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Optimizing Urinary Tract Infection Treatment: The Role of Antibiograms in Bridging Microbiology and Clinical Practice

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Abstract

Urinary tract infections (UTIs) are among the most common bacterial infections, affecting approximately 150 million individuals globally. Over 95% of cases are bacterial, with fungal and rare viral infections also contributing. The extensive use of empirical antimicrobials has led to rising resistance, necessitating regular surveillance.

This study sought to determine the prevalence, associated risk factors, and antibiotic susceptibility patterns of uropathogenic isolates. A retrospective analysis was conducted from January 2022 to January 2023 in the Department of Microbiology. Midstream urine samples (10 ml) were collected in sterile containers and processed using a semiquantitative method. Significant bacteriuria ($\geq 10^5$ CFU/ml) was identified based on the Kass criteria. Pathogen identification was performed using culture characteristics, Gram staining, and biochemical assays. Antimicrobial susceptibility was determined by the Kirby–Bauer disc diffusion method. A chi-square test for independence was employed, and proportions of dominant pathogens were compared utilizing a Z-test. A chi-square test or Fisher's exact test was utilized to evaluate differences in antimicrobial susceptibility rates between organisms. Of a total of 5,257 samples, 1,231 (23.41%) had significant bacteriuria. Gram-negative bacteria (65%) were predominant, followed by Gram-positive bacteria (26%) and *Candida* spp. (8%). Among Enterobacteriaceae, amikacin had the highest susceptibility (73%), followed by piperacillin-tazobactam (51.4%) and imipenem (51.2%). Gram-positive isolates were 100% sensitive to vancomycin and linezolid.

Conclusions: The rising antimicrobial resistance in uropathogens underscores the need for continuous surveillance and antibiogram updates to guide empirical therapy and improve treatment outcomes

Manuscript Text

Introduction: Urinary tract infections (UTIs) represent the most common infectious ailments worldwide, impacting individuals spanning all age demographics, from the youngest to the oldest members of the population.[1] While a variety of microorganisms can cause UTIs, bacterial infections account for more than 90% of cases, with fungal and rarely viral infections also contributing. [2] UTIs are the leading cause of healthcare-associated infections among hospitalized patients and the second leading cause of hospital visits.[3] Urinary tract infections (UTIs) are predominantly caused by a variety of microbial pathogens. These include gram-negative bacilli such as *Escherichia coli* (*E. coli*), *Klebsiella pneumoniae*, *Acinetobacter* species, *Pseudomonas aeruginosa*, and

Proteus species. Additionally, gram-positive organisms contribute to UTIs, notably *Staphylococcus aureus*, coagulase-negative *staphylococci*, and *Enterococcus* species.[4] Compared with men, women are more frequently affected by UTIs because of factors such as a smaller urethra, a lack of bactericidal prostatic secretions, a higher bacterial load in the urothelial mucosa, urinary tract obstructions, sexual activity, and pregnancy.[5] Elderly males, individuals with enlarged prostates, and those experiencing neurogenic bladder dysfunction are at an increased risk of developing urinary tract infections.[6] The classification of urinary tract infections (UTIs) encompasses two primary categories: uncomplicated and complicated. Uncomplicated UTIs manifest in the absence of functional or anatomical abnormalities within the urinary system. In contrast, complicated UTIs develop when such abnormalities are present, consequently elevating the risk of infection.[7] Failure to treat urinary tract infections can result in serious sequelae, such as scarring of the kidneys, elevated blood pressure, and long-term renal dysfunction.[8] The diversity of uropathogens varies by region, and treatment has become increasingly challenging owing to evolving antibiotic resistance.[9] Regular monitoring is therefore crucial for developing effective empirical antimicrobial therapy guidelines. The objective of this investigation was to elucidate the predominant uropathogens associated with urinary tract infections (UTIs) and to determine their respective antimicrobial susceptibility profiles.

