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Bacteriological profile and antimicrobial susceptibility patterns of symptomatic Urinary Tract Infection among patients of different age groups in a tertiary care hospital of Bangladesh

Mohammad Zakerin Abedin[1], Farida Yeasmin[2], Samim Mia[1,3], Hasanul Bari Helal[3], Rasheda Yasmin Shilpi[4]

[1]*Department of Microbiology, School of Biomedical Science, Khwaja Yunus Ali University, Bangladesh*
[2]*Faculty of Basic Medical and Pharmaceutical Science, University of Science and Technology, Chittagong, Bangladesh*
[3]*Department of Microbiology, Al-Hera General Hospital, Sadar, Sirajganj, Bangladesh*
[4]*Department of Botany, Jahangirnagar University, Savar, Dhaka, Bangladesh*

Abstract

Urinary tract infection (UTI) is one among the foremost prevalent diseases affecting people of both genders and all age groups. The management of UTI relies on the likely etiology of UTI and knowledge of their antimicrobial susceptibility patterns. Extensive application of antibiotics has led to the emergence of resistant microorganisms. This study was conducted to research the prevalence rate of UTI among patients of different age groups and to analyze the recent antibiotic susceptibility pattern of uropathogen in UTI because the antibiogram of the microorganisms is repeatedly changing. In a prospective study undertaken at Al-Hera General Hospital, Sadar, Sirajganj over a 9 month period, 159 samples from patients suspected of having UTI were analyzed, of which 57 were culture positive. Identification of the isolated bacteria was performed by standard tests, and antibiotic susceptibility was measured by disc diffusion method. The total predominance of UTI was 35.8% of which 78.9% were females and 21.1% were from males. Greater prevalence was detected in females as compared to males. The overall prevalence in females was high in the age range of 21-60 yr (57.8%), followed by 1-20 yr (22.2%) and > 60 yr (20%). In males the frequency is high in 21-40 yr (41.7%); both the age range of 41-60 yr and > 60 yr was 50%, and the rest of 8.3% for the age group of 1-20 yr. From the total 57 uropathogens, *E. coli* was estimated for 63.1% of all the isolate, followed by *Pseudomonas* spp. (7%), *Proteus* spp. (5.2%), *Klebsiella pneumoniae* (5.2%), *Staphylococcus saprophyticus* (5.2%), *Salmonella* spp. (3.5%), *Serratia* spp. (3.5%), *Citrobacter frundii* (5.2%), and *Candida albicans* (1.8%). In the present study, the most potent antibiotics were Amikacin, Imipenem, Nitrofurantoin, Meropenem, and Amoxiclav, while on the contrary higher resistance was observed among the commonly used drugs like Cephradine, Amoxicillin, Cotrimoxazole, and Ceftazidime. UTI may be a serious health problem if untreated. Initial diagnosis and immediate treatment will prevent the probabilities of developing further complication of UTI. Both the patient's age and gender can increase accuracy in defining the causative agents and providing a useful guideline to treat UTI. As the drug resistance pattern of the bacterial pathogens of UTI is greatly varied with time, regular surveillance and monitoring are vital for giving updated information to the physician for effective management of UTI.

Keywords: Predominance, Urinary tract infection, Uropathogen, Antibiogram susceptibility.

Introduction

The inhabitance of microbial pathogens in the urinary tract is termed as Urinary tract infection (UTI). The involvement of the bladder and urethra is referred to as lower UTI, whereas the involvement of the kidney and urethra is

CONTACT Mohammad Zakerin Abedin ✉ zakerin.du2016@gmail.com 🖃 Department of Microbiology, School of Biomedical Science, Khwaja Yunus Ali University, Bangladesh

referred to as upper UTI (John *et al*., 2016).

UTI is very common infectious diseases affecting people from all age groups, including neonate and old-age groups. Every year approximately 150 million people are being diagnosed with UTI globally. 60% of women have a lifelong risk of getting UTI in comparison to men who have a risk of only 13% (Nalini *et al*., 2014).

