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Original Article

Comparative Evaluation of Antimicrobial Susceptibility Patterns in Klebsiella spp. and Proteus spp. Isolates from Urine Cultures in a Tertiary Care Hospital, Lahore

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ABSTRACT

Background: Urinary tract infections caused by antimicrobial-resistant Gram-negative organisms are a growing challenge for empirical therapy, particularly in tertiary care settings. Local susceptibility data are essential for guiding antibiotic selection and supporting antimicrobial stewardship. **Objective:** To compare the antimicrobial susceptibility patterns of Klebsiella spp. and Proteus spp. isolated from urine cultures in a tertiary care hospital in Lahore, Pakistan. **Methods:** This observational, cross-sectional, laboratory-based study was conducted in the Pathology Department of Ameer Ud Din Medical College/Post Graduate Medical Institute, Lahore, from January 2024 to December 2024. A total of 8,745 midstream clean-catch urine specimens were processed using standard microbiological culture methods. Significant bacterial isolates were identified by conventional biochemical testing, and antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method. Susceptibility patterns of Klebsiella spp. and Proteus spp. were summarized as frequencies and percentages. **Results:** Of 8,745 urine specimens, 3,326 (38.0%) showed significant bacterial growth. Among culture-positive samples, Klebsiella spp. were isolated in 312 (9.4%) cases and Proteus spp. in 48 (1.4%) cases. Proteus spp. showed complete susceptibility to imipenem and meropenem (100% each), while Klebsiella spp. showed lower susceptibility to imipenem (73%) and meropenem (72%). Ceftazidime-avibactam susceptibility was 81% in Proteus spp. and 64% in Klebsiella spp. Piperacillin/tazobactam susceptibility was higher in Proteus spp. (79%) than Klebsiella spp. (46%). Fluoroquinolone susceptibility was reduced in both organisms, with ciprofloxacin and levofloxacin susceptibility of 42% in Klebsiella spp. and 46% in Proteus spp. **Conclusion:** Proteus spp. retained high susceptibility to carbapenems, whereas Klebsiella spp. showed comparatively reduced susceptibility across several antibiotic classes. The low fluoroquinolone susceptibility in both organisms highlights the need for routine culture-based testing, updated local antibiograms, and susceptibility-guided therapy for urinary tract infections. **Keywords:** Antimicrobial resistance; Urinary tract infection; Klebsiella spp.; Proteus spp.; Antibiogram; Kirby–Bauer disk diffusion.

INTRODUCTION

Urinary tract infections (UTIs) remain among the most frequently encountered bacterial infections in both community and hospital settings, and their management has become increasingly complicated by the global rise of antimicrobial resistance. Gram-negative uropathogens are of particular concern because they often acquire resistance to commonly used empirical agents, leading to delayed effective therapy, increased healthcare costs, prolonged illness, and greater reliance on broader-spectrum or last-line antibiotics. In tertiary care hospitals, where prior antibiotic exposure, recurrent infections, catheter

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use, and comorbidities are frequently encountered, local antimicrobial susceptibility data are essential for guiding rational treatment decisions and supporting antimicrobial stewardship (1,2).

Among Gram-negative organisms causing UTIs, Klebsiella spp. and Proteus spp. are clinically important because they are associated with complicated infections and variable antimicrobial susceptibility profiles. Klebsiella species, particularly in hospital environments, have been increasingly linked with reduced susceptibility to several conventional beta-lactam antibiotics and other commonly used agents. Proteus species, especially Proteus mirabilis, are also important uropathogens in complicated urinary tract infections, particularly among patients with long-term catheterization, structural or functional urinary tract abnormalities, and renal calculi. Their urease production promotes alkaline urine, crystalline biofilm formation, stone development, catheter encrustation, and bacterial persistence, which makes them clinically relevant in recurrent and catheter-associated infections (3,9–12).

Empirical treatment of UTIs depends heavily on knowledge of local pathogen distribution and antibiotic susceptibility patterns. However, resistance profiles vary substantially between countries, regions, hospitals, and even between organisms isolated from the same clinical specimen type. Fluoroquinolones, cephalosporins, beta-lactam/beta-lactamase inhibitor combinations, aminoglycosides, carbapenems, fosfomycin, nitrofurantoin, and other reported agents may show differing in-vitro activity against Klebsiella and Proteus isolates. Therefore, organism-specific antibiogram data are necessary for more appropriate clinical decision-making (4,5).