Materials and Methods:

Study design & duration: A retrospective observational investigation spanning from January 2022 to January 2023 was undertaken within the Microbiology Department, encompassing a full calendar year.

Sample processing: During the investigation period, we conducted analyses and processing on a cohort of 5,257 clean-catch midstream urine specimens.

Samples were collected from patients of all age groups, from pediatrics to the older age group, with suspected UTI symptoms. A semiquantitative methodology was employed for the urine cultures, involving the inoculation of 0.001 ml of urine onto cysteine lactose electrolyte-deficient (CLED) agar using a calibrated loop. [10, 11] The results were assessed following the standard Kass criteria used for significant bacteriuria. [10,11] Significant bacteriuria was considered when the culture showed growth of ≥10^5 colony-forming units (CFU)/ml and symptoms of UTI. Microbial cultures demonstrating greater than three non-identical colony morphologies were designated as substantial contaminants.[12]

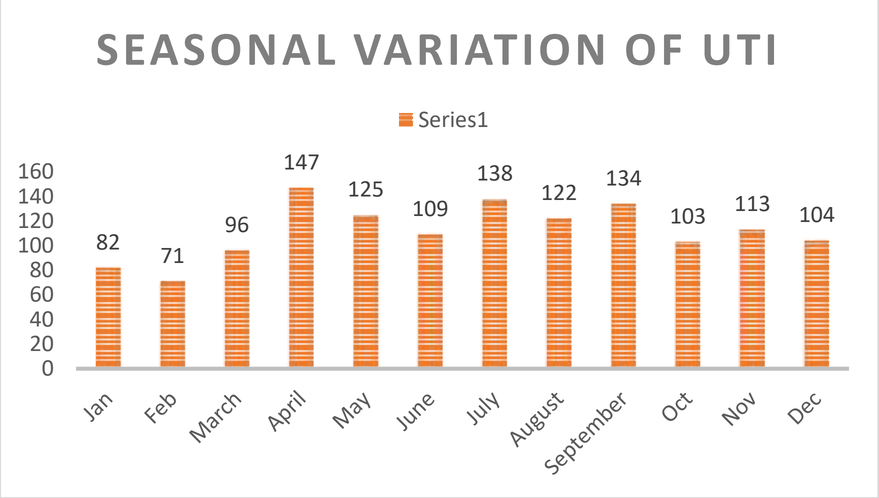
Identification of organisms: Organisms were identified through Gram stain (gram-positive/gram-negative), motility (motile/nonmotile), and biochemical characteristics per established microbiological methods, whether pathogenic or not.[13]

Antimicrobial susceptibility: The evaluation of antimicrobial susceptibility was conducted utilizing the Kirby–Bauer disk diffusion technique on the Mueller–Hinton agar medium.[14] Antimicrobials such as amikacin (30 mcg), amoxicillin-clavulanic acid (20/10 mcg), ceftazidime (30 mcg), cefotaxime (30 mcg), cefoxitin (30 mcg), ciprofloxacin (5 mcg), norfloxacin (5 mcg), gentamicin (10 mcg), imipenem (10 mcg), nitrofurantoin (300 mcg), piperacillin-tazobactam (100/10 mcg), cotrimoxazole (25 mcg), doxycycline (30 mcg), clindamycin (2 mcg), erythromycin (15 mcg), penicillin (10units), and vancomycin (30 mcg) were used, The interpretation of the data was conducted in alignment with the standards set forth by the Clinical Laboratory Standards Institute guidelines(CLSI).[15] The results were analyzed via Microsoft Excel, 2010 version.

Statistical analysis: A chi-square test for independence was performed to assess the association between gender and UTI prevalence and, the significance of UTI prevalence across different age groups. No significant seasonal variation was reported. Proportions of dominant pathogens were compared using a Z-test for proportions or chi-square tests. Relative frequencies of bacterial groups tested for significance using an ANOVA or Kruskal-Wallis test. A chi-square test or Fisher's exact test was used to evaluate differences in antimicrobial susceptibility rates between organisms. A paired t-test was applied to compare susceptibility patterns of individual antibiotics across multiple isolates.

