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Antibiotic Resistance Trends in Urinary Tract Infections: A Study from the Center of Iran

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Abstract

Background: Antibiotic resistance is a critical global issue, particularly in developing countries where high population density and poverty exacerbate the problem. This study examined the prevalence and trends of antibiotic resistance in urinary tract infections (UTIs) in western Iran from 2021 to 2023.

Materials & Methods: 2,185 patients with UTI symptoms were sampled at Qaem Clinic. Urine samples were collected and cultured on Eosin Methylene Blue (EMB) and blood agar. Bacterial isolates were identified using biochemical tests, and antibiotic sensitivity testing was performed using the standard disc diffusion method.

Results: *Escherichia coli* was the most prevalent pathogen (56%), followed by *Staphylococcus aureus* (32%). The study found that 71% of isolated bacteria were multidrug-resistant (MDR), with significant resistance against oxacillin (79%) and cotrimoxazole (60%). Among Gram-negative bacteria, the highest resistance was noted for cephalothin (41%) and cotrimoxazole (36.5%).

Conclusions: These findings highlight the urgent need to regularly monitor antibiotic sensitivity patterns to inform effective treatment strategies for UTIs. Treatment should be guided by urine culture results to ensure optimal outcomes and address the rising threat of antibiotic resistance.

Keywords: Antibiotic Resistance, Multidrug Resistance, Urinary Tract Infection, *Escherichia coli*, *Staphylococcus aureus*

Introduction

The urinary system is crucial in filtering blood, regulating ion concentration, and maintaining blood volume and pressure. In healthy individuals, urine is typically sterile or contains very few microorganisms. However, UTIs are among the most common bacterial infections worldwide, affecting millions of people annually and leading to significant medical costs and a substantial impact on patients' quality of life. UTIs are particularly prevalent among women, with approximately one in three women experiencing a UTI by the age of 24 and 50 to 60% of women experiencing at least one UTI in their lifetime [1]. The financial burden of UTI management is substantial, with the United States alone spending over $2 billion annually on treatment [2]. Furthermore, UTIs contribute significantly to hospital-acquired infections, accounting for a considerable portion of the annual expenditure on such infections [3].

The emergence of antibiotic resistance has complicated the treatment of UTIs, making them more challenging to manage. Inappropriate antibiotic use, both in clinical settings and in the community, has led to a significant increase in resistance rates among uropathogens. This trend is particularly concerning in developing countries, where antibiotic stewardship programs are often lacking, and the misuse of antibiotics is widespread [4]. The situation in Iran is no different, with studies reporting high rates of resistance among common

uropathogens, particularly E. coli, the most frequently isolated bacterium in UTIs [5]. For instance, resistance rates to commonly used antibiotics such as ciprofloxacin and co-trimoxazole have been reported to be as high as 50% and 60% in some regions of Iran [6, 7].

Globally, the resistance patterns of uropathogens vary significantly by region. In developed countries like the United States and Germany, resistance rates to first-line antibiotics like ciprofloxacin and trimethoprim-sulfamethoxazole are relatively lower, ranging from 10% to 20% [8, 9]. However, in developing countries, resistance rates are often much higher, with studies from India and Pakistan reporting resistance rates of up to 70% for ciprofloxacin and 80% for co-trimoxazole [10, 11]. These regional differences highlight the importance of local surveillance and tailored treatment strategies to address the specific resistance patterns in each region.

In Iran, the rising trend of antibiotic resistance among uropathogens is a growing public health concern. Studies have shown that E. coli, the most common cause of UTIs, exhibits high resistance rates to multiple antibiotics, including beta-lactams, quinolones, and aminoglycosides [12,13]. For example, a study conducted in Tehran reported that 40% of E. coli isolates were resistant to ciprofloxacin, while 50% were resistant to co-trimoxazole [14]. Similarly, resistance to third-generation cephalosporins has been on the rise, primarily due to the production of extended-spectrum beta-lactamases (ESBLs) by uropathogens [15]. These findings underscore the urgent need for improved antibiotic stewardship, enhanced surveillance systems, and the development of alternative treatment options to combat the growing threat of antibiotic resistance in Iran.