In Pakistan, antimicrobial misuse and empirical prescribing remain important contributors to resistance, making updated local surveillance data from tertiary care laboratories important for clinical practice. Although UTIs are common, comparative local data on susceptibility patterns of Klebsiella spp. and Proteus spp. from urine cultures remain useful for selecting appropriate empirical therapy and preserving the effectiveness of higher-tier antimicrobial agents. Therefore, this study aimed to compare the antimicrobial susceptibility patterns of Klebsiella spp. and Proteus spp. isolated from urine cultures processed at a tertiary care hospital in Lahore, Pakistan, and to identify organism-specific susceptibility trends relevant to empirical treatment decisions and local antibiogram development.

MATERIALS AND METHODS

This observational, cross-sectional, laboratory-based study was conducted in the Pathology Department of Ameer Ud Din Medical College/Post Graduate Medical Institute, Lahore, Pakistan, over a one-year period from January 2024 to December 2024. The study included urine specimens submitted for routine microbiological culture and antimicrobial susceptibility testing during the study period. A total of 8,745 midstream clean-catch urine specimens were processed, of which 3,326 showed significant bacterial growth. Among the culture-positive specimens, 312 Klebsiella spp. and 48 Proteus spp. isolates were included for comparative antimicrobial susceptibility analysis.

Midstream clean-catch urine samples were collected aseptically in sterile, leak-proof containers. Approximately 10–20 mL of urine was obtained from each patient and transported to the laboratory for processing. Samples were processed immediately where possible; when delay was unavoidable, specimens were stored at 4°C for a limited duration before culture. Before inoculation, each urine specimen was mixed thoroughly to ensure homogeneity. For microbiological culture, urine samples were inoculated onto Cysteine-Lactose-Electrolyte-Deficient agar using a calibrated loop and the semi-quantitative streaking technique. Culture plates were incubated aerobically at 37°C for 18–24 hours. Significant bacterial growth was assessed according to routine laboratory criteria, and preliminary identification of bacterial isolates was performed using colony morphology, Gram staining, and catalase testing. Further identification and characterization of isolates were carried out using standard biochemical tests, including oxidase, citrate utilization, urease activity, methyl red, and Triple Sugar Iron tests. Isolates identified as Klebsiella spp. and Proteus spp. were selected for comparative analysis.

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method on Mueller–Hinton agar. A standardized bacterial inoculum equivalent to 0.5 McFarland turbidity standard was prepared and spread evenly across the agar surface to produce a uniform lawn of growth. After the inoculated plates were allowed to dry for approximately 3–5 minutes, antibiotic disks were applied aseptically at appropriate distances and gently pressed to ensure complete contact with the agar surface. Antibiotic disks were stored at 4°C in sealed containers and brought to room temperature before use to maintain disk potency. Following incubation, inhibition zones were measured and interpreted according to CLSI/EUCAST laboratory guidelines used by the institutional microbiology laboratory. Quality-control procedures were followed during culture and susceptibility testing; where available, standard control strains were used to verify media performance, disk potency, and zone-size interpretation.

The antibiotic panel included agents reported in the laboratory susceptibility records, including aminoglycosides, beta-lactam/beta-lactamase inhibitor combinations, carbapenems, cephalosporins, fluoroquinolones, fosfomycin, nitrofurantoin, tetracycline-class agents, tigecycline, and colistin. However, tigecycline, minocycline, and colistin were interpreted cautiously because these agents are not routinely preferred urinary antimicrobial options and require guideline-based interpretive validity.

The primary study variables were organism type and antimicrobial susceptibility pattern. The main outcome was the proportion of Klebsiella spp. and Proteus spp. isolates reported as susceptible to each tested antibiotic. Additional descriptive variables included total urine samples processed, culture-positive and culture-negative results, sex distribution of submitted samples, and frequency of the two target organisms among culture-positive samples. Culture positivity was calculated using the total number of urine samples as the denominator, whereas organism frequency was calculated among culture-positive samples.