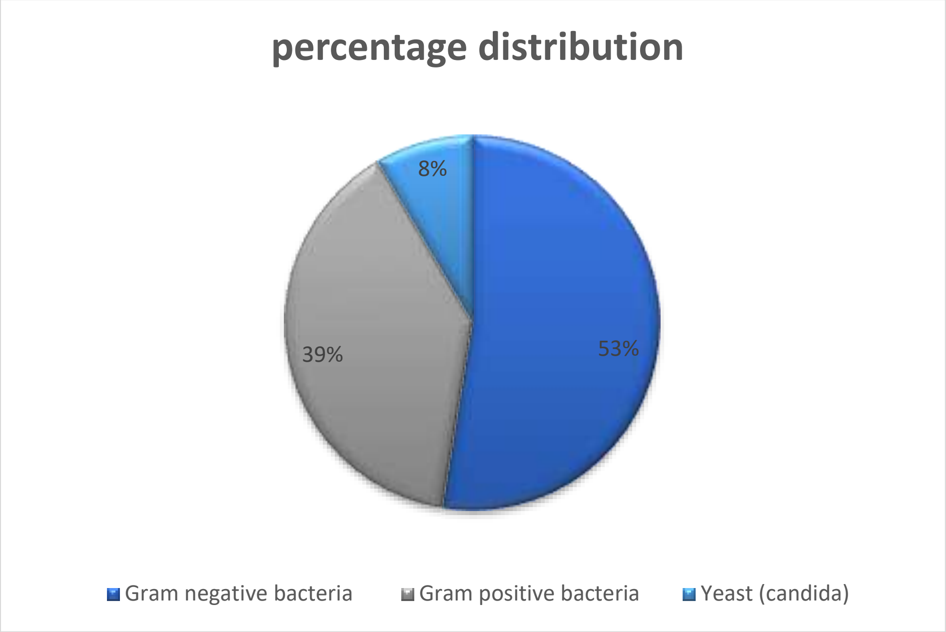
Results: During the study period, a total of 5257 cases of symptomatic UTIs were registered, 4026 of which were excluded because there was no significant growth. The overall prevalence of UTIs was 23.41%. A total of 1231 positive urine samples presented significant bacterial/fungal growth, including 467 (38%) samples from males and 764 (62%) from female patients. A high prevalence was observed among females of middle-aged (31-45 years) patients, and a high prevalence was observed among elderly (>45 years) male patients. The prevalence of infection was not affected by seasonal variation, although the hot–humid summer (April-September) was responsible for a large number of cases. [Figure1]

Figure 1: **Prevalence of UTI for seasonal variation**



The analysis of total positive samples revealed a predominance of Gram-negative isolates, accounting for 53% of the cases. Gram-positive isolates constituted 39% of the samples, while Candida species isolates represented the remaining 8% [Figure 2].

