ORIGINAL RESEARCH PAPER

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ANTIMICROBIAL RESISTANCE SURVEILLANCE OF UROPATHOGENIC ESCHERICHIA COLI: A ONE-YEAR STUDY IN A TERTIARY CARE HOSPITAL, BANGLADESH

KEY WORDS: Urinary Tract Infection (UTI), E. coli (UPEC), Multidrug-Resistant Bacteria (MDR), surveillance, Bangladesh

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ABSTRACT

Background: Urinary tract infections (UTIs) are among the most common bacterial diseases globally, with Escherichia coli recognized as the predominant etiological agent. The increasing prevalence of antimicrobial resistance in uropathogenic E. coli (UPEC) presents a major clinical and public health concern, particularly in developing countries such as Bangladesh. **Methods:** A prospective cross-sectional surveillance study was conducted from July 2023 to June 2024 at the Department of Microbiology and Immunology, Bangabandhu Sheikh Mujib Medical University, Bangladesh. A total of 17,139 midstream urine specimens from patients with clinically suspected UTIs were cultured and analyzed. Significant bacteriuria was defined as $\geq 10^5$ CFU/mL. Isolates were identified through standard microbiological and biochemical methods, and antimicrobial susceptibility was determined using the Kirby-Bauer disk diffusion method according to CLSI guidelines. **Results:** Of the 17,139 specimens, 5,266 (30.73%) yielded significant bacterial growth, with E. coli isolated from 2,842 cases (69%). Female patients constituted 62.45% of positive cases, with the highest prevalence among the 21–40-year age group (33.21%). Resistance was markedly high against Nalidixic Acid (91.31%), Cefotaxime (83.21%), and Ceftazidime (81.92%), as well as Co-trimoxazole (59.32%) and fluoroquinolones. In contrast, aminoglycosides, particularly Amikacin (90.58%) and Netilmicin (81.45%), along with Nitrofurantoin (69.49%), demonstrated the highest sensitivity. **Conclusion:** This study highlights the predominance of E. coli in UTIs and the alarming resistance to commonly used antibiotics. Amikacin, Netilmicin, and Nitrofurantoin remain viable therapeutic options. Regular surveillance and stringent antimicrobial stewardship are essential to mitigate the spread of multidrug-resistant UPEC strains.

INTRODUCTION

Urinary tract infections (UTIs) represent one of the most common infectious diseases encountered in clinical practice worldwide, affecting individuals across all stages of life. These infections involve either the lower urinary tract (cystitis and urethritis) or the upper urinary tract (pyelonephritis and ureteritis) (1,2). Escherichia coli, a Gram-negative, facultatively anaerobic, non-spore-forming bacillus belonging to the family Enterobacteriaceae, is the predominant etiological agent of UTIs. While commensal strains of E. coli colonize the human gastrointestinal tract as part of the normal microbiota, uropathogenic variants possess virulence determinants enabling invasion, colonization, and persistence within the urinary tract. In addition to UTIs, pathogenic E. coli strains are implicated in gastrointestinal infections, sepsis, intra-abdominal abscesses, and other systemic illnesses (3).

The global burden of UTIs is substantial owing to their high incidence, recurrence, and the risk of complications, particularly in the context of rising antimicrobial resistance. Epidemiological studies indicate that E. coli accounts for over 80% of community-acquired UTIs worldwide (4). The prevalence is disproportionately higher among women due to anatomical and physiological predispositions, including a shorter urethra and its proximity to the perianal region, which facilitates microbial ascension into the bladder (4,5,6). Complicated UTIs frequently occur in patients with structural or functional abnormalities of the urinary tract, indwelling catheters, or immunocompromised conditions (7).

The current diagnostic gold standard remains urine culture coupled with antimicrobial susceptibility testing (8). However, therapeutic management is increasingly constrained by the emergence of antibiotic resistance, particularly in regions where indiscriminate prescription practices, over-the-counter availability, and insufficient antimicrobial stewardship prevail (9). Over the last two decades, clinical E. coli isolates have demonstrated escalating resistance to conventional first-line agents, culminating in the emergence of multidrug-resistant (MDR) strains—defined as those resistant to three or more antimicrobial classes (10,11). Infections caused by MDR E. coli are associated with limited therapeutic options, increased treatment costs, prolonged hospital stays, and elevated morbidity and mortality, especially in resource-constrained healthcare systems such as those in South Asia (12,13).

Objectives of the Study

This Study Aimed to:

1. Detect the presence of E. coli in clinical specimens obtained from patients with suspected UTIs.
2. Assess the prevalence of E. coli infections across different age groups.
3. Characterize the antimicrobial resistance patterns of the isolated strains.