Data were entered and analyzed using SPSS software. Categorical variables were summarized as frequencies and percentages. Antimicrobial susceptibility patterns of Klebsiella spp. and Proteus spp. were compared descriptively across antibiotic classes. No inferential statistical testing was retained because the available dataset was summarized as aggregate susceptibility percentages rather than isolate-level susceptibility counts for each antibiotic. Ethical approval for the study was obtained from the relevant institutional ethics committee before data collection. All urine specimens were processed as part of routine diagnostic microbiology services, and patient confidentiality was maintained throughout the study by using anonymized laboratory data. Duplicate isolates, contaminated specimens, and samples with incomplete laboratory records were excluded from the final analysis.

RESULTS

A total of 8,745 urine specimens were processed during the study period. Of these, 3,326 specimens showed significant bacterial growth, giving an overall culture positivity rate of 38.0%, while 5,419 specimens were culture-negative. Among all submitted specimens, 6,123 were from female patients and 2,622 were from male patients. Because culture positivity was not stratified by sex, these figures represent the sex distribution of submitted samples only and should not be interpreted as sex-specific UTI prevalence.

Table 1. Culture Results and Sex Distribution of Submitted Urine Specimens

Variable	Category	n	%
Total urine specimens processed	—	8,745	100.0
Culture result	Culture-positive	3,326	38.0
Culture result	Culture-negative	5,419	62.0
Sex distribution of submitted specimens	Female	6,123	70.0
Sex distribution of submitted specimens	Male	2,622	30.0

Among the 3,326 culture-positive samples, Klebsiella spp. were isolated in 312 cases, representing 9.4% of culture-positive samples. Proteus spp. were isolated in 48 cases, representing 1.4% of culture-positive

samples. Together, Klebsiella spp. and Proteus spp. accounted for 360 isolates, or 10.8% of all culture-positive urine samples.

Table 2. Frequency of Klebsiella spp. and Proteus spp. Among Culture-Positive Urine Samples

Organism	Number of isolates	Percentage among culture-positive samples, N = 3,326
Klebsiella spp.	312	9.4
Proteus spp.	48	1.4
Combined Klebsiella spp. and Proteus spp. isolates	360	10.8
Other culture-positive organisms	2,966	89.2

Comparative antimicrobial susceptibility patterns showed organism-specific variation across several antibiotic classes. Among aminoglycosides, Klebsiella spp. showed susceptibility rates of 64% to amikacin, 65% to gentamicin, and 51% to tobramycin, while Proteus spp. showed susceptibility rates of 73%, 58%, and 44%, respectively. Piperacillin/tazobactam showed higher reported susceptibility among Proteus spp. than Klebsiella spp., with susceptibility rates of 79% and 46%, respectively.

Carbapenems showed the highest reported activity against Proteus spp., with 100% susceptibility to both imipenem and meropenem. In comparison, Klebsiella spp. showed susceptibility rates of 73% to imipenem and 72% to meropenem. Ceftazidime-avibactam also showed higher susceptibility among Proteus spp. than Klebsiella spp., with rates of 81% and 64%, respectively. Among cephalosporins, Proteus spp. showed higher susceptibility to cefepime and ceftriaxone than Klebsiella spp. However, ceftriaxone susceptibility of 0% in Klebsiella spp. and cefotaxime susceptibility of 1% in Proteus spp. should be verified against raw laboratory records because these values appear unusual and may substantially affect interpretation. Fluoroquinolone susceptibility was reduced in both organisms, with ciprofloxacin and levofloxacin susceptibility of 42% among Klebsiella spp. and 46% among Proteus spp.

Table 3. Comparative Antimicrobial Susceptibility Patterns of Klebsiella spp. and Proteus spp. Urine Isolates

Antibiotic	Klebsiella spp. susceptible, %	Proteus spp. susceptible, %
Amikacin	64	73
Gentamicin	65	58
Tobramycin	51	44
Amoxicillin/clavulanic acid	23	19
Ampicillin	1	11
Piperacillin/tazobactam	46	79
Co-trimoxazole	43	29
Imipenem	73	100
Meropenem	72	100
Cefepime	38	67
Cefixime	31	40
Cefotaxime	32	1*
Ceftazidime-avibactam	64	81
Ceftriaxone	0*	54
Cefuroxime	NA	NA
Ciprofloxacin	42	46
Levofloxacin	42	46
Fosfomycin	37	13
Nitrofurantoin	28	3
Doxycycline	42	3
Minocycline	3†	NA
Tigecycline	3†	NA
Colistin	22†	NA

*The ceftriaxone and cefotaxime findings were retained as reported in the laboratory dataset and should be interpreted in the context of local testing practices and organism-specific susceptibility reporting.