UTI may be asymptomatic, symptomatic and complicated or uncomplicated. The symptoms of UTI depend on the part of the urinary tract involved, the etiologic organisms, the severity of the infection, and the patient's capability to augment an immune response to it. The symptoms of UTI include fever, burning sensation while urinating, LAP, genital and supra pubic pain and pyuria (Amali *et al*., 2008).

Healthy individuals are affected mostly by uncomplicated UTI which includes cystitis and pyelonephritis. The factors which cause complicated UTI include urinary obstruction, pregnancy, renal failure, kidney transplantation, and the existence of external bodies like indwelling catheters, calculi or other drainage devices (Flores-Mireles *et al*., 2015). UTI is more common in females than males due to the short length of the urethra, pregnancy, easy contamination of the tract with fecal flora and absence of prostatic secretion (Sing Randhir *et al*., 2016). UTI commonly develops in the two genders and transversely all age groups. Despite several different microorganisms which can cause UTI including viruses and fungi, the major causative agents are bacteria and are liable for more than 95% of UTI causes. *Escherichia coli* is the most common organism which accounts for up to 90% of UTI cases, whereas *P. aeruginosa*, *Klebsiella* spp., *P. mirabilis*, and *Enterobacter* spp. are less frequent malefactors. Gram-positive organisms are less common which includes Group B *Streptococcus*, *S. haemolyticus*, *S. Saprophyticus*, and *S. aureus* (Loh & Sivalingam, 2007).

UTI is mostly treated tentatively without urine culture or susceptibility testing which may be relevant to the recurrent misapplication of antibiotics. The statistics on antimicrobial susceptibility of UTI-causing microorganisms is inconstant. Most frequently UTI are treated provisionally. So the antimicrobial agents should be selected on the basis of the possible pathogen and its expected resistance pattern in a certain geographic region.

The repeated supervision of the causes of UTI and their susceptibility pattern in the community is therefore very essential. Hence the study was done to determine the etiology of UTI and susceptibility pattern of isolated uropathogens to recent antimicrobial agents in an area of Sirajganj sadar, Bangladesh in addition to evaluate the age and gender wise distribution which may help and guide the medical practitioners to carry out practical treatment.

Material and methods

A recollective study was done on UTI at the Microbiology department of Al-Hera General Hospital, Sadar, Sirajganj throughout the term of January to September 2020. A total of 159 samples were collected throughout this study term.

Specimen collection

To obtain a urine sample, patients were instructed to collect midstream clean catch urine in a wide, sterile container supplied by the laboratory.All patients were advised to maintain a proper aseptic procedure prior to urine collection. All the samples were collected before the beginning of antibiotic treatment.

Cultivation and identification of isolates

Specimens of urine were plated using calibrated wire loops (0.002 ml) on

Nutrient agar, 5% sheep blood agar, and MacConkey's agar following standard bacteriological technique then incubated aerobically at 37 °C for 24 hours. After incubation the numbers of bacterial colonies were counted.

The number of bacteria was measured as colony-forming units (CFU) per ml of urine. Urine culture having a colony count $\geq 10^5$ CFU/ml was considered as significant. Identification of the isolated uropathogen was done by colony morphology of the organisms on culture media and biochemical tests specific for uropathogens.

Antimicrobial susceptibility testing

Kirby-Bauer disk diffusion method was used to determine the susceptibility of the isolates to the antimicrobial agents (Bauer *et al.*, 1966).

Muller Hinton agar was inoculated with standard inoculum and incubated for 24 h at 37 °C. The antimicrobial agents tested in this study were: Amoxicillin (30 µg), Amikacin (30 µg), Amoxiclav (30 µg), Aztreonam (30 µg), Ceftazidime (30 µg), Ceftriaxone (30 µg), Cefixime (5 µg), Ciprofloxacin (5 µg), Cefuroxime (30 µg), Cotrimoxazole (25 µg), Gentamicin (10 µg), Cephradine (25 µg), Imipenem (10 µg), Nitrofurantoin (300 µg), Meropenem (10 µg), Levofloxacin (5 µg), and Azithromycin (15 µg). Results were interpreted as stated by the Clinical Laboratory Standards Institute (CLSI, 2009).

