.7%)*	1 *(100%)*		1 *(25%)*	3 *(75%)*	1 *(100%)*	
	Sulfamethoxazole	27 (38%)	44 (52%)	4 *(66.7)*	2*(33.3 %)*	1*(100 %)*		2*(50 %)*	2 *(50%)*	1*(100 %)*	
Macrolides	Azithromycin	22(30.9 %)	49(69.1 %)	3 (50%)	3 (50%)		1*(100 %)*		4*(100 %)*		1*(100 %)*

Table 4: Sensitive Testing against different species of gram positive bacteria

Classes	Antibiotics	*S. aureus (8)*		*S. Saprophyticus (4)*		*S. Agalactica (2)*		*E. faecalis (7)*	
		S	R	S	R	S	R	S	R
Aminoglycoside									
	Gentamicin	4*(50%)*	4*(50%)*	3*(75%)*	1*(25%)*	1*(50%)*	1*(50%)*	-	-
Quinolones	Ciprofloxacin	6*(75%)*	2*(75%)*	4*(100%)*		2*(100%)*		3*(42.8%)*	4*(57.2%)*
	Levofloxacin	7*(87.5%)*	1*(12.5%)*	4*(100%)*		2*(100%)*		4*(57.2%)*	3*(42.8%)*
Penicillin	Ampicillin	-	-	-	-	-	-	4*(57.2%)*	3*(42.8%)*
	Vancomycin	-	-	-	-	2*(100%)*		6*(85.7%)*	1*(14.3%)*
Sulfonamides	Trimethoprim	3*(37.5%)*	5*(62.5%)*	3*(75%)*	1*(25%)*	2*(100%)*		-	-
	Sulfamethoxazole	4*(50%)*	4*(50%)*	3*(75%)*	1*(25%)*	2*(100%)*		-	-
Macrolides	Azithromycin	2*(25%)*	6*(75%)*	2*(50%)*	2*(50%)*	1*(50%)*	1*(50%)*	3*(42.8%)*	4*(57.2%)*

S; Sensitive, R; resistant

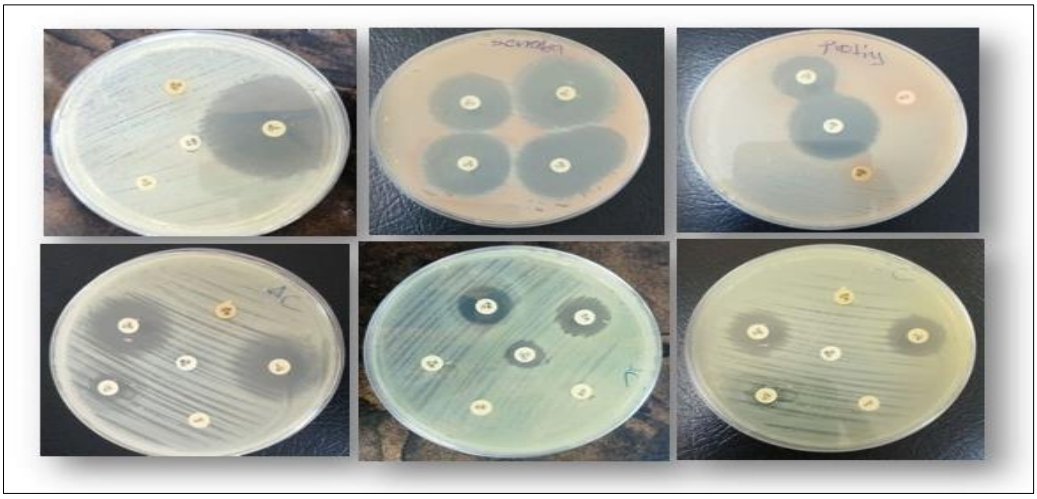

Figure 4: Antibiotic susceptibility test for bacterial isolates

DISCUSSION

According to the data, the present study found that females make up the majority of UTI cases compared to males, which is consistent with the findings of most prior studies [18–20], because of the structural differences between males and females, the length of the urethra in women is shorter than in men. As a result, bacteria have a shorter distance to travel to reach the bladder in women [21, 22]. In addition, hormonal fluctuations in women across the menstrual cycle are essential in increasing susceptibility to UTIs [21-24]. There appears to be a clear statistical significance in the age distribution between males and females, and as shown in Table 1, there is a high percentage of females who are less than 30 years old compared to males. This result was confirmed by a previous study conducted on 13,820 samples positive for bacterial infection, in which the ratio of females to males was highest in the age group 18 to 29 years [25].

As shown in the table (1), the average age of females was less than that of males, and the difference between the ages was statistically significant, as it was p = 0.012. In addition, the standard deviation for females, 8.01, was higher than that of males, 5.50. This means that the ages of females are more widespread than those of males. In general, the current study found that females with UTIs tended to be younger than males with UTIs.

