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Original Research Article

Antimicrobial sensitivity pattern of bacterial pathogens for uncomplicated urinary tract infection in female patients at a tertiary level hospital

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ABSTRACT

Background: Urinary tract infections (UTIs) remain the common infections in outpatients as well as hospitalized patients. Antimicrobials are frequently used drugs for the treatment of UTIs. Periodic evaluation of antimicrobial activity of different antimicrobial agents is essential as the pattern of antimicrobial sensitivity may vary over period. The aim of this study was to identify the antimicrobial sensitivity pattern of the isolated uropathogens in female patients in urinary tract infection at a tertiary care hospital in Bangladesh.
Methods: This observational cross-sectional type of study was conducted in the department of pharmacology and therapeutics in collaboration with department of microbiology SBMC, outpatient department of medicine, and gynaecology and obstetrics, SBMCH, Barishal, from January 2017 to December 2017.
Results: In this study, age of the subjects ranging from 18 to 65 years, majority subjects (57.0%) belonged to age group of 31-44 years. The mean age was found 44.5±9.1 years. Out of 200 cases, 83% cases hailing from rural area and 17% from urban site. In this study microbial culture result of uncomplicated UTI revealed that 103 (51.5%) of urine samples had significant bacteriuria. *E. coli* was found to be the most prevalent 47 (45.6%), followed by *Klebsiella pneumoniae* 18 (17.4%), *Proteus* spp. 11 (10.6%) and *Enterobacter* spp. 9 (8.7%).
Conclusions: The pattern of resistance to commonly used antimicrobials for treating UTI alerts us against indiscriminate usage of antimicrobials.

Keywords: Urinary tract infection, Bacterial pathogens, Antimicrobial sensitivity, *Escherichia coli, Enterobacter* spp.

INTRODUCTION

Urinary tract infections (UTIs) are one of the most common infectious diseases, and it is estimated that 150 million UTIs occur yearly on a global basis, resulting in more than 6 billion dollars are used as direct health care expenditures.[1] All individuals are susceptible to UTIs; but prevalence of infections differs with age, sex and some other predisposing factors. The incidence of UTIs greater in females than males with two exceptions, infants and catheter related infections.[2] Females are more infected due to the proximity of genital tract and urethra/anus and anatomical predisposition or urothelial mucosa adherence to mucopolysaccharides lining or other host factors.[3] In women, the ascent of organisms into the bladder is facilitated by the short urethra and absence of bactericidal prostatic secretions. Sexual intercourse causes minor urethral trauma and may transfer bacteria from the perineum into the bladder. The prevalence of UTI in women is about 3% at the age of 20 years, increasing by about 1% in each subsequent decade. UTI is an infection in any part of urinary system-kidneys, ureters, bladder and urethra. Most infections involve the lower urinary tract-the bladder and the urethra. It implies multiplication of

organisms in the urinary tract, and is defined by the presence of more than 100000 organisms per ml in a midstream sample of urine (MSU).[4] UTIs are classified according to site of infection: kidney (pyelonerhritis), bladder (cystitis), urethra (urethritis) or urine (bacteriuria). It can be classified as symptomatic or asymptomatic, on the other way it may be classified as uncomplicated and complicated UTIs. UTIs that occur in a normal genitourinary tract with no prior instrumentation and no concomitant disease that would promote the UTIs are considered as "uncomplicated" UTIs, whereas UTIs are diagnosed as complicated when the infections are associated with some functional, metabolic or structural abnormalities in genitourinary tract, e.g., renal cyst/calculi/abscess, certain forms of pyelonephritis, indwelling catheter, diabetes, AIDS, and H/O renal transplant. Both complicated and uncomplicated UTIs are frequently symptomatic.[5]

It is considered to be a lower UTI (AUC) when symptoms are restricted to the lower urinary tract, e.g., pain on micturition (dysuria), urgency, pollakisuria or pain above the symphysis and in case of an upper UTI (AUP), symptoms are restricted to the upper urinary tract, those include, flank pain, pain on percussion on the renal area, and/or fever (>38^{0}C).[6] The probability of infection is approximately 50% if women present with 1 or more symptoms of UTIs. Probability of UTIs raises to more than 90% in presence of specific combinations of symptoms, e.g., dysuria and frequency without vaginal discharge or irritation, effectively ruling in the diagnosis based on history alone. In contrast, history taking, physical examination and dipstick urinalysis are not able to reliably lower the posttest probability of disease to a level where a UTI can be ruled out when a patient present with 1 or more symptoms.[7]

Objectives

General objective of the study was to identify the antimicrobial sensitivity pattern of the isolated uropathogens in female patients in urinary tract infection at a tertiary care hospital in Bangladesh.