Data processing and analysis

Excel 2016 and SPSS version 20 were used to analyze the data. Descriptive statistics and chi-square tests were done to check the statistical evaluation. "< 0.5" was the significant value of the p-value considered in this study.

Results

A total of 159 cases of symptomatic UTI were studied in 9 months among these 102 (64.2%) samples showed no growth of bacteria and 57 (35.8%) urine samples showed positive cases. Among the positive cases, 45 (78.9%) of females and 12(21.1%) of males cases. In females high prevalence was seen with age group 21-60 years (57.8%), followed by 1-20 yr (22.2%), and > 60 yr (20%). In males high prevalence was seen with age group 21-40 yr (41.7%), both the age range of 41-60 yr and > 60 yr was 50%, and the remaining 8.3% for the age group of 1-20 yr (Table 1).

From the total 57 positive culture, *Esherichia coli* was the foremost common isolate causing UTI in both genders with a prevalence rate of 63.1%. The second most prevalence isolates were *Pseudomonas aeruginosa* (7%), followed by *Klebsiella pneumoniae* (5.3%), *Proteus* spp. (5.3%), *Staphylococcus saprophyticus* (5.3%), *Citrobacter* spp. (5.3%), *Salmonella* spp. (3.5%), *Serratia* spp. (3.5%), and *Candida albicans* (1.7%) (Figure 1). It was also observed that the prevalence of Gram negative bacteria (93%) was too much higher than the both Gram positive bacteria (5.3%) and fungi (1.7%) (Table 2).

The isolation rate of all the isolated bacterial species is significantly different among the four selected age groups (Table 3*).* In the adult subjects (19-59 yr), *Escherichia coli* was more common (29.8%), followed by senior adult (> 60 yr), 21% and less in child (< 12 yr), 5.3% (Table 3).

Table 1. Age and gender based distribution of cases of urinary tract infections.

Age groups	Number of patient (%)	Female patients (%)	Male patients (%)
1-20 yr	11 (19.3)	10 (22.2)	1 (8.3)
21-40 yr	18 (31.6)	13 (28.9)	5 (41.7)
41-60 yr	16 (28.1)	13 (28.9)	3 (25.0)
> 60 yr	12 (21.0)	9 (20.0)	3 (25.0)

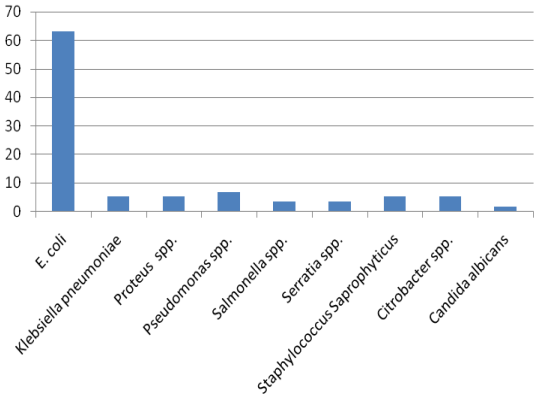

Figure 1. Distribution of causative agents of urinary tract infection.

Table 2. Prevalence of Gram positive, Gram negative bacterial isolates and fungi of urinary tract infection.

Organisms	Percentage (%)
Gram negative	
Escherichia coli	63.1
Klebsiella pneumoniae	5.3
Proteus spp.	5.3
Pseudomonas spp.	7
Salmonella spp.	3.5
Serratia spp.	3.5
Citrobacter spp.	5.3
Gram positive	
Staphylococcus saprophyticus	5.3
Fungi	
Candida albicans	1.7

Susceptibility of the main isolated uropathogens to frequently used antibiotics is shown in Figure 2. Susceptibility of *E. coli* to test antibiotics ranged from 13.9% (Cephradine) to 86.1% (Amikacin). Nitrofrantoin and Imepenem activity rate was 83.3%, followed by Amoxiclav, Meropenem (58.3%), Cefuroxime, and Gentamicin (52.8%). *E.coli* showed resistance to Ceftazidime and Amoxicilin (52.8%, 55% respectively), whereas *Pseudomonas* spp., *Klebsiella* spp., *Serratia* spp., and *Salmonella* spp. exhibit 100% resistance to Amoxicilin.