MATERIALS AND METHODS

Sample Collection

This cross-sectional study was conducted at the Department of Microbiology and Immunology, Bangabandhu Sheikh Mujib Medical University from July 2023 to June 2024. Patients visiting the OPD and IPD with any symptoms indicative of a UTI were included in the study. Freshly voided, clean-catch midstream urine samples were collected from each patient into sterile screw-capped universal containers. Specimens collected in unsterile containers, those that were delayed in transport for culture, not kept refrigerated at 4°C, and inadequate samples for urine culture were excluded from the study. A total of 17139 midstream urine specimens were collected aseptically in sterile, wide-mouth glass containers from both male and female patients across various age groups. Each specimen was labelled and transported to the microbiology laboratory for further processing and analysis.

Isolation of Escherichia Coli

Quantitative bacteriuria was assessed using a calibrated 0.01 mL wire loop to inoculate urine samples onto Blood Agar, which supports the growth of a broad range of bacterial species, including E. coli. The same procedure was followed for inoculation onto MacConkey Agar, selective for Gram-negative organisms, and Chromogenic Agar, which facilitates the identification of E. coli through its characteristic colony morphology. All culture plates were incubated aerobically at 37°C for 24 hours.

Post-incubation, colony counts were determined to evaluate bacteriuria. A threshold of $\geq 10^5$ colony-forming units (CFU)/mL was considered indicative of significant bacteriuria, suggestive of pathogenic E. coli, while counts $<10^5$ CFU/mL were classified as non-significant growth. Gram staining was performed on isolated colonies, and pure cultures were prepared for further confirmatory biochemical and morphological identification of E. coli.

Biochemical Identification

A series of standard biochemical tests were performed on the pure cultures to confirm the identity of E. coli. These included the Indole test, Methyl Red test, Citrate utilization test, Urease test, Catalase test, Oxidase test, Hydrogen Sulphide (H_2S) production, Gas production test, and Motility test.

Antimicrobial Susceptibility Testing

Antibiotic susceptibility of the E. coli isolates was determined using the Kirby-Bauer disk diffusion method following the guidelines of the Clinical and Laboratory Standards Institute (CLSI) (8). Commercially prepared antibiotic discs (Oxoid Ltd., England) were used to evaluate the antimicrobial profile of the isolates. The following antibiotics and their respective disc concentrations were tested: Cefotaxime (30 µg), Co-trimoxazole (23.75 µg sulfamethoxazole/ 1.25 µg trimethoprim), Levofloxacin (5 µg), Ceftazidime (30 µg), Ceftriaxone (30 µg), Ciprofloxacin (30 µg), Gentamycin (10 µg), Amikacin (30 µg), Piperacillin-Tazobactam (20/5 µg).

To prepare the bacterial inoculum, a loopful of pure E. coli colony was inoculated into 2 mL of Mueller-Hinton broth and incubated at 37°C for 4 hours to obtain a fresh culture. The turbidity of the culture was then adjusted to match the 0.5 McFarland standard. A sterile cotton swab was dipped into the standardized suspension and used to uniformly inoculate the surface of a Mueller-Hinton agar plate. Plates were allowed to dry at room temperature for a few minutes before the antibiotic discs were aseptically placed on the agar surface using sterile forceps. The plates were incubated aerobically at 37°C for 24 hours. After incubation, the diameters of the zones of inhibition were measured in millimetres using a vernier calliper to determine susceptibility patterns.

Quality Control

A reference strain of Escherichia coli (ATCC 25922) was used as a quality control organism to validate culture and susceptibility testing procedures. The results of the antibiotic susceptibility tests were interpreted according to the CLSI interpretive criteria, categorizing the isolates as sensitive or resistant based on the measured inhibition zone diameters (14,15).

RESULTS

A total of 17139 urine specimens from patients clinically suspected of urinary tract infection (UTI) were analyzed for the presence of Escherichia coli, followed by antimicrobial susceptibility testing. During the study period, 5266 (30.73%) of these samples demonstrating significant growth. Out of these, 2842 samples (69%) showed growth of E. coli, defined by colony counts $\geq 10^5$ CFU/mL. The presence of E. coli was further confirmed through standard microbiological and biochemical methods.

A total of 17,139 urine specimens from patients clinically suspected of urinary tract infection (UTI) were examined for the presence of Escherichia coli and subsequently subjected to antimicrobial susceptibility testing. Of these, 5,266 samples (30.73%) demonstrated significant bacterial growth. Among the culture-positive samples, 2,842 (69%) yielded E. coli, defined by a colony count of $\geq 10^5$ CFU/mL The identity of E. coli isolates was confirmed using standard microbiological and biochemical techniques.