Specific objectives of the study were: to assess the antimicrobial sensitivity pattern, and to find out the causative organism for UTI.

METHODS

This was an observational cross sectional type of study conducted in the department of pharmacology and therapeutics in collaboration with department of microbiology and outpatient department of female medicine and gynaecology and obstetrics, Sher–E-Bangla Medical College Hospital, Barishal, Bangladesh from January 2017 to December 2017. A total of 200 women were selected who were attended in female medicine and gynaecology and obstetrics OPD, Sher-E-Bangla Medical College hospital, Barishal, with clinical features of UTIs.

Inclusion criteria

Patients with clinical features of UTI, female patients, age >18 year, and patients with informed consent were included in the study.

Exclusion criteria

Seriously ill patients who need ICU support, patients with features of UTI but without microbiological proof, pregnant women, women who received antibiotic within two months, complicated UTI, and patients who were not interested to participate in this study were excluded.

Data collection

This prospective study was conducted in department of pharmacology and therapeutics with collaboration with department of microbiology and outpatient department of female medicine, and gynaecology and obstetrics, Sher-E-Bangla Medical College Hospital, Barishal. Subjects were selected purposively as patients with clinical features of UTI. The demographic information includes age, residence, occupation, socioeconomic status, marital status of the woman. Data were collected for the analysis. Samples with incomplete data, women who received antibiotics recently were excluded from the study. After fulfilling the inclusion and exclusion criteria, patient was enrolled with unique ID. Subjects were briefed about the objectives of the study, risk and benefits, freedom for participating in the study and confidentiality.

Operational definition

UTI defined as multiplication of organisms in the urinary tract. It is usually associated with the presence of WBC and ≥100,000 microorganism/ml in a midstream sample of urine (MSU).

Laboratory procedure

Urine samples were collected using a clean catch mid-stream method and processed within 1 hour. Samples were inoculated on MacConkey agar and blood agar, then incubated at 37°C for 24-48 hours. UTI was diagnosed if a single organism was cultured at a concentration of 10^5 CFU/ml and 5 leukocytes per high-power field observed.[8] Bacterial identification relied on standard culture and biochemical characteristics. *E. coli, K. pneumoniae, P. mirabilis, Pseudomonas, Staphylococcus aureus,* and *Staphylococcus saprophyticus* were identified based on specific tests.

Antimicrobial susceptibility test

Antimicrobial susceptibility tests were conducted on Mueller-Hinton agar using the disk diffusion method. Standard inoculums were incubated at 37°C for 24 hours, and susceptibility/resistance was determined based on isolate growth zone diameter. Inhibition zones greater than

10 mm were considered sensitive, 5-10 mm moderately sensitive, and no inhibition zone as resistant. Antimicrobials tested included amikacin, gentamicin, ciprofloxacin, nitrofurantoin, cefixime, imipenem, and co-trimoxazole. *E. coli* ATCC 25922, *S. aureus* ATCC 25923, and *Pseudomonas aeruginosa* ATCC-27853 were used as reference strains for quality control.

Data analysis

Statistical analysis was performed using the statistical package for social sciences (SPSS, Chicago, IL) version 22.0 software for Windows. Descriptive statistics was performed, and all data was expressed as mean±SD and percentage ratio. The quantitative observations were analyzed by frequencies and percentages.

Quality assurance strategy

It is extremely important that the data collection procedure, method becomes good in quality, for that purpose at first work manual was made. Then a sample size was selected. A standard questionnaire was made.