†Minocycline, tigecycline, and colistin were included as reported in the laboratory susceptibility panel; their clinical interpretation for urinary isolates should be guided by institutional protocols and applicable antimicrobial susceptibility testing standards.

The highest reported susceptibility among Klebsiella spp. was observed for imipenem, meropenem, gentamicin, ceftazidime-avibactam, and amikacin. Among Proteus spp., the highest susceptibility was observed for imipenem, meropenem, ceftazidime-avibactam, piperacillin/tazobactam, and amikacin.

Table 4. Antibiotics Showing Highest Reported Susceptibility by Organism

Organism	Antibiotic	Reported susceptibility, %
Klebsiella spp.	Imipenem	73
Klebsiella spp.	Meropenem	72
Klebsiella spp.	Gentamicin	65
Klebsiella spp.	Ceftazidime-avibactam	64
Klebsiella spp.	Amikacin	64
Proteus spp.	Imipenem	100
Proteus spp.	Meropenem	100
Proteus spp.	Ceftazidime-avibactam	81
Proteus spp.	Piperacillin/tazobactam	79
Proteus spp.	Amikacin	73

The lowest reported susceptibility among Klebsiella spp. was observed for ceftriaxone, ampicillin, minocycline, tigecycline, and amoxicillin/clavulanic acid. Among Proteus spp., the lowest reported susceptibility was observed for cefotaxime, nitrofurantoin, doxycycline, ampicillin, and fosfomycin. The low nitrofurantoin susceptibility in Proteus spp. should be interpreted cautiously because Proteus spp. are generally considered poor targets for nitrofurantoin therapy.

Table 5. Antibiotics Showing Lowest Reported Susceptibility by Organism

Organism	Antibiotic	Reported susceptibility, %
Klebsiella spp.	Ceftriaxone*	0
Klebsiella spp.	Ampicillin	1
Klebsiella spp.	Minocycline†	3
Klebsiella spp.	Tigecycline†	3
Klebsiella spp.	Amoxicillin/clavulanic acid	23
Proteus spp.	Cefotaxime*	1
Proteus spp.	Nitrofurantoin	3
Proteus spp.	Doxycycline	3
Proteus spp.	Ampicillin	11
Proteus spp.	Fosfomycin	13

*Reported according to the available laboratory susceptibility dataset. †Included as part of the laboratory susceptibility panel; clinical interpretation for urinary isolates should follow institutional protocols and applicable susceptibility-testing standards.

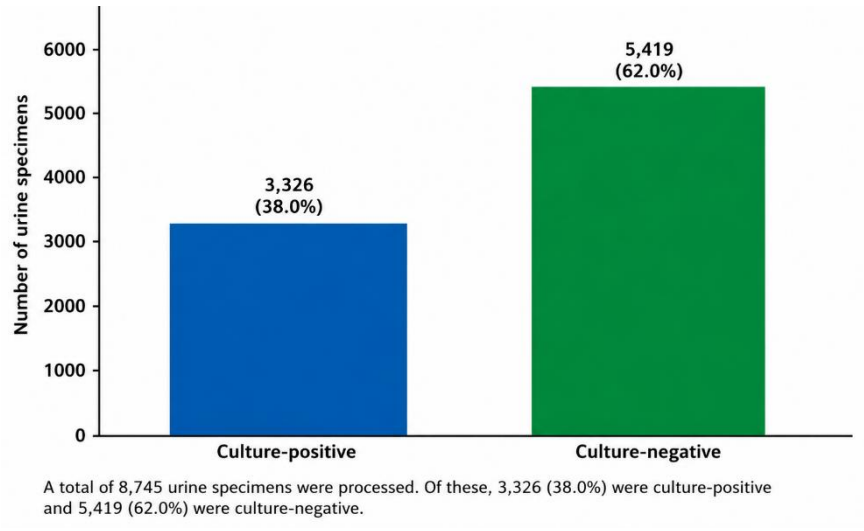

Figure 1. Distribution of Urine Culture Results Among Submitted Specimens

Figure 1 shows that, among 8,745 urine specimens processed during the study period, 3,326 (38.0%) yielded significant bacterial growth, while 5,419 (62.0%) were culture-negative. This indicates that