Pseudomonas spp. showed highest sensitivity to Meropenem (100%), followed by Imipenem (75%) and Amikacin (50%). Highest sensitivity was observed by *Klebsiella* spp. to Amikacin, Meropemem, Gentamicin, and Nitrofurantoin (100%), followed by Amoxiclav, Cefuroxime, and Ciprofloxacin (66.7%). *Proteus* spp. also showed higher sensitivity to Amikacin and Nitrofurantoin (100%), followed by Gentamicin (66.7%), Cefixime, Amoxiclav, and Amoxicillin (33.3%).

Table 3. Distribution (%, frequency among patients) of uropathogens that cause urinary tract infection according to age category.

Uropathogens	Child (< 12 yr) (n=5)	Adolescent (13-18 yr) (n=4)	Adult (19-59 yr) (n=34)	Senior Adult (> 60 yr) (n=14)
E.coli	5.3	7.1	29.8	21.0
Klebsiella pneumoniae	0	0	5.3	0
***Proteus* spp.**	0	0	3.5	1.7
***Pseudomonas* spp**	0	0	7.1	0
***Salmonella* spp**	3.5	0	0	0
***Serratia* spp.**	0	0	3.5	0
Staphylococcus saprophyticus	0	0	3.5	1.7
***Citrobacter* spp.**	0	0	5.3	0
Candida albicans	0	0	1.7	0

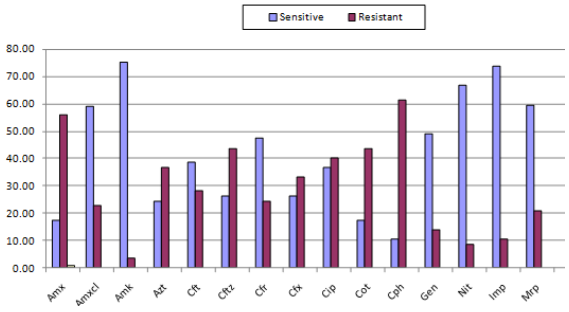

Figure 2. Overall antibiotic susceptibility pattern of uropathogens. Amx-Amoxycillin, Amxcl-Amoxyclav, Amk-Amikacin, Azm-Azithromycin, Cft-Ceftriaxone, Cftz-Ceftazidime, Cfr-Cefuroxime, Cfx-Cefixime, CIP-Ciprofloxacin, Cot-Cotrimoxazole, Cph-Cephradine, Gen-Gentamycin, Nit-Nitrofurantoin, Ipm- Imipenem, Mrp-Meropenem.

Salmonella spp. showed 100% sensitivity to Amikacin, Ceftriaxone, Cefuroxime, Ciprofloxacin, Gentamicin,

Table 4. Overall susceptibility (%) of the uropathogens to the antibiotics.

Antibiotics	*E. coi* (n=36)	*Pseudomonas* spp. (n=4)	*Klebsiella* spp. (n=3)	*Proteus* spp. (n=3)	*Salmonella* spp. (n=2)	*Serratia* spp. (n=2)	*Staph.* spp. (n=3)	*Citrobacter* spp. (n=3)
AMX (30 µg)	50	100	100	66.8	100	100	33.3	0
AMCL (30 µg)	25	50	33.3	0	0	0	33.3	0
AK (30 µg)	2.8	25	0	0	0	0	0	100
ATM (30 µg)	41.7	0	66.7	100	50	0	0	100
CRO (30 µg)	41.7	25	0	0	0	0	0	100
CAZ (30 µg)	52.8	25	33.3	33.3	100	0	0	0
CFM (5 µg)	41.7	50	0	0	0	0	33.3	0
CFX(30 µg)	33.3	0	0	33.3	0	50	0	100
CIP (5 µg)	44.4	25	33.3	100	0	50	33.3	100
COT (25 µg)	47.2	0	66.7	100	100		33.3	66.7
CPH (25 µg)	66.7	50	66.7	100	50	100	0	0
CN (10 µg)	16.7	0	0	33.3	0	0	33.3	66.7
NIT (300 µg)	13.9	0	0	0	0	0	0	66.7
IMP (10 µg)	5.6	25	33.3	33.3	0	0	0	33.3
MEM (10 µg)	27.8	0	0	0	100	0	0	66.7
LEV (5 µg)	41.7	---	---	---	---	---	---	---
AZM (15 µg)	---	---	---	---	100	---	---	---