Ethical issues

There is minimum physical, psychological, social and legal risk during history taking, physical examination and investigation. Proper safety measures were ensured in every steps of the study. Ethical clearance was obtained from the ethical committee of Sher-E-Bangla Medical College, Barishal. According to Helsinki Declaration for Medical Research involving Human Subjects 1964, all the patients were informed about the study design, the right for the study subjects to withdraw themselves from the study at any time for any reason. Informed written consent was obtained from each subject who was voluntarily provide consent to participate in the study.

RESULTS

Table 1 analyzes the socio-demographic characteristics of our study patients. The majority of study subjects fall within the age range of 31-44 years (57%), followed by 15-30 years (9%), 45-60 years (26%), and 60-75 years (8%). The mean age of the study subjects is 42.6 years, with a standard deviation of 11.4 years. The age range spans from 18 to 74 years. The majority of study subjects are married (73%), while the remaining is single (27%). The most common occupation among study subjects is housewife (41.5%), followed by service holder (20%), daily worker (22.5%), business (9.5%), and others (6.5%). The majority of study subjects belong to the poor class (46.5%), followed by middle class (38.0%) and upper class (15.5%). Most study subjects reside in rural areas (83%), while a smaller proportion resides in urban areas (17%) respectively.

Figure 1 shows the result of microbial culture of study subjects. Cultures with colony count ≥105 cfu/ml was considered as significant bacteriuria. Culture results shows culture positive cases in 51.5% of urine samples.

Table 1: Socio demographic characteristics of the study subjects (n=200).

Characteristics	Frequency	Percentage
Age		
15-30	18	9
31-44	114	57
45-60	52	26
60-75	16	8
Mean±SD	42.6±11.4	
Range	18-74	
Marital status		
Single	54	27
Married	146	73
Occupation		
Service holder	40	20
House wife	83	41.5
Daily worker	45	22.5
Others	13	6.5
Business	19	9.5
Socioeconomic condition		
Poor class	93	46.5
Middle class	76	38.0
Upper class	31	15.5
Residence		
Urban	34	17
Rural	166	83.0

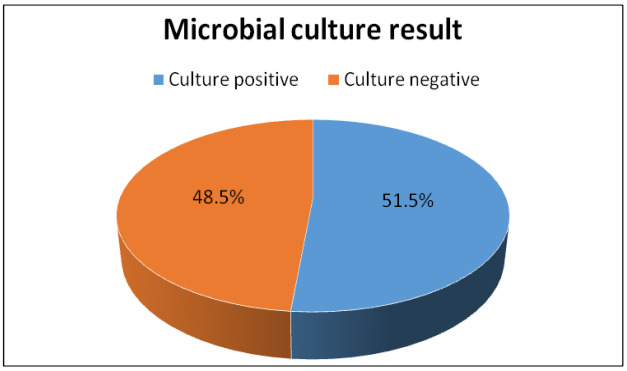

Figure 1: Microbial culture result of uncomplicated UTI patients (n=200).

Table 2 and Figure 2 shows the percentage prevalence of isolated microorganisms in uncomplicated UTIs. *E. coli* was found to be the most prevalent 47 (45.6%), followed by *Klebsiella pneumoniae* 18 (17.4%), *Proteus* sp. 11 (10.6%) and *Enterobacter* sp. 9(8.7%). The least prevalent organisms were *Staphylococcus aureus, Pseudomonas* and *Streptrococcus saprophyticus.*

Table 3 shows the antimicrobials sensitivity and resistance pattern those are used in UTI patients. Nitrofurantoin was found lowest resistant antimicrobial and Cotrimoxazole

was highest resistance amongst all antimicrobials. About 58 (56.3%) of the uncomplicated UTI patients had resistant to Amoxycillin, 60 (58.3%) to Cotrimoxazole, 29 (28.2%) to Cefixime, 19 (18.4%) to Ciprofloxacin and 16 (15.5%) detected to Gentamicin.

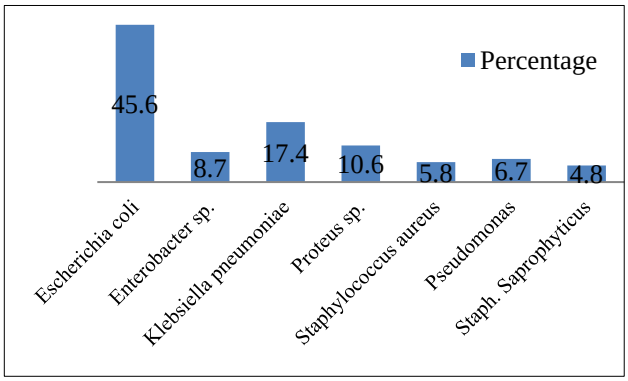

Figure 2: Isolated microorganism from urine samples with frequency (n=103).

