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Original Article

Study of Antimicrobial susceptibility test for *Moraxella catarrhalis* in Hospitalized Patients of respiratory tract Infection

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ABSTRACT

Urinary tract infections (UTIs), mainly caused by *Escherichia coli*, are a major health concern, affecting over 150 million people globally each year, with a lifetime prevalence of 40-50% among females. This study, conducted from May 1, 2023, to February 29, 2024, in three hospitals in Kirkuk City, aimed to assess the frequency of multidrug-resistant (MDR) bacteria in UTI patients and examine the effect of seasonal variation. A total of 250 patients aged 17-78 were enrolled, with 110 confirmed UTI cases. Bacterial identification and antibiotic susceptibility testing were performed using the VITEK2 system and the disk diffusion method. Results showed a higher incidence of UTIs during the summer months, likely due to dehydration and increased bacterial growth. *E. coli* was the most common pathogen (39.1%), followed by *Klebsiella* spp. (22.7%) and *Proteus mirabilis* (14.5%), all exhibiting significant multidrug resistance. The findings highlight a seasonal pattern in UTIs, particularly among females, with higher prevalence in warmer months.

Keywords: UTIs, uropathogenic bacteria, Kirkuk city, Antibiotic resistance, *E. coli*

Introduction

UTIs are associated with increased morbidity, mortality, and treatment costs, in addition to being among the most prevalent bacterial infections in community healthcare settings (1, 2). *Escherichia coli* is the most frequently isolated uropathogen; however, *Klebsiella*, *Staphylococcus*, *Enterococcus*, *Enterobacter*, and *Citrobacter* are also commonly implicated (1, 3). Recent research indicates that Gram-negative bacteria are the primary cause of UTIs (4). Globally, over 150 million cases are recorded annually (5, 6). Uropathogenic *E. coli* (UPEC) strains possess multiple virulence factors including fimbrial and afimbrial adhesins, toxins, siderophores, and capsular polysaccharides that facilitate bacterial adhesion and infection within the urinary tract (7, 8). Clinical studies have highlighted a notable prevalence of antibiotic resistance among uropathogens (9-11). The excessive use of antibiotics is considered the primary driver behind the rise of multidrug-resistant (MDR) UPEC isolates (12).

The emergence of MDR UPEC is increasingly observed in both hospital and community settings, with MDR-UPEC strains reported in 68% of hospitalized patients and 61% of community-acquired UTI cases (13-15). Contributing factors include inappropriate antibiotic use, limited laboratory infrastructure, and insufficiently trained healthcare personnel, particularly in resource-limited facilities (16). The spread of MDR strains leads to recurrent UTIs, complications such as pyelonephritis with sepsis and preterm birth, and increased morbidity and mortality (17, 18).

Urine culture and antibiogram results typically require up to four days; therefore, empirical antibiotic therapy is often initiated before culture outcomes are available, given the complex clinical manifestations of UTIs (19-21). While the prevalence of multidrug-resistant UPEC slightly declined in the United States between 2016 and 2020, resistance to penicillin and co-resistance to penicillin and trimethoprim-sulfamethoxazole (TMP-SMX) remain concerning (22). Despite rising multidrug resistance over the past two decades, contemporary data on UPEC resistance patterns remain limited (23, 24). The increased use of broad-spectrum antibiotics reduces treatment options and contributes to higher treatment failure, morbidity, healthcare costs, and hospitalization rates (25).

Studies have also reported seasonal trends in UTI incidence. Adults, particularly females, exhibit higher susceptibility during summer, while children are more prone in winter months (26, 27). The seasonal pattern is most evident in younger adults due to behavioral changes in warmer months, whereas it is less prominent in individuals over 70, a group with generally higher UTI prevalence (28). This study aimed to (i) determine the prevalence of multidrug-resistant bacteria among individuals with UTIs and (ii) analyze the effect of seasonal variation on the incidence of UTIs caused by multidrug-resistant bacteria.

Methods

Sample Collection and Bacterial Isolation

The study was conducted from May 1, 2023, to February 29, 2024, including 250 patients under medical supervision at Azadi Teaching Hospital, Kirkuk General Hospital, and Gynecology and Pediatric Hospital in Kirkuk City. Patient information such as age, gender, marital status, education level, pregnancy status, symptoms, menstrual cycle, and medical history was collected. Participants were aged 17-78 and presented with symptoms including fever, back and abdominal pain, hematuria, vomiting, foul-smelling urine, dysuria, and frequent urination.

Patients were required to abstain from antibiotics for three days prior to urine sample collection. Samples were collected in sterile containers, inoculated onto Nutrient, MacConkey, and Blood agar plates, and incubated at 37°C for 48 hours. Bacterial isolates were identified using Gram staining, standard biochemical tests, and confirmed via the VITEK-2 compact system. Monthly UTI incidence was recorded to evaluate seasonal trends.