The changing sensitivity patterns of bacteria over time and across regions pose significant challenges for treatment strategies. Therefore, antibiotic therapy should be guided by specific sensitivity and resistance patterns of the pathogens involved. This study aims to evaluate the antibiotic resistance patterns of E. coli causing UTIs in urine culture samples from patients at Qaem Clinic in Qazvin between January 2021 and January 2023. By understanding the local resistance patterns, this study seeks to contribute to developing more effective treatment guidelines and inform public health interventions aimed at reducing the burden of antibiotic resistance in Iran.

Materials and Methods

This cross-sectional study was conducted on urine samples collected from outpatients of various age groups and genders at the Qaem Clinic Clinic laboratory in Qazvin, Iran, between January 2021 and January 2023. A total of 2,185 patients presenting with clinical symptoms of UTIs were included in the study, of which 473 patients (21.64%) tested positive for UTIs based on laboratory confirmation.

Urine Culturing and Bacterial Identification: Urine specimens were cultured on Sheep Blood Agar and MacConkey Agar plates using a 1 µL calibrated loop. The plates were incubated at 37 °C for 24 hours. Positive samples had more than 10^5 colony-forming units (CFU) per milliliter. Isolates were identified using standard laboratory techniques based on morphological and biochemical characteristics [16-17].

Antibiotic Susceptibility Testing: Antibiotic susceptibility testing was performed using the disc diffusion method on Mueller-Hinton Agar (MHA). The following antibiotics were evaluated for antimicrobial resistance: Cephalexin (30 µg), Ceftazidime (30 µg), Ciprofloxacin (5 µg), Gentamicin (10 µg), Imipenem (10 µg), Cotrimoxazole (1.25/23.75 µg), Ampicillin (10 µg), Vancomycin (30 µg), Nitrofurantoin (300 µg), Amikacin (30 µg), Ofloxacin (10 µg), Norfloxacin (10 µg), Oxacillin (1 µg), and Nalidixic Acid (30 µg). Antibiotic discs were obtained from Padtanteb, an Iranian company. After placing the discs on the agar plates, the plates were inverted and incubated at 37 °C for 24 hours. Following incubation, the diameters of the inhibition zones were measured to the nearest millimeter, and the results were categorized as susceptible or resistant.

Data collected during the study were analyzed using one-way analysis of variance (ANOVA) with SPSS version 19 software. A p-value of ≤ 0.05 was considered statistically significant.

Results

Frequency of Bacteria Causing Urinary Infections: A total of 2,185 patients were sampled, with 473 (21.64%) testing positive for urinary tract infections (UTIs). Among the positive cases, 379 patients (80%) resisted at least one antibiotic. The predominant bacteria isolated included *Escherichia coli* (263 isolates, 55.6%), *Staphylococcus aureus* (153 isolates, 32.3%), and Streptococcus (38 isolates, 8%). The frequency distribution of uropathogenic bacteria is illustrated in Fig. 1.

Antibiotic Resistance Patterns: The analysis revealed that Gram-positive bacteria accounted for 57% of isolates, while Gram-negative bacteria constituted 43%. The resistance rates for E. coli to various antibiotics were as follows: oxacillin (79%), vancomycin (76%), cotrimoxazole (60%), and ciprofloxacin (58%). Among Gram-negative bacteria, the highest resistance was noted against cephalothin (41%) and cotrimoxazole (36.5%). The results are shown in Fig. 2 and Table 1.