Table 4 analyzes the antimicrobial sensitivity pattern of isolated uropathogens. The highest sensitivity was observed with imipenem (91.48%), followed by amikacin (87.23%), nitrofurantoin (85.10%), and gentamicin (85.10%). Moderate sensitivity was observed with ciprofloxacin (80.85%), cefixime (61.70%), and co-trimoxazole (46.80%). The lowest sensitivity was observed with amoxicillin (57.44%). The highest sensitivity was observed with cefixime (88.88%) and nitrofurantoin (94.44%). Moderate sensitivity was observed with ciprofloxacin (77.77%), imipenem (77.77%), and amikacin (83.33%). Lower sensitivity was observed with co-trimoxazole (33.33%) and amoxicillin (38.88%). These analyses provide insights into the antimicrobial sensitivity patterns of different uropathogens, which can inform treatment decisions and antimicrobial stewardship efforts.

Table 2: Isolated microorganism from urine samples with frequency (n=103).

Organism	Number of urine samples	Percentage
Escherichia coli	47	45.6
Enterobacter sp.	9	8.7
Klebsiella pneumoniae	18	17.4
Proteus sp.	11	10.6
Staphylococcus aureus	6	5.8
Pseudomonas	7	6.7
Sta. saprophyticus	5	4.8

Table 3: Overall sensitivity pattern of antimicrobials used in uncomplicated UTI patients (n=103).

Antimicrobials	Sensitive (n/%)	Resistant (n/%)
Amoxycillin	45 (43.7)	58 (56.3)
Cefixime	74 (71.8)	29 (28.2)
Cotrimoxazole	43 (41.7)	60 (58.3)
Imipenem	89 (86.4)	14 (13.6)
Ciprofloxacin	84 (81.6)	19 (18.4)
Nitrofurantoin	91 (88.3)	12 (11.7)
Gentamicin	87 (84.5)	16 (15.5)
Amikacin	90 (87.4)	13 (12.6)

Table 4: Antimicrobial sensitivity pattern of isolated uropathogens.

Antimicro-bials	*E. coli (%)*	*Enterobact -er sp. (%)*	*Klebsiella pneumonia (%)*	*Proteus sp. (%)*	*Staph. aureus (%)*	*Pseudomo-nas (%)*	*Staph. saprophyt --icus (%)*
Amoxicillin	27/47 (57.44)	1/9 (11.11)	7/18 (38.88)	3/11 (27.27)	4/6 (66.66)	0/7 (0)	3/5 (60)
Cefixime	29/47 (61.70)	7/9 (77.77)	16/18 (88.88)	9/11 (81.81)	5/6 (83.33)	5/7 (71.42)	3/5 (60)
Co-trimoxazole	22/47 (46.80)	5/9 (55.55)	6/18 (33.33)	4/11 (36.36)	1/6 (16.66)	3/7 (42.85)	2/5 (40)
Imipenem	43/47 (91.48)	9/9 (100)	14/18 (77.77)	2/11 (18.18)	4/6 (66.66)	6/7 (85.71)	4/5 (80)
Ciprofloxa-cin	38/47 (80.85)	8/9 (88.88)	14/18 (77.77)	9/11 (81.81)	5/6 (83.33)	6/7 (85.71)	4/5 (80)
Nitrofuran-toin	40/47 (85.10)	9/9 (100)	17/18 (94.44)	9/11 (81.81)	6/6 (100)	6/7 (85.71)	4/5 (80)
Gentamicin	40/4 (85.10)	7/9 (77.77)	15/18 (83.33)	11/11 (100)	5/6 (83.33)	5/7 (71.42)	4/5 (80)
Amikacin	41/47 (87.23)	8/9 (88.88)	15/18 (83.33)	10/11 (90.90)	5/6 (83.33)	7/7 (100)	4/5 (80)

Data were presented as frequency, percentage, n=number of study subjects

DISCUSSION

This cross sectional study was conducted to assess the isolation, identification and antimicrobial sensitivity pattern in female patients of uncomplicated urinary tract infections attending in the gynaecology and obstetrics and female medicine outpatient department in Sher-E-Bangla Medical College Hospital, Barishal.