AMX-Amoxycillin, AMXCL-Amoxyclav, AK-Amikacin, ATM-Aztreonam, CRO-Ceftriaxone, CAZ-Ceftazidime, CFM-Cefuroxime, CFX-Cefixime, CIP-Ciprofloxacin, COT-Cotrimoxazole, CPH-Cephradine, CN-Gentamycin, NIT-Nitrofurantoin, IPM-Imipenem, MEM-Meropenem, LEV-Levofloxacin, AZM-Azithromycin. (---) Not used.

Azithromicin, and also showed 100% resistance to Ceftazidime, Cotrimoxazole, and Meropenem.

100% sensitivity was also shown by *Serratia* spp. to Amoxiclav, Amikacin, Ceftazidime, Cotrimoxazole, and, Nitrofurantoin, and showed higher resistance to Amoxicilin and Cephradine (100%).

100% sensitivity to Cefuroxime, Meropenem, and Imipenem were found in *Staphylococcus saprophyticus.* Highest sensitivity (100%) to Amikacin, Aztreonam, Ceftriaxone, Cefuroxime, and Ciprofloxacin were showed by *Citrobacter frundii*, followed by 66.7% to Cotromoxazole, Meropenem, Gentamicin, and Nitrofurantoin.

Discussion

UTI is diagnosed as one of the most frequent diseases worldwide and developing countries in particular like Bangladesh. Reachability of new antimicrobials has improved the treatment of UTI. Monitoring of native UTI's etiology and antimicrobial susceptibility is considered helpful to design empirical treatment, as the frequency of pathogens and their characteristics may change with geographical range and time (Livermore & Pearson, 2007).

UTI is predominantly a disease of the female due to short urethra and its closeness to the anus (Jawetz & Melninick, 1995). The ubiquity of UTI in female 78.9% compared to that of male was observed in present study which was similar to that reported by Abedin *et al*. (2020). The prevalence of urinary tract infections varies in consonance with sex and age (Kumamoto *et al.*, 1996). In the present study regarding age and sex distribution, the majority of the cases are female in the age group of 21 to 60 yr (57.8%). According to works cited by Yasmeen *et al.*, (2015), the majority of the cases are in the age group of 21-30 y

(32.3%). Other studies on UTI in Bangladesh revealed that most of the cases of UTI occurred between the 21-30 yr and 41-50 yr age groups (44% and 37% in the same order) which support our studies mostly.

In the present study, the Gram negative bacilli contributed to 93% of the total isolates, while the Gram positive cocci constituted 7%. In our study *E.coli* was the most commonly detected species. There were 36 (63.2%) out of 57 positive cases of *E. coli*. This result is persistent with reports from other studies by Agbawa & Ifeanacho-Emeka (2015), 63.3% and Das & Banerjee (2015), 60.3% and it was most frequent pathogen causing UTI in all these studies. The logic for the highest rate of *E.coli* is that they are normal fecal flora and fimbriae or pili act as an adherence factor to mediate the attachment of *E.coli* to epithelial cells of the urinary tract (Sood & Gupta, 2012). Other researchers also reported a higher association with *E. coli* (Abubakar, 2009; Haque *et al.*, 2015).

In this study *Pseudomnas* spp. (7%), *Klebsiella* spp., *Proteus* spp., *Staphylococcus saprophyticus* (5.3%), *Serratia* spp., *Salmonella* spp., and *Citrobacter frundii* (3.5%) are the other frequently identified isolates all having been reported to be highly prevalent species in UTI. In our study *Pseudomnas* spp. (7%) was the second most prevalent uropathogen, whereas in other studies *Klebsiella* spp., was the second most prevalent isolate (Sanjee *et al.*, 2017) in UTI which indicated that Gram negative bacteria, mostly *E.coli*, *Klebsiella* spp., and *Pseudomnas* spp., are the common pathogens isolated in the patient having UTI.