Most studies found that Gram-negative bacteria are the most common cause of UTIs [18-26], rather than Gram-positive bacteria [27–30], and this is consistent with the results of the current study. The rationale for this is the availability of Gram-negative bacteria in high proportions in the intestine, in addition to the presence of unique virulence factors in Gram-negative bacteria, for example, the bacteria having a unique structural structure or the presence of special adhesion proteins, These proteins facilitate the attachment of bacteria to urinary epithelial cells, thus increasing the rate of spread of urinary tract infection [31].

The current study revealed that the most common cause of UTIs is *E. coli*, which belongs to the Enterobacteriaceae family and is responsible for more than half of the cases.

The elevated incidence of E. coli infection may be attributed to its presence as a constituent of the natural microbial community in the human intestinal tract. As a result, it may readily establish itself in the urinary system [18]. Moreover, many researches in different regions have concluded that E. coli is the dominant bacteria

causing UTIs (20,24,26,27,32,33). The present study found that *E. coli* was sensitive to Imipenem, Meropenem, Ciprofloxacin, and Levofloxacin (98.5%,91.5%,90.1%, 80.2%), respectively, while there were resistant to Tobromycin(80.3%) and Amikacin(70.5%).

However, another study reported that *E. coli* was resistant to Imipenem and Meropenem [34]. Another studys conducted in Iraq reported that *E. coli* was highly sensitive to Imipenem and Meropenem, At the same time,it had high resistance to Cephalosporin, including the third generation [18-35], which was approved by current study. However, the pattern of antibiotic sensitivity to these bacteria varies in different studies. The effectiveness of antibiotics arises from their restricted usage under medical supervision and their exclusive use with a prescription.

Moreover, the high antibiotic resistance can be partly explained the high antibiotic abuse and overuse rate in some areas. As for Gram-positive bacteria, *Staphylococcus aureus* was the dominant bacteria and second most common cause of UTIs, followed by *Enterococcus faecalis*. Previous studies confirmed that *S. aureus* is one of the most common types of Gram-positive bacteria UTIs [18-37].

S. aureus has shown sensitivity to Ciprofloxacin (75%) and Levofloxacin (87.5%) while it has shown resistance to Trimethoprim and Azithromycin. These results are supported by many researches around the world [18-39]. *E. faecalis* was resistant to Azithromycin and Ciprofloxacin (57.2%) for both, while it was sensitive to Vancomycin (85.7%). The results reached by Alzahrani [40], showed agreement with the results of the current study, as *E. faecalis* was completely sensitive to Vancomycin, in contrast, it was resistant to Ciprofloxacin by 66.6%. While ALfuadi 2023 in Al-Diwaniyah came up with variant results, his showed that *E. faecalis* was resistant to both Vancomycin and Ciprofloxacin [41].

The results obtained by the Al-Naqid in 2020 were not consistent with ALfuadi study but rather were consistent with the results of current study, where he concluded that the rate of resistance of *E. faecalis* to Vancomycin was 12.5%, while the rate of resistance to Levofloxacin was 41% [18].

In the current study found that *Klebsiella pneumoniae* is the most common pathogen after *E. coli* among Gram-negative bacteria. *K. pneumoniae* was highly susceptible to Ciprofloxacin (100%) and Imipenem (100%), followed by Gentamicin, Meropenem, Trimethoprim, and Ceftriaxone (83.3%) for all of them. While, it was completely resistant to Piperacillin (100%), as many studies support this study [16-27].

In addition, *Pseudomonas aeruginosa* was highly resistant to Azithromycin, Tobramycin, Ceftriaxone, and Levofloxacin (100%, 75%, 75%, 100%), respectively, while it was highly susceptible to Imipenem (100%) and Meropenem (75%). These results are almost identical to results of a study conducted by Alzahrani in Saudi Arabia [40], where *P. aeroginosa* was sensitive to Imipenem and Meropenem at a rate of 73% and 71%, respectively, and was completely resistant to Levofloxacin.

Al-Naqid showed a different sensitivity pattern, as he reported that *P. aeroginosa* was sensitive to Tobramycin and Piperacillin at a rate of 77% to both [18]. The regional variation in antibiotic resistance of bacteria that cause UTIs is evident due to different treatment and prevention strategies and the result of excessive use of medications without a prescription by populations in different geographical areas [23-42]. This variation calls for an urgent need to monitor and provide Up-to-date information to improve the selection of appropriate treatments.

CONCLUSIONS

The results revealed that females are more susceptible to urinary tract infections than males. Furthermore, females with UTIs tend to be younger than males with UTIs. Gram-negative bacteria are prevalent and E. coli is the most common cause of UTIs.

Imipenem remains the most effective treatment for inhibiting most bacteria causing UTIs. Meropenem, ciprofloxacin, and levofloxacin are viable treatment alternatives. Regarding antibiotics in Iraq, the study recommended a dynamic guideline for uncomplicated UTIs based on the annual updating of culture and sensitivity reports.

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Conflict of Interest: No conflict of interest is present in the current study.

Ethics Approval: The current research has followed the accepted principles of ethical conduct by University of Al-Qadisiah, Medicine College.

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