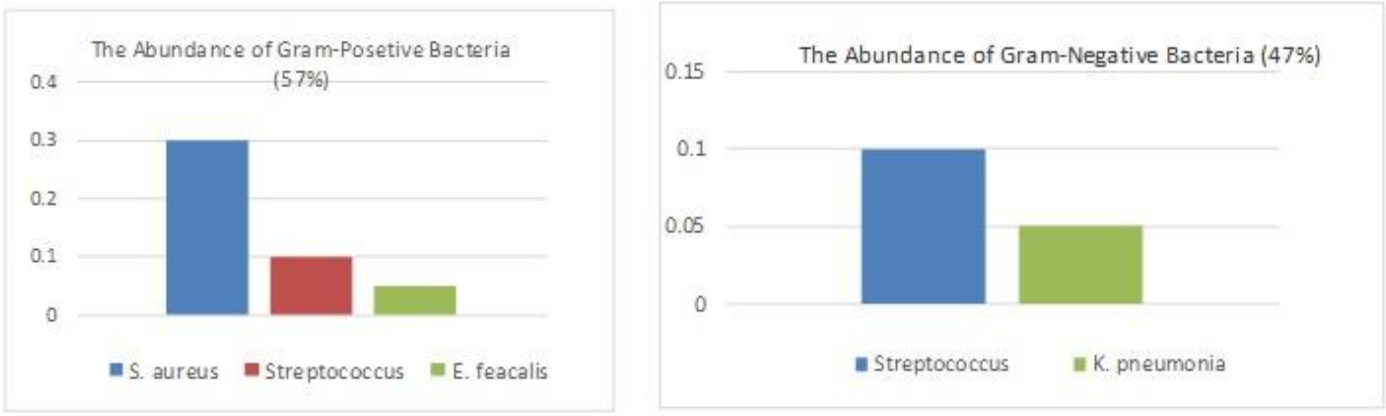

Fig. 1. Frequency distribution of UTI between different Bacteria.Source: Author

Table 1. The percentage of resistance to gram-negative and gram-positive bacteria in urine

Year / Names of microorganisms	January 2022 to March 2022			March 2022 to March 2023			March 2023 to December 2023		
	R	S	R%	R	S	R%	R	S	R%
E. coli	10	2	83%	88	32	73%	94	36	72%
E. feacalis	0	0	0	7	0	100%	4	0	100%
S. aureus	3	0	100%	78	16	83%	45	11	80%
Streptococcus	-	-		-	-		-	-	
K. pneumonia	-	-		-	-		-	-	

R: resistance; **S:** Sensitive

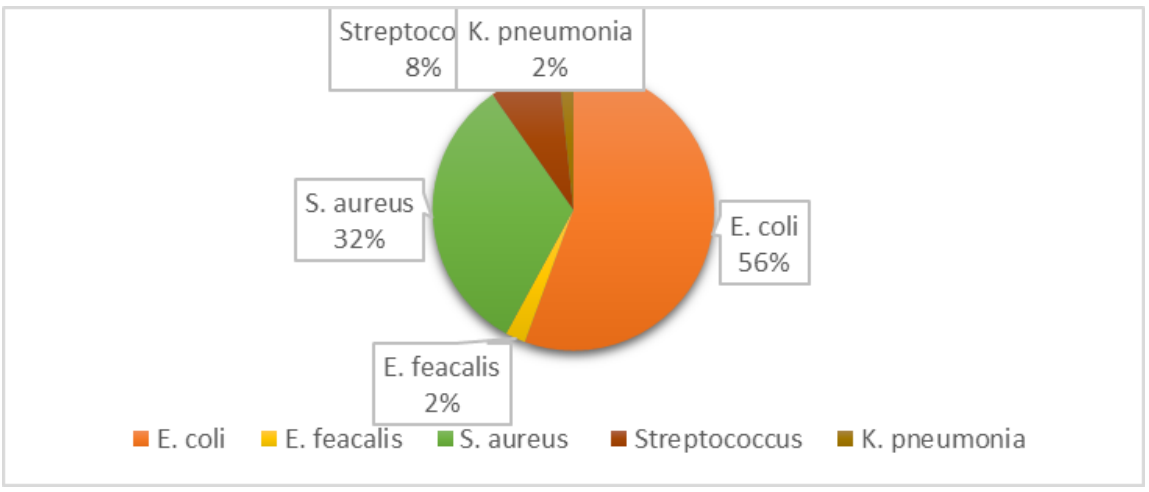

Fig. 2. Prevalence of uropathogenic bacteria obtained from urine specimens Source: Author

Classification and percentage of MDR/XDR/VRSA/VRE/PDR bacteria isolated from the urinary tract: The study identified that 71% of the isolated bacteria were multidrug-resistant (MDR), while 13% exhibited extensive drug resistance (XDR). The most frequently isolated MDR/XDR Gram-negative bacteria showed significant resistance to cotrimoxazole, nalidixic acid, and ciprofloxacin but remained relatively sensitive to ceftazidime, amikacin, and nitrofurantoin. The result is shown in Fig. 3.