Among the 2842 positive cases, 1775 (62.45%) were female and 1067 (37.55%) were male (Figure 1). The highest prevalence of E. coli-positive UTI was observed in the 21–40 years age group (33.21%), followed by 41–60 years (25.36%), >60 years (22.30%), and 0–20 years (19.10%) (Table 1).

The 2842 confirmed E. coli isolates were subjected to antimicrobial susceptibility testing using the Kirby-Bauer disk diffusion method. High resistance was observed against Nalidixic Acid (91.31%), Cefotaxime (83.21%), Ceftazidime (81.92%), Co-trimoxazole (59.32%), Ceftriaxone (57.55%), Ciprofloxacin (58.52%), and Levofloxacin (52.59%). Conversely, high sensitivity was seen to Amikacin (90.58%), Tobramycin (79.72%), Netilmicin (81.45%), Gentamycin (68.89%), and Nitrofurantoin (69.49%). (Table 2).

DISCUSSION

Escherichia coli is a ubiquitous microorganism, widely distributed in the environment and as part of the commensal flora of the human gastrointestinal tract, but it also represents a major opportunistic pathogen in both community and healthcare settings (9). It is universally recognized as the principal etiological agent of urinary tract infections (UTIs), being implicated in approximately 70–80% of global cases (16,17). In the present study, E. coli was isolated in 53.97% of positive urine cultures, underscoring its continued predominance as the leading uropathogen. The overall distribution of pathogens identified is presented in Table I, where E. coli constituted 56.8% of the isolates, which is consistent with the findings of previous investigations (18).

Sex-specific distribution revealed a greater prevalence of UTI cases among females (62.45%), with E. coli being isolated more frequently in women (68%) compared to men. This observation is concordant with the studies of Malik et al and Ramírez et al. (19,20). These include the relatively short female urethra, its close proximity to the anus, absence of antimicrobial factors such as prostatic secretions, and hormonal influences during reproductive age (7). The highest prevalence of E. coli-mediated UTIs was noted in females aged 21–30 years, followed by the 31–40 years group, reflecting the impact of sexual activity and reproductive physiology on susceptibility. Comparable findings have been reported in Pakistan (69.80%), Saudi Arabia (60.35%), Iran (86.24%), and Turkey (82.30%) (21,22,23,24).

Age-stratified analysis indicated that the 21–40 years group accounted for the highest incidence (33.21%), aligning with the findings of Tanzina et al. in Bangladesh [7]. Infections were also observed among 41–60 years (25.36%), >60 years (22.30%), and 0–20 years (19.10%) groups. Interestingly, the infection rate in the youngest cohort was higher than that reported by Tanzina et al. but remains in agreement with epidemiological trends described by Foxman, who demonstrated that nearly 50% of women will develop a UTI in their lifetime, with one-third requiring antimicrobial therapy by early adulthood (25).

Antimicrobial resistance (AMR) among uropathogens, particularly E. coli, represents a formidable public health challenge worldwide, with the burden being disproportionately higher in resource-limited settings due to injudicious antibiotic use, limited diagnostic infrastructure, and inadequate stewardship programs (26). Several studies from Bangladesh have reported alarmingly high resistance rates among urinary E. coli isolates (7,27). Consistent with these reports, the present investigation identified extensive resistance across multiple antibiotic classes.

Antimicrobial susceptibility testing, performed using the Kirby–Bauer disk diffusion method, revealed the highest resistance to Nalidixic Acid (91.31%). This parallels the findings of Setu et al. in Bangladesh (83.31%) , as well as reports from India (77.7%) and Saudi Arabia (50.6%) (22,27.28), though notably higher than data from Iraq (25.9%) (30). The pervasive resistance to Nalidixic Acid is likely attributable to its longstanding use as a first-generation quinolone in UTI management and possible zoonotic dissemination of resistant strains (7,31). In addition, markedly high resistance was observed against third-generation cephalosporins, including Cefotaxime (83.21%), Ceftazidime (81.92%), and Ceftriaxone (57.55%), as well as to Co-trimoxazole (59.32%), Ciprofloxacin (58.62%), and Levofloxacin (52.59%). These findings indicate a worrying escalation in cephalosporin resistance compared to earlier Bangladeshi studies that reported resistance rates below 40% (7). However, they are supported by more recent national data and studies from India, which reported resistance rates above 60% (27,32).