Sensitivity of uropathogens to antimicrobial agents is also known to differ among countries and over time. In our study the foremost common uropathogen *E.coli* were sensitive to Amikacin (86.1%), followed by Nitrofrantoin and Imepenem (83.3%), Amoxiclav, Meropenem, (58.3%), Cefuroxime, and Gentamycin (52.8%). *E.coli* showed resistance to commonly used drugs like Cephradine, Ceftazidime, Amoxicilin, Cotrimoxazole, Ciprofloxacin, and Ceftriaxone (66.7%, 52.8%, 50%, 47.2%, 44.4%, and 41.7% respectively). Parveen & Rahim (2018) has cited a highest sensitivity to Amikacin and Nitrofrantoin (87.5% and 83.3% respectively) which is similar with present study but different findings for sensitivity was observed to Imipenem which is 100% and also found that *E.coli* was most resistant to commonly used drugs like Cephalexin, Cefuroxime, and Cotrimoxazole (100%, 75%, and 54.2% in the same order). Khondakar *et al*. (2016) have likewise the highest sensitivity to Imipenem (100%) and Amikacin (93%).

It was noted that the susceptibility of the isolates to the tested antibiotics varied with the species in this study. Susceptibility to Amikacin was higher in *Klebsiella* spp., *Proteus* spp., *Seratia* spp., *Staphylococcus saprophyticus*, and *Citrobacter* spp. (100%) in comparison to *E.coli* (86.1%).

Pseudomonas spp. and *Klebsiella* spp, showed 100% susceptibility to Meropenem, whereas *Salmonella* spp. and *Citrobacter* spp. showed 100% sensitivity to Ceftriaxone, Cefuroxime, and Cotrimoxazole. 100% resistance to Amoxicilin was observed by *Pseudomonas* spp., *Klebsiella* spp., *Serratia* spp., and *Salmonella* spp., whereas *Proteus* spp. and *Staphylococcus saprophyticus* showed 66.7% and 33.3% resistance respectively.

The overall sensitivity to the commonly used antibiotics like Cephradine (61.4%), Amoxicilin (56.1%), Cotrimoxazole (43.9%), Ceftazidime (43.8%), and Ciprofloxacin (40.4%) in present study is observed significantly low. The best active antimicrobial agents in our study were Amikacin (75.4%), Imepenem (73.7%), Nitrofurantoin (66.7%), and Meropenem (59.7%). Nowadays, resistance to antibiotics is an increasing problem throughout the world. Continuous estimation of the susceptibility pattern of

uropathogens serves as a protocol for antibiotic therapy because these organisms showed resistance to most of the commonly prescribed antibiotics for the treatment of UTI. To overcome this present challenge of drug resistance appropriate therapy as per bacterial sensitivity pattern is required to be initiated.

Conclusion

It has been concluded that *E.coli* was the principal uropathogen, followed by *Pseudomonas* spp., *Klebsiella* spp., *Proteus* spp., *Citrobacter* spp., *Serratia* spp., *Salmonalla* spp., *Staphylococcus saprophyticus*, and *Candia albicans*. Despite being an infection that affects both genders and all ages in female the high prevalence of UTI was observed. In females high prevalence was seen among the 21-40 yr age group, followed by the 41- 60 yr one, and in males high prevalence was seen among the 21-40 yr age group, followed by the > 60 yr one. In our study elevated resistance was found to commonly used antibiotics like Amoxicillin, Cephradine, Cotrimoxazole, and Ceftazidime. However majority of organisms are sensitive to Amikacin, Imepenem, Nitrofurantoin, and Amoxiclav, hence given a better choice for treatment of UTI. For this reason, the choice of antimicrobials for UTI should be guided by culture and sensitivity, and empirical therapy must be considered on the recent antibiogram of a particular geographical area which takes into an account the patient's gender and age.

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Conflicts of interest

The authors declare that they have no conflicts of interest.

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