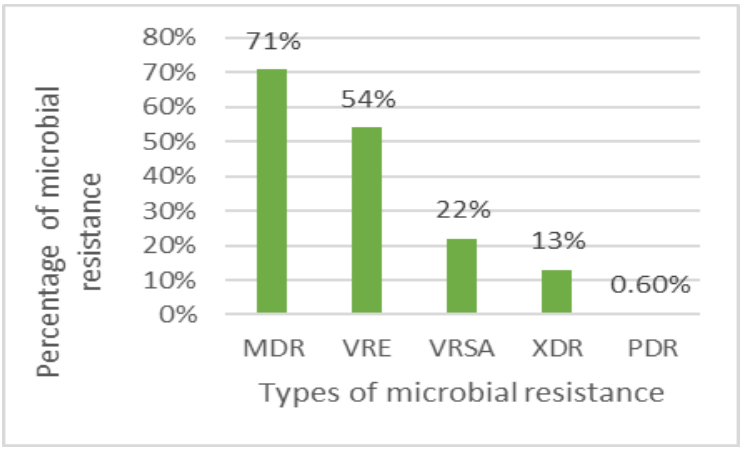

Fig. 3. Classification and percentage of MDR/XDRk/VRSA/VRE/PDR bacteria isolated from urinary tract

Table 2. Resistant Gram-positive bacteria and percentage of their antibiotic resistance

Bacteria	AM	AN	CP	GM	OX	P	SXT	V
Enterococcus spp.	4(36%)	3(27%)	3(27%)	3(27%)	3(27%)	1(9%)	3(27%)	6(54%)
Staphylococcus spp.	1(0.6%)	4(2.6%)	47(31%)	9(6%)	79(52%)	5(3.5%)	51(33%)	34(22%)

Disc diffusion susceptibility testing: Among the isolated bacteria, Staphylococcus spp. Demonstrated the highest resistance among Gram-positive bacteria (32% of total), while E. coli was the most resistant among Gram-negative bacteria (56% of the total). Both Gram-positive and Gram-negative bacteria exhibited the highest resistance to oxacillin and cotrimoxazole, respectively, while showing greater sensitivity to ceftazidime and amikacin. The results are shown in Table 2 and Table 3.

Table 3. Resistant Gram-negative bacteria and percentage of their antibiotic resistance

Bacteria	CT	NI	AM	AN	CP	GM	OX	SXT	NA	CF	IMP	NOR	OFX
Escherichia coli	1 (0.4)	16 (6%)	4 (2%)	14 (5%)	58 (22%)	21 (8%)	35 (13%)	63 (24%)	63 (24%)	42 (16%)	34 (13%)	15(6%)	13(5%)
Klebsiella spp.	0	1 (12.5%)	1 (12.5%)	0	1 (12.5%)	1 (12.5%)	1 (12.5%)	1 (12.5%)	0	2 (25%)	0	0	0

Discussion

Prevalence of Pathogens in Urinary Tract Infections (UTIs): The findings of this study confirm that bacteria from the Enterobacteriaceae family, particularly E. coli, are the most common causative agents of UTIs. This is consistent with global epidemiological data, where E. coli has been identified as the predominant pathogen in UTIs across various regions, with prevalence rates ranging from 45% to 80%. For instance, studies from Taiwan (65.9%) [6], Pakistan (79.2%) [7], and the USA (72.8%) [13] have reported similar findings, highlighting the global dominance of E. coli in UTIs. However, regional variations in pathogen prevalence are evident. For example, in South-Western Uganda, K. pneumoniae has been reported as the leading cause of UTIs [18], contrasting with our findings where Klebsiella was the second most common isolate. This discrepancy underscores the influence of geographical, environmental, and socioeconomic factors on the epidemiology of UTIs. Additionally, our study's high prevalence of E. coli aligns with findings from other regions, such as the USA [14] and Nigeria [15], where E. coli and Klebsiella are among the top pathogens. These regional differences emphasize the importance of local surveillance and tailored intervention strategies to manage UTIs effectively [19-20].