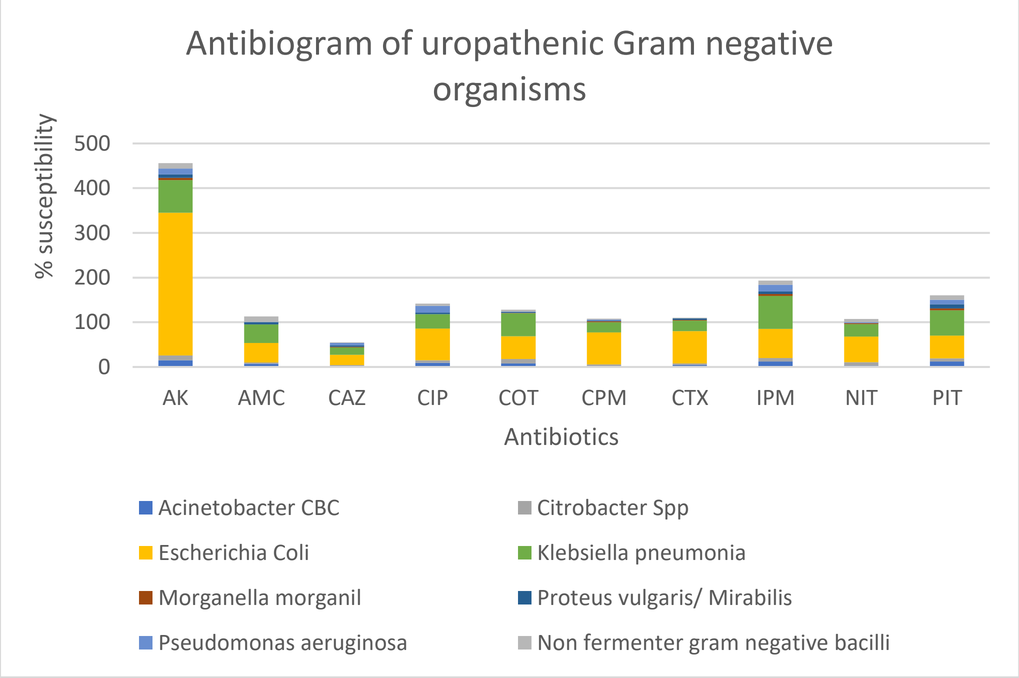
Figure 2: Organism distribution among positive isolates



Among the uropathogens, Enterobacteriaceae, i.e., *E. coli* (68.91%), followed by *Klebsiella pneumoniae* (15%), *Citrobacter* spp. (2.60%), *Proteus* spp. (1.68%), and *Morganella morganii* (0.91%), were the second most common pathogens, followed by nonfermenter gram-negative bacilli (NF-GNB), i.e., *Acinetobacter* spp. (5.51%), *Pseudomonas aeruginosa* (3.21%), and other nonfermenter GNB, account for 1.83% of the cases causing UTIs [Table 1]. Among the gram-positive cocci, *Staphylococcus aureus* (67.2%), *Enterococcus* spp. (26.4%), and other coagulase-negative staphylococci (6.25%) were ruled out.

The overall antimicrobial susceptibility patterns of the urinary bacterial isolates were as follows: amikacin (71%), imipenem (54%), and piperacillin-tazobactam (37%), followed by ciprofloxacin (35%), amoxicillin-clavulanic acid (33%), nitrofurantoin (28%), and sulfamethoxazole/trimethoprim (Cotrimoxazole-27%). The organisms showing considerable resistance to cefotaxime (19%), cefepime (18%), and ceftazidime (16%) had low susceptibilities, ranging from 16-19%. However, amikacin (71%), carbapenem (54%), and piperacillin-tazobactam (37%) have relatively good susceptibility, thus providing a better option for UTI treatment. Percentage susceptibility pattern of gram-negative organisms to antimicrobial agents. [Figure 3]