Antimicrobial Susceptibility Testing

The Kirby-Bauer disk diffusion method was used following Clinical and Laboratory Standards Institute (CLSI) guidelines (29). Twelve antibiotics were tested: ciprofloxacin, trimethoprim-sulfamethoxazole, nitrofurantoin, ampicillin, amoxicillin/clavulanic acid, tetracycline, ceftriaxone, gentamicin, cefuroxime, and cephalexin. Bacterial suspensions were prepared in 0.9% saline to match McFarland turbidity standards, and antibiotic discs were applied. Zones of inhibition were measured in millimeters and isolates resistant to three or more antibiotic classes were classified as multidrug-resistant (MDR).

Results and Discussion

Out of 250 participants, 110 (44%) were confirmed to have UTIs. Table 1 illustrates the monthly and seasonal variations in UTI incidence from May 2023 to February 2024.

Table 1. Seasonal fluctuation in the frequency of UTI incidents during the investigative period.

Month	UTI among the number of isolates	Percentage%
May	10	9
June	14	12.70
July	22	20
August	19	17.30
September	8	7.30
October	11	10
November	7	6.40
December	8	7.30

January	5	4.55
February	6	5.45
Total	110	100

The results of this study are consistent with previous research demonstrating a clear association between elevated temperatures and increased urinary tract infection (UTI) prevalence, particularly among females (30, 31). A notable peak in UTI cases occurred during the summer months, which may be attributed to factors such as dehydration. Reduced urine output during dehydration can limit the expulsion of urinary pathogens, increasing susceptibility to UTIs (32). Additionally, warm weather may cause the perineum to remain moist, facilitating bacterial migration from the rectum to the urethra, as supported by other studies (33, 34).

UTIs are caused by a variety of bacterial species, with uropathogenic *Escherichia coli* (UPEC) responsible for 80-90% of cases worldwide (35). In this study, *E. coli* was the most prevalent pathogen, accounting for 39.09% of isolates, consistent with previous reports (36-39). Other bacterial isolates included *Klebsiella* spp. (22.73%), *Proteus mirabilis* (14.55%), and *Staphylococcus aureus* (8.18%). *Pseudomonas aeruginosa* and *Streptococcus* spp. each accounted for 6.36%, while *Shigella* spp. showed the lowest occurrence at 2.73%. These findings align closely with results from local studies (40, 41).

Table 2. Illustrates the distribution of bacterial isolates causing urinary tract infections from the total number of positive growth samples.

UTI bacterial isolates	NO. of Isolates	Percentage
Escherichia coli	43	39.091%
Klebsiella spp.	25	22.727%
Proteus mirabilis	16	14.545%
Staphylococcus aureus	9	8.181%
Pseudomonas aeruginosa	7	6.363%
Streptococcus spp.	7	6.363%
Shigella spp.	3	2.727%
Total	110	100%

The findings of this study revealed that different bacterial species exhibited varying degrees of resistance to multiple antibiotics, as shown in Table 3. *Escherichia coli* demonstrated resistance to ciprofloxacin, tetracycline, trimethoprim-sulfamethoxazole, and ceftazidime. *Klebsiella* spp. showed resistance to amoxicillin/clavulanic acid, trimethoprim-sulfamethoxazole, and vancomycin. *Proteus mirabilis* was resistant to amoxicillin/clavulanic acid, ciprofloxacin, and tetracycline, while *Staphylococcus aureus* exhibited resistance to amoxicillin/clavulanic acid, vancomycin, and ampicillin. *Pseudomonas aeruginosa* showed resistance to ceftriaxone, gentamicin, and piperacillin, whereas *Streptococcus* spp. and *Shigella* spp. were resistant to cefuroxime and cephalexin. These results are consistent with previous studies (42-44). Overall, these findings underscore the critical importance of accurate antibiotic susceptibility testing in addressing the growing issue of multidrug-resistant (MDR) UTIs. Effective management of MDR UTIs requires consideration of patient-specific factors, rapid diagnostic testing, and awareness of local resistance patterns to optimize treatment outcomes.

Table 3. illustrates Antibiotic Resistance Profiles of Various Bacterial Species

Bacteria Species	Resistant Antibiotics
Escherichia coli	Ciprofloxacin
	Tetracycline
	Trimethoprim-sulfamethoxazole
	Ceftazidime
Klebsiella spp.	Amoxicillin/clavulanic acid
	Trimethoprim-sulfamethoxazole
	Vancomycin
Proteus mirabilis	Amoxicillin/clavulanic acid

	Ciprofloxacin
	Tetracycline
	Amoxicillin/clavulanic acid
Staphylococcus aureus	Vancomycin
	Ampicillin
	Ceftriaxone
P. aeruginosa	Gentamicin
	Piperacillin
	cefuroxime
Streptococcus spp.	cephalexin
	cefuroxime
Shigella spp.	cephalexin

Conclusion

The observed seasonal peak in urinary tract infection (UTI) cases during the summer months indicates a strong correlation between elevated temperatures and increased UTI incidence, particularly among females. *Escherichia coli* was identified as the primary pathogen, followed by *Klebsiella* spp., *Proteus mirabilis*, and *Staphylococcus aureus*. Further research into the mechanisms by which temperature influences UTI prevalence, as well as the dynamics of pathogen distribution, is warranted to improve understanding and inform effective preventive strategies against UTIs.

Conflict of Interest

The authors declare that there is no competing of interests.

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