Antibiotic Resistance Patterns: The antibiotic resistance patterns observed in this study reveal significant concerns, particularly in the context of co-trimoxazole and quinolone antibiotics. Resistance rates to co-trimoxazole were 24% for E. coli and 12.5% for Klebsiella, which are lower than those reported in Tanzania (41.3% for E. coli) [21] and Iran (66% for both species) [22]. However, these rates are still alarmingly high compared to developed countries like Germany (21%) [23] and the USA (6.7%) [24], where resistance rates are significantly lower. This discrepancy may be attributed to the misuse of antibiotics, over-the-counter availability of drugs, and lack of robust antibiotic stewardship programs in developing countries. [25-26] The high resistance rates in developing countries highlight the urgent need for improved prescribing practices, public awareness campaigns, and strengthened regulatory frameworks to control the inappropriate use of antibiotics.[27-28]

Similarly, resistance rates to quinolone antibiotics, such as nalidixic acid (22%) and ciprofloxacin (24%), were notably high for E. coli. These findings are consistent with studies from Bangladesh, where nalidixic acid resistance was reported at 87.58% [29-30]. The high resistance rates observed in developing countries may be due to the widespread use of quinolones as first-line treatments for UTIs, leading to the selection of resistant strains. Given their importance in treating complicated UTIs and pyelonephritis, the rising resistance to quinolones is particularly concerning.[31-32] This underscores the need for alternative treatment options and continuous monitoring of resistance patterns to guide clinical decision-making.[33]

Multi-Drug Resistance (MDR): The emergence of multi-drug resistance (MDR) in uropathogenic bacteria, particularly E. coli and K. pneumoniae, poses a significant public health challenge. Studies have reported MDR rates as high as 42.5% for E. coli and 36.0% for K. pneumoniae in various regions [34-36]. These findings emphasize the critical need for continuous surveillance of antibiotic resistance patterns and the implementation of effective infection control measures. The high prevalence of MDR pathogens may be driven by several factors, including the overuse of broad-spectrum antibiotics, inadequate infection prevention practices, and horizontal gene transfer among bacterial species. The rise of MDR pathogens not only complicates the treatment of UTIs but also increases the risk of treatment failure, prolonged hospital stays, and higher healthcare costs. To address

this growing threat, there is a pressing need for research and development of new antibiotics, alternative therapeutic strategies (such as phage therapy or immunotherapy), and strengthened global collaboration to combat antimicrobial resistance.[37, 38]

This study has several limitations that should be acknowledged. Firstly, the lack of comprehensive data on antibiotic resistance rates of uropathogenic bacteria in Iran restricted the depth of our analysis. This limitation highlights the need for more extensive local studies to understand better the region's epidemiology of UTIs and antibiotic resistance patterns. Secondly, due to insufficient literature reporting, we could not stratify resistance rates by gender. This gap in data limits our ability to explore potential gender-based differences in antibiotic resistance, which could be important for tailoring treatment strategies. Thirdly, inconsistent age categorization in the reviewed studies made analyzing age-related differences in resistance patterns challenging [39-40]. Age is a known risk factor for UTIs, and understanding how resistance varies across different age groups could inform targeted interventions. Lastly, the absence of data on resistance rates based on patient admission types (e.g., inpatient vs. outpatient) further limited our ability to provide a comprehensive overview. Future studies should address these gaps by collecting detailed demographic and clinical data to enhance understanding antibiotic resistance dynamics in UTIs.

Conclusion

In conclusion, *E. coli* is the most common cause of urinary tract infections, followed by Klebsiella and S. aureus. The study highlights significant resistance to commonly used antibiotics, indicating careful antibiotic selection is necessary. It is recommended to use broad-spectrum antibiotics for severe UTIs, particularly in patients with a history of UTIs and advanced age. Effective treatment options for E. coli infections include ceftazidime, amikacin, and nitrofurantoin, while gentamicin and nalidixic acid should generally be avoided. The rising issue of antibiotic resistance necessitates responsible prescribing practices and ongoing surveillance to mitigate its impact on public health.

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Conflict of interest

None declared.

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Ethical Considerations

All procedures performed in this study involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Code of Ethics

This research was approved by Zanjan University of Medical Sciences under the ethical code ZUMS.REC.1394.322.

Authors' Contributions

Mahfam Alijaniha: Contributed to the original writing, conceptualized the study, and collected the data; Mahdin Alijaniha: Was responsible for language editing and data analysis; Mahdi Mirzaalimohammadi: Contributed to the manuscript revision and performed the statistical work.

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