slightly more than one-third of submitted urine samples were culture-positive in this tertiary care laboratory setting. The finding supports the relevance of routine urine culture for identifying clinically significant bacterial growth and generating local antimicrobial susceptibility data. However, because the figure presents overall culture results only, it does not identify organism-specific burden, patient-level risk factors, or sex-specific culture positivity.

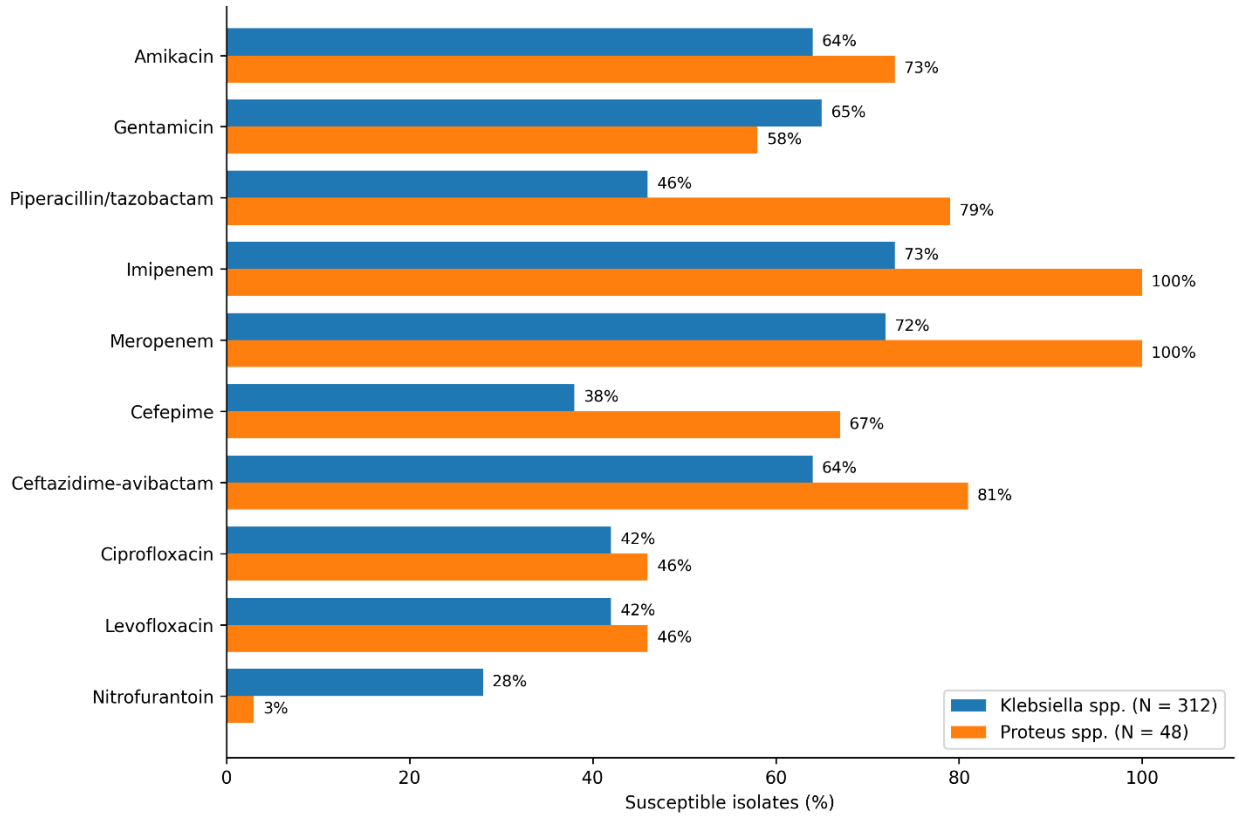

Figure 2. Comparative Antimicrobial Susceptibility Patterns of Klebsiella spp. and Proteus spp. Urine Isolates