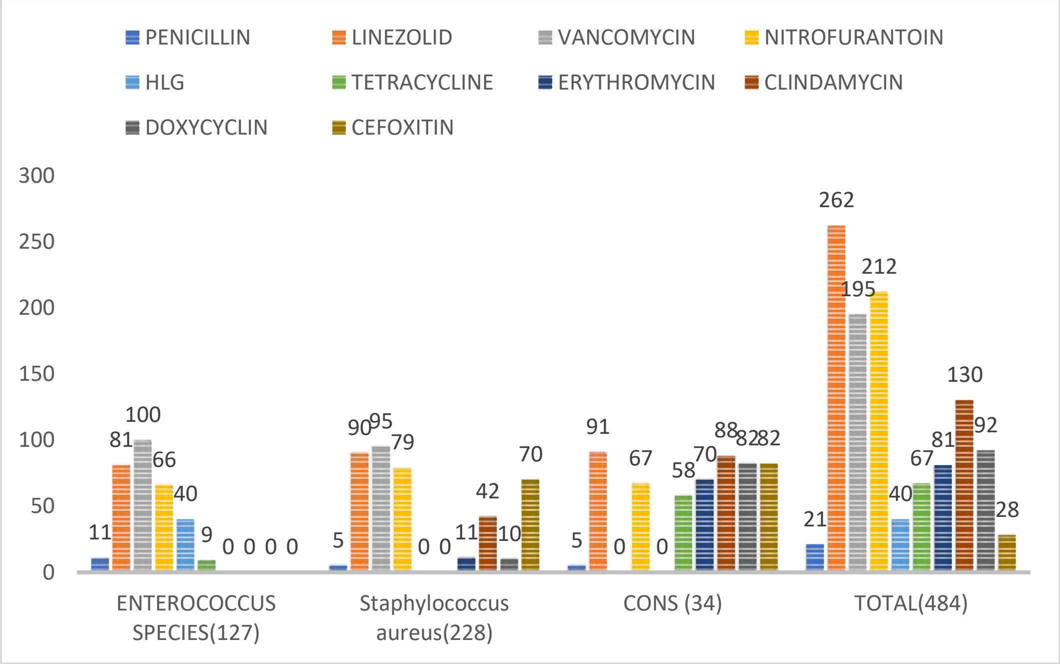
Figure 3: Uro-pathogenic gram-negative Organism & their Antibiogram



Abbreviations: N= number of isolates, AK-Amikacin, AMC-Amoxycillin-clavulanic CAZ-Ceftazidime, CIP-Ciprofloxacin, CPM-Cefepime, CTX-Cefotaxime, IPM-Imipenem, PIT- Piperacillin-tazobactam

In the analysis of gram-positive microorganisms, Staphylococcus aureus isolates demonstrated complete susceptibility to both linezolid and vancomycin (100% each), with nitrofurantoin showing efficacy against 80% of the isolates. The majority of these strains were resistant to erythromycin (88%), followed by doxycycline (90%). All *enterococcus* spp. were sensitive to linezolid (100%) and vancomycin (100%), followed by nitrofurantoin and amoxicillin-clavulanic acid, with sensitivities of 69.2% and 53.8%, respectively. Most of the *Enterococcus* spp. Isolates were resistant to norfloxacin (92.3%), followed by erythromycin (62%) and gentamicin (69.2%). [Figure 4]

Graph 4: Gram-positive bacterial isolates and their antimicrobial susceptibility pattern (In percentage)



Discussion

The prevalence of UTIs can vary based on geographic location and regional differences within a country.[15] In our study, the occurrence of UTIs was 23.41%. Comparable findings were reported by Arjuna M et al., who observed a prevalence of 34.5% in India, and by Mehta et al., who documented a prevalence of 36.6%. [16,17] These variations in findings may be attributed to differences in climate, host factors, healthcare practices, standards of living, educational levels, and hygiene practices across geographic regions.[18]

The elevated incidence of urinary tract infections (UTIs) in females can be attributed to multiple factors, including the anatomical proximity of the urethral meatus to the anus, a comparatively shorter and wider urethra, pregnancy, and a relatively less acidic pH of the vaginal surface. [19,20] The isolated microorganisms predominantly included gram-negative bacteria (70.8%) from the Enterobacteriaceae family. This prevalence can be attributed to several factors, including their ability to adhere to host tissues, the presence of pili and fimbriae, and interactions with P1 blood group phenotype receptors.[21] Our research revealed that 89.43% of gram-negative bacilli belong to the Enterobacteriaceae family, whereas 10.56% are nonfermenter gram-negative bacilli. Within the Enterobacteriaceae family, *E. coli* was the most frequently isolated microorganism (68.91%), followed by *Klebsiella* species (15%) and nonfermenters (10.5%). These results align with findings from several other studies conducted in India. [22,23,24] The gastrointestinal tract harbors Escherichia coli as a typical commensal organism, which possesses the potential to induce urinary tract infections.[25] In our study, among Enterobacteriaceae, amikacin had the highest susceptibility (73%), followed by piperacillin-tazobactam (51.4%) and imipenem (51.2%). The highest resistance was observed with ciprofloxacin (94%), followed by amoxicillin-clavulanic acid (Amoxiclav) and cefotaxime, particularly among Enterobacteriaceae and nonfermenters.