In this study, age of the patient ranging from 18 to 74 years, majority of the patients 114 (57.0%) belonged to age group of 31-44 years. The mean age was found 42.6±11.4

years. Frequency of uncomplicated UTIs was predominant in rural female subject. Out of 200 cases 166 (83%) cases hailing from rural areas and 34(17%) from urban sites. Rural to urban ratio was 4.88:1. Present study shows that frequency of uncomplicated UTIs are predominant at middle to elderly age but more urban affected at elder age and rural female at earlier than urban. Highest incidence of urban and rural patients was in age group between 31-44 years, 52.9% and 57.83% respectively. It was observed that maximum patients e.g., 83 (41.5%) were housewives. Socioeconomically patients are grouped into three classes. Among the patients, the poor class 93 (46.5%) comprising the major percentage of the patients. In our study majority patients 114 (57.0%) belongs to age group of 31-44 years. A similar study done by Akram et al showed that more cases of UTI were among young and middle aged patients (20-49 years, 51.04%).[9] In 2009 another study by Ullah et al revealed that middle aged patients accounted for 54.35% of UTI.[10]

Maximum numbers of respondents were married (73.0%) in this study. Regarding occupational status of the patients, it was observed that maximum patients e.g., 83 (41.5%) were housewives followed by daily workers 22.5%, service holders 20.0%. Majority of the patients were poor class 93 (46.5%) population followed by middle class 76 (38.0%) and upper class 31 (15.5%). Study done by Gururaju, et al has also found that there was a high incidence of UTI among low socioeconomic group (62%), followed by middle class (30%), and high class (8%).[11]

In this study microbial culture result of uncomplicated UTI revealed that 103 (51.5%) of urine samples had significant bacteriuria. *E. coli* was found to be the most prevalent 47(45.6%), followed by *Klebsiella pneumoniae* 18 (17.4%), *Proteus* sp. 11(10.6%), *Enterobacter* sp. 9(8.7%) *Pseudomonas* (6.7%), *Staphylococcus aureus* (5.8%) and *Staphylococcus saprophyticus* (4,8%). This finding pattern was similar with the study of Ahmed et al showed *E. coli* (34%), *Klebsiella* species (16%), *Proteus* species (9.60%), and *Staphylococcus aureus* (8.40%).[12]

It also correlates with the study conducted by Biswas et al in which they demonstrated that out of 200 urine samples processed 110 (55%) gave significant growth of pathogens and E. coli remain the leading uropathogen being responsible for UTI (58.18%) followed by *Enterococci* (13.63%), *Pseudomonas* (9.09%), *Proteus* (8.18%), *Staphylococcus aureus* (4.54%), and *Klebsiella pneumonia* (3.63%).[13] In 2001 a study conducted by Abdul et al showed the similar findings with the present study where *Escherichia coli* (43.4%), *Klebsiella* (12.4%), *Proteus* sp. (9.7%) and *Pseudomonas* (8%) were found.[14] Study conducted by Shaifali in 2012 showed that the sensitivity to Nitrofurantoin is 86.95% against *E. coli* and 90.90% against *Klebsiella* which correlates with my study.[15]

Present study demonstrated that Nitrofurantoin (11.7%) was found less resistant and co-trimoxazole (58.3%) was highest group of resistant agent amongst all antibiotics those included in my study. About 58 (56.3%) of the uncomplicated UTI patients had resistant to Amoxycillin, 19 (18.4%) to Ciprofloxacin, 29 (28.2%) to cefixim. Biswas et al observed that cotrimoxazole and amoxicillin are virtually useless against uropathogens as they were effective against 31.81% and 11.81% of all isolated organisms respectively.[13]