Figure 2 demonstrates clear organism-specific differences in antimicrobial susceptibility between Klebsiella spp. and Proteus spp. urine isolates. Proteus spp. showed the highest susceptibility to carbapenems, with 100% susceptibility to both imipenem and meropenem, compared with 73% and 72%, respectively, among Klebsiella spp. Ceftazidime-avibactam and piperacillin/tazobactam also showed higher activity against Proteus spp. than Klebsiella spp., whereas fluoroquinolone susceptibility was reduced in both organisms, with ciprofloxacin and levofloxacin susceptibility of 42% in Klebsiella spp. and 46% in Proteus spp. Overall, the figure supports the study's main finding that Klebsiella spp. exhibited a less favorable susceptibility profile than Proteus spp., reinforcing the need for organism-specific susceptibility testing and local antibiogram-guided empirical therapy.

DISCUSSION

The present study compared the antimicrobial susceptibility patterns of Klebsiella spp. and Proteus spp. isolated from urine cultures processed in a tertiary care hospital laboratory in Lahore, Pakistan. Among 8,745 urine specimens processed during the study period, 3,326 (38.0%) showed significant bacterial growth. Of the culture-positive samples, Klebsiella spp. accounted for 312 (9.4%) isolates and Proteus spp. for 48 (1.4%) isolates. The principal finding of the study was that Proteus spp. demonstrated a comparatively more favorable susceptibility profile than Klebsiella spp., particularly against carbapenems and selected beta-lactam/beta-lactamase inhibitor combinations. In contrast, Klebsiella spp. showed reduced susceptibility across several commonly used antimicrobial classes, highlighting its importance as a resistant uropathogen in this hospital setting.

The overall culture positivity rate observed in this study indicates a substantial diagnostic yield among submitted urine specimens. However, these findings should be interpreted as laboratory culture positivity rather than population-level prevalence because the study was based on specimens submitted for microbiological testing rather than a defined community cohort. Although a greater proportion of submitted specimens originated from female patients, sex-specific infection rates could not be determined because culture positivity was not stratified according to sex (9,10).

Klebsiella spp. demonstrated only moderate susceptibility to carbapenems, with susceptibility rates of 73% for imipenem and 72% for meropenem. These findings are clinically important because carbapenems are commonly reserved for severe infections caused by resistant Gram-negative organisms. Reduced susceptibility among Klebsiella isolates may reflect increasing antimicrobial selection pressure in tertiary care settings and emphasizes the importance of antimicrobial stewardship and susceptibility-guided therapy (11-13). In contrast, Proteus spp. demonstrated complete susceptibility to both imipenem and meropenem in the present dataset. Although this finding suggests preserved in-vitro activity of carbapenems against Proteus isolates in this setting, interpretation should be cautious because the number of Proteus isolates was relatively small compared with Klebsiella isolates. The findings related to beta-lactam and beta-lactam/beta-lactamase inhibitor agents further demonstrated organism-specific variability. Proteus spp. showed higher susceptibility to piperacillin/tazobactam and ceftazidime-avibactam than Klebsiella spp., suggesting comparatively better retained activity of these agents against Proteus isolates in the current study setting. These findings support the continued usefulness of selected inhibitor-protected beta-lactam agents when guided by susceptibility testing.

Reduced susceptibility to cephalosporins was observed in both organisms. Klebsiella spp. demonstrated low susceptibility to cefepime and cefotaxime, while ceftriaxone susceptibility was reported as 0% in the available laboratory data. Proteus spp. showed higher susceptibility to cefepime and ceftriaxone but markedly low susceptibility to cefotaxime. Because cefotaxime and ceftriaxone are both third-generation cephalosporins, the observed discrepancy should be interpreted cautiously and verified against original laboratory antimicrobial susceptibility records before definitive conclusions are drawn. Nevertheless, the overall findings suggest reduced reliability of several conventional cephalosporins for empirical treatment of urinary tract infections caused by these organisms in the present setting.

Fluoroquinolone susceptibility was reduced in both organisms, with susceptibility rates of 42% among Klebsiella spp. and 46% among Proteus spp. for both ciprofloxacin and levofloxacin. This finding is clinically important because fluoroquinolones remain commonly prescribed oral agents for urinary tract infections. The reduced susceptibility observed in the present study suggests that empirical fluoroquinolone therapy may be unreliable in this hospital setting unless supported by current local antibiogram data or isolate-specific susceptibility testing.