Imipenem and amikacin exhibited significant efficacy against gram-negative pathogens, which is consistent with findings from other studies conducted.[16,26] Amikacin (73%), piperacillin-tazobactam (51.4%), and imipenem (51.2%) demonstrated high sensitivity among Enterobacteriaceae, which aligns with findings from similar research.[27] Furthermore, nitrofurantoin was identified as the most effective drug for treating Enterobacteriaceae infections, which is consistent with results from other studies.[16,28,29] Among the nonfermenting isolates, amikacin was 68% susceptible, imipenem was 59%, and ceftazidime was 41.3%, which aligns with findings from studies conducted in India. [30] In this study, *Pseudomonas aeruginosa* exhibited 47.6% sensitivity to piperacillin-tazobactam, which contrasts with the higher sensitivity rates of 70% and 76.4% reported in other studies. [27,31] Among the gram-negative bacilli, amikacin (70%), imipenem (30%), and piperacillin-tazobactam (25%) demonstrated the highest overall susceptibility. All the gram-positive *Staphylococcus aureus* isolates presented 100% sensitivity to linezolid and vancomycin and 80% sensitivity to nitrofurantoin. Conversely, most isolates were resistant to erythromycin (88%) and doxycycline (90%). Linezolid and vancomycin have shown 100% susceptibility toward *enterococcus* species. Nitrofurantoin and amoxicillin-clavulanic acid had sensitivities of 69.2% and 53.8%, respectively. Our research revealed a high level of resistance to norfloxacin, which stands in contrast to the outcomes reported in previous studies. The Enterococcus isolates displayed significant resistance to norfloxacin (92.3%), as well as notable resistance to erythromycin (62%) and gentamicin (69.2%).

The gram-positive isolates demonstrated complete susceptibility to vancomycin, linezolid, and teicoplanin, aligning with observations reported in analogous investigations.[32]

Conclusion: Given the evolving patterns of antimicrobial susceptibility among various pathogens, understanding the antibiogram for common uropathogens within specific hospital settings is crucial to ensure effective empirical treatment. As resistance patterns among bacterial pathogens shift frequently, ongoing surveillance is essential to provide up-to-date information concerning the most effective antimicrobials for uropathogens. This perception of antibiograms for different isolates in different samples and different areas is highly valuable for identifying prevalent isolates and their susceptibility and resistance patterns. This would significantly help guide the selection of appropriate empirical antimicrobials and help mitigate the rise of antimicrobial resistance in our hospital.

Declaration statements

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- **Funding Source Statement:** None
- **Authors' Contribution:**
  - **RS**: Conceptualization, Methodology, Writing- Original draft preparation, Investigation, Reviewing and Editing
  - **SS**: Literature search, Supervision, Reviewing and Editing
  - **RKC:** Literature search, Supervision, Reviewing and Editing
  - **GD:** Literature search, Data curation, Reviewing and Editing

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Table 1: Distribution of gram-negative bacteria

	No of isolates
Enterobacteriaceae	584
Escherichia Coli	450 (68.91%)
Klebsiella pneumonaie	100 (15%)

Citrobacter species	17 (2.60%)
Proteus mirabilis	11 (1.68%)
Morganella morganii	06 (0.91%)
Non fermenter GNB	69
Acinetobacter CBC	36 (5.51%)
Pseudomonas aeruginosa	21 (3.21%)
Other NFGNB	12 (1.83%)
Total	653

Footnotes: NFGNB- Nonfermenter Gram-negative bacilli, *Acinetobacter* CBC- *Acinetobacter calcoaceticus baumannii* complex, GNB- Gram-negative bacilli