Global surveillance studies similarly document rising resistance to Co-trimoxazole and cephalosporins among urinary E. coli isolates (33,34). In regions such as Iraq, resistance to cephalosporins including Cefazolin and Cefixime has been particularly pronounced (30). The drivers of this resistance in Bangladesh and comparable developing countries include unrestricted access to antibiotics without prescription, inappropriate selection of antimicrobials, use in veterinary and agricultural settings, and dissemination of plasmid-mediated resistance genes (27).

Encouragingly, the present study demonstrated preserved sensitivity of urinary E. coli isolates to aminoglycosides and nitrofurans. More than 90% of isolates were susceptible to Amikacin, followed by Netilmicin (81.45%), Gentamicin (69.49%), and Nitrofurantoin (68.89%). These findings are concordant with earlier Bangladeshi studies as well as international reports from Saudi Arabia, India, Pakistan, and Iraq (3,17.22,30). Particularly, Amikacin remains highly effective, with sensitivity rates exceeding 95% in several regions. Nitrofurantoin and Gentamicin also continue to demonstrate robust efficacy, with studies from Turkey and India reporting sensitivity rates above 90% (34).

Collectively, these results suggest that aminoglycosides (Amikacin, Netilmicin, Gentamicin) and Nitrofurantoin remain viable therapeutic options for the management of UTIs caused by E. coli in Bangladesh. Nevertheless, empirical therapy should be guided by antimicrobial susceptibility testing to ensure optimal treatment outcomes and to mitigate the further propagation of multidrug-resistant E. coli strains.

CONCLUSION

The present study reaffirms that Escherichia coli constitutes the predominant etiological agent of urinary tract infections (UTIs), with a disproportionately higher prevalence among female patients compared to males. The antimicrobial susceptibility patterns highlight Amikacin and Netilmicin as the most efficacious therapeutic options against E. coli isolates, supporting their continued use as potential first-line agents. Nevertheless, reliance on empirical therapy should be approached with caution, as treatment decisions must be guided by routine antimicrobial susceptibility testing to ensure optimal clinical outcomes and to limit the emergence of resistant strains.

Despite these important findings, the study is not without limitations. The investigation was conducted at a single tertiary care centre, which may restrict the generalizability of results to wider populations. Additionally, only a limited panel of antibiotics was assessed, potentially underestimating the resistance spectrum against newer or less commonly used antimicrobials. The study also relied on conventional culture-based methods without incorporating molecular approaches that could have provided deeper insights into resistance mechanisms, virulence factors, and clonal relatedness of isolates. Furthermore, patient-level factors such as comorbidities, prior antibiotic exposure, or hospitalization history were not comprehensively evaluated, which may have influenced both pathogen distribution and resistance patterns.

To mitigate the escalating challenge of antimicrobial resistance, stringent antibiotic stewardship strategies, rational prescribing practices, and strengthened surveillance systems are urgently required. Future research incorporating multicentre data, broader antimicrobial panels, and molecular epidemiological tools is warranted to better characterize the evolving resistance landscape of uropathogens in Bangladesh and beyond.

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Declarations
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Ethical Approval: The study was approved by the Institutional Ethics Committee.

Table 1: Distribution of E. coli Positive Urine Samples by Age Group (n = 2842)

Age Group (years)	Number of Patients (n)	Percentage (%)
0–20	543	19.10
21–40	944	33.21
41–60	721	25.36
>60	634	22.33
Total		100.00

Table 2: Antimicrobial Susceptibility Pattern of E. coli Isolates (n = 138)

Antibiotic	Resistance (%)	Sensitivity (%)
Nalidixic Acid	91.31	6.69
Cefotaxime	83.21	16.79
Ceftazidime	81.92	18.08
Co-trimoxazole	59.32	40.68
Ceftriaxone	57.55	42.45
Ciprofloxacin	58.52	41.48
Levofloxacin	52.59	47.41
Amikacin	9.42	90.58
Netilmicin	18.55	81.45
Gentamycin	30.51	69.49
Nitrofurantoin	31.11	68.89

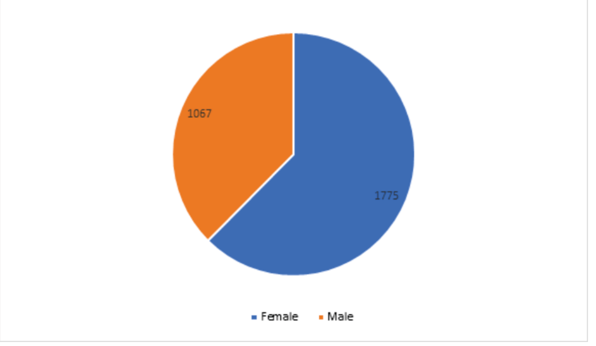

Figure 1: Distribution of E. coli Isolates of Urinary Samples Among Different Gender Groups (n=2842).

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