In my study antimicrobial susceptibility testing to Escherichia coli strains isolated from urine samples showed highest sensitivity to Imipenem (91.48%) followed by Amikacin (87.23%), Nitrofurantoin (85.10%), Gentamicin (85.10%) and Ciprofloxacin (80.85%). It showed highest resistance to Co-trimoxazole (53.19%) followed by Amoxicillin (42.55%) and Cefixime (38.29%). *Enterobacter* sp. showed 100% sensitivity to Imipenem and Nitrofurantoin followed by Amikacin (88.88%), Ciprofloxacin (88.88%) and highest resistance to Amoxicillin (88.88%) and co-trimoxazole (44.44%). *Klebsiella* showed highest sensitivity to Nitrofurantoin (94.44%) followed by cefixime (88.88%), Amikacin (83.33%) and showed highest resistance to co-trimoxazole (66.66%) followed by Amoxicillin (61.11%). *Proteus* showed 100% sensitivity to Gentamicin followed by 90.90% to Amikacin, 81.81% to Nitrofurantoin, Ciprofloxacin and Cefixime and highest resistance to Amoxicillin (72.72%) and Co-trimoxazole (63.63%). *Staphylococcus aureus* showed highest sensitivity to Nitrofurantoin (100%) followed by 83.33% to Amikacin, Gentamicin, Ciprofloxacin and Cefixime and showed highest resistance to co-trimoxazole (83.33%) followed by 33.33% to Imipenem and Amoxicillin. *Pseudomonas* showed 100% sensitivity to Amikacin followed by 85.71% sensitivity to Nitrofurantoin, Ciprofloxacin and Imipenem and showed 71.42% sensitivity to Gentamicin and Cefixime. It showed 100% resistance to Amoxicillin followed by 57.14% to co-trimoxazole, 28.57% to Gentamicin and Cefixime. *Staphylococcus saprophyticus* showed 80% sensitivity to Imipenem, Ciprofloxacin, Nitrofurantoin, Gentamicin and Amikacin and highest resistance to co-trimoxazole (60%) followed by 40% resistance to Cefixime and Amoxicillin. A study conducted by Yasmeen et al showed that E. *coli* was found to be most sensitive to Imipenem and Amikacin and resistant to Cefixime and Co-trimoxazole. *Pseudomonas* was 100% sensitive to nitrofurantoin which is similar with my study.[16] Biswas et al showed that *E. coli* showed high sensitivity to Imipenem and Amikacin (100%), Gentamicin (94.1%), Ciprofloxacin (88.23%).[13] *Pseudomonas* showed sensitivity to Amikacin, Ciprofloxacin, Imipenem and Gentamicin. *Staphylococcus aureus* showed sensitivity to Nitrofurantoin and Amikacin (100%). Another study showed high sensitivity of *E. coli* to Amikacin and Nitrofurantoin and Ciprofloxacin. *Klebsiella* showed high sensitivity to Amikacin (89.65%) which also correlates with my study.[17] So isolation, identification and antimicrobial sensitivity pattern is essential to avoid drug resistance in the treatment of uncomplicated urinary tract infection.

Limitations

The study population was selected from one hospital in metropolitan city, so that the results of the study may not be reflect the exact picture of the country. A large-scale study needs to be conducted to reach to a definitive conclusion. Study was conducted in a tertiary care hospital which may not represent primary or secondary centre. Sample were taken by purposive method in which question of personal biasness might arise.

CONCLUSION

The antimicrobial resistance patterns are constantly evolving and vary from region to region and changing with time. Uncomplicated UTI is a frequently reported infection in women. Diagnosis is usually based on clinical symptoms and the demonstration of significant bacteriuria. A high prevalence of uncomplicated UTI was established in middle aged population and mainly rural and poor class population. The main pathogen was *E. coli* and this organism is beginning to acquire resistance to some of the clinically used antibiotics. This is mainly attributable to inappropriate indication, dose, and/or duration of therapy with antibiotics.

Recommendations

Need to develop educational campaigns aimed at raising awareness about the importance of appropriate antimicrobial use and the dangers of antimicrobial resistance. Implement antimicrobial stewardship programs in healthcare settings to promote the rational use of antibiotics and reduce unnecessary prescribing. Regular updated training program within health care providers. Facilities for prompt and adequate treatment of risk factors and screening should be available in all hospitals. Study should be conducted in multiple centers with a large sample size to get the real scenario of our country.

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