Lower susceptibility was also observed for several commonly used antimicrobial agents. Klebsiella spp. demonstrated very low susceptibility to ampicillin and limited susceptibility to amoxicillin/clavulanic acid, nitrofurantoin, and fosfomycin. Proteus spp. also demonstrated low susceptibility to ampicillin, fosfomycin, nitrofurantoin, and doxycycline. The low reported susceptibility of Proteus spp. to nitrofurantoin should be interpreted cautiously because Proteus species are generally considered intrinsically less susceptible to nitrofurantoin and are not ideal targets for nitrofurantoin therapy (14,15).

The present findings are broadly consistent with previous studies reporting increasing antimicrobial resistance among Gram-negative uropathogens. Studies by Caneiras et al., Bobbadi et al., Vaez et al., and Cohen-Nahum et al. similarly reported variable susceptibility profiles among Klebsiella and Proteus isolates, particularly with reduced activity of fluoroquinolones and cephalosporins (15-18). However, direct comparisons should be interpreted cautiously because antimicrobial panels, study populations, laboratory methodologies, and local resistance patterns differ between institutions and regions.

From a clinical perspective, the present study highlights the importance of routine culture and antimicrobial susceptibility testing for urinary tract infections in tertiary care settings where resistant Gram-negative organisms are frequently encountered. The findings support regular updating of institutional antibiograms and careful selection of empirical therapy based on local susceptibility trends rather than generalized assumptions regarding organism susceptibility.

This study has several limitations. First, it was conducted in a single tertiary care hospital laboratory, which may limit generalizability to other healthcare settings or geographic regions. Second, the sample size of Proteus spp. isolates was relatively small compared with Klebsiella spp. isolates, which may affect comparative interpretation. Third, patient-level clinical data, including prior antibiotic exposure, inpatient or outpatient status, catheterization history, and comorbidities, were not available for analysis. Finally, molecular characterization of resistance mechanisms such as ESBL or carbapenemase production was not performed. Despite these limitations, the study provides useful local laboratory evidence regarding the comparative antimicrobial susceptibility patterns of Klebsiella spp. and Proteus spp. isolated from urine cultures in a tertiary care hospital setting. The findings reinforce the practical importance of local microbiology surveillance, organism-specific antibiogram reporting, and antimicrobial stewardship programs in guiding empirical therapy and preserving the effectiveness of remaining therapeutic options.

CONCLUSION

This laboratory-based observational study demonstrated distinct antimicrobial susceptibility patterns among Klebsiella spp. and Proteus spp. isolated from urine cultures in a tertiary care hospital setting in Lahore, Pakistan. Proteus spp. retained high susceptibility to carbapenems, whereas Klebsiella spp. showed comparatively reduced susceptibility across several commonly used antimicrobial classes. Reduced susceptibility to fluoroquinolones and multiple beta-lactam agents in both organisms highlights the growing challenge of antimicrobial resistance in urinary tract infections. These findings support the routine use of culture-based antimicrobial susceptibility testing, regular updating of local antibiograms, and susceptibility-guided therapy to improve empirical treatment decisions and strengthen antimicrobial stewardship practices.

RECOMMENDATIONS AND LIMITATIONS

Empirical therapy for urinary tract infections should be supported by regularly updated local antibiogram data and culture-based susceptibility testing wherever feasible. Rational antimicrobial prescribing and strengthened antimicrobial stewardship programs are important to reduce unnecessary use of broad-spectrum antibiotics and preserve the effectiveness of higher-tier agents such as carbapenems and newer beta-lactam/beta-lactamase inhibitor combinations. Continued microbiological surveillance and periodic reassessment of local resistance trends are also recommended.

The present study was limited by its single-center design and unequal isolate distribution between Klebsiella spp. and Proteus spp. The study also lacked patient-level clinical information, including prior antibiotic exposure, hospitalization status, catheterization history, and associated comorbidities. In addition, molecular characterization of resistance mechanisms such as ESBL or carbapenemase production was not performed. Some reported cephalosporin susceptibility findings should also be verified against original laboratory antimicrobial susceptibility records before definitive interpretation.

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