

Antibiotic Resistance Pattern of Uropathogens Causing Urinary Tract Infection

*Azad MK,¹ Abedin MJ,² Das DC,³ Sultana S,⁴ Belal MM,⁵ Rahman MA⁶

Urinary tract infection is a very common disease that can affect anyone at any age where the infections rate is higher in women than man. This study aimed to ascertain the current situation of urinary tract infections (UTIs) and drug resistance determination caused by uropathogens in different age groups of patients. A retrospective data analysis of culture results of urinary pathogens was performed. This study determined the antibiotic susceptibility of 134 isolates of uropathogens from the urine culture of 497 patients. Urine samples were aseptically collected in sterile containers and then cultured, susceptibility to 13 antibiotics was tested by the disc diffusion technique. Among 134 cultured confirmed patients 69% were females and 31% were males. It state that female are more vulnerable to UTI than male ($p < 0.001$). The onset of UTIs in female was high at the age between 21-40 years (17.2%). Studies revealed that the most resistance rate for uropathogens from urine culture were amoxicillin, azithromycin, cefixime, ceftriaxone, ciprofloxacin and cotrimoxazole. Whereas, meropenem was found to be the most effective against the uropathogens followed by amikacin.

[SSNI Med Col J 2016 Jul; 1 (2):103-109]

Key words: Urinary tract infection, antibiotic resistance, uropathogens

Introduction

Urinary tract infection (UTI) is one of the most common bacterial infections in many developing countries like Bangladesh where proper sanitation is not maintained adequately. Each year upto 150 million individuals are affected by UTIs worldwide.¹ UTIs affect all age group individuals and diagnosed in both hospitalized as well as out patients. This burden causes serious impact on the socioeconomic life of individuals and also leads to a large proportion of antibacterial drug consumption.² In the third world, causative factors for UTI include poor hygiene, long time catheterization, uncontrolled sexual intercourse, pregnancy and spermicidal contraception.³ Colonization of normal and opportunistic microflora is a main causal agent for UTIs. Majority of pathogens are

gram-negative species, with predominance of members of Enterobacteriaceae.⁴ Unfortunately, several studies have reported that uropathogens may become resistant to a wide range of antibiotics due to abuse, over dosage use, non-prescribed use, incomplete dosages and ease of access to antimicrobial drugs.⁵ Increasing antibiotic resistance in pathogens is a worldwide concern. The prevalence of antibiotic resistance among UTI agents is also increasing,⁶ and its treatment has become more complicated due to increasing resistance and empirical therapy leading to treatment failures mostly associated with gram-negative bacteria.⁷ The present study investigated the pattern of gram-negative uropathogens and their antibiotic resistance pattern among the clinical isolates to commercially available antibiotics that are often prescribed in UTI cases.

1. *Dr. Mohammad Abul Kalam Azad, Assistant Professor, Department of Medicine, Shaheed Syed Nazrul Islam Medical College, Kishoreganj. drmakazad80@gmail.com
2. Dr. Md. Joyanal Abedin, Assistant Professor, Department of Medicine, Shaheed Syed Nazrul Islam Medical College, Kishoreganj
3. Dr. Dulal Chandra Das, Assistant Professor, Department of Medicine, Shaheed Syed Nazrul Islam Medical College, Kishoreganj
4. Dr. Safia Sultana, Assistant Professor, Department of Microbiology, Shaheed Syed Nazrul Islam Medical College, Kishoreganj
5. Dr. Munshi Mohammad Belal, Consultant (Medicine), 250-Bedded General Hospital, Kishoreganj
6. Dr. Md. Aatur Rahman, Consultant (Medicine), 250-Bedded General Hospital, Kishoreganj

*For correspondence

Methods

This a retrospective study of the antibiotic resistance profile of 134 positive urine culture samples from 497 urine samples over a period of 6 months (From December, 2015 to May 2016) at 250 Bed District Hospital, Kishoreganj, Bangladesh. Patients were identified and data were extracted using the hospital information and support system.

Laboratory Techniques

In brief, one calibrated loopful urine samples were streaked on to MacConkey agar (MAC) and Blood agar media and incubated aerobically at 37⁰C for 24 hours for isolation and identification of urinary pathogens. The colony characteristics were then examined. The pink colonics on MAC were lactose fermenters and most likely to be E. coli, Klebsiella or other coliforms. If colonies were colorless, they were lactose non-fermenters. The clinical isolates were characterized by gram-stain, oxidase test, motility, citrate utilization and biochemically for species identifications.⁸

Antibiogram

Bacterial susceptibility to antimicrobial agents was determined in vitro by using agar disc diffusion method following CLSI guidelines.⁹ The following antibiotic discs were used : amikacin (30 µg), amoxicillin (10 µg), azithromycin (15 µg), cefixime (5 mg), ceftriaxone (30 µg), ciprofloxacin (5 µg), cotrimoxazole (25 µg), gentamycin (10 µg), nitrofurantoin (30 µg), meropenem (10 µg), cefuroxime (30 µg), linezolid (30 µg), ceftazidime (30 µg). After incubation at 37⁰C for 24 hour, the diameter of inhibition zone was measured by digital slide calipers and interpreted as “Sensitive”, “Intermediate” or “Resistant” according to CLSI standard interpretative charts.

Statistical Analysis

The data were analyzed using SPSS version 16 statistical software for Confidence Interval (CI) and P value. Statistical significance was defined when P value is <0.05. The confidence interval was set at 95% level of significance for all the proportions.

Results

A total of 134 UTIs caused by uropathogens were confirmed on urine culture of 497 patients. Among them 69% patients were females (93) and 31% were males (41). It revealed that female were more vulnerable to UTI than male (p<0.001). To analyze the prevalence of UTI at different age group, a gender based age group variation of UTI was analyzed. It was observed that, in each and every age group the percentage of female patients were higher than male. The percentage of male and female patients was highest in the age group above 50 years which was 14.9% and 16.4%, respectively. The lowest percentage of male and female patients was in the age group <10 years which was 0.75% and 3% respectively. The present data stated that, the on set of UTIs in female were higher at the age between 21-40 years 17.2% (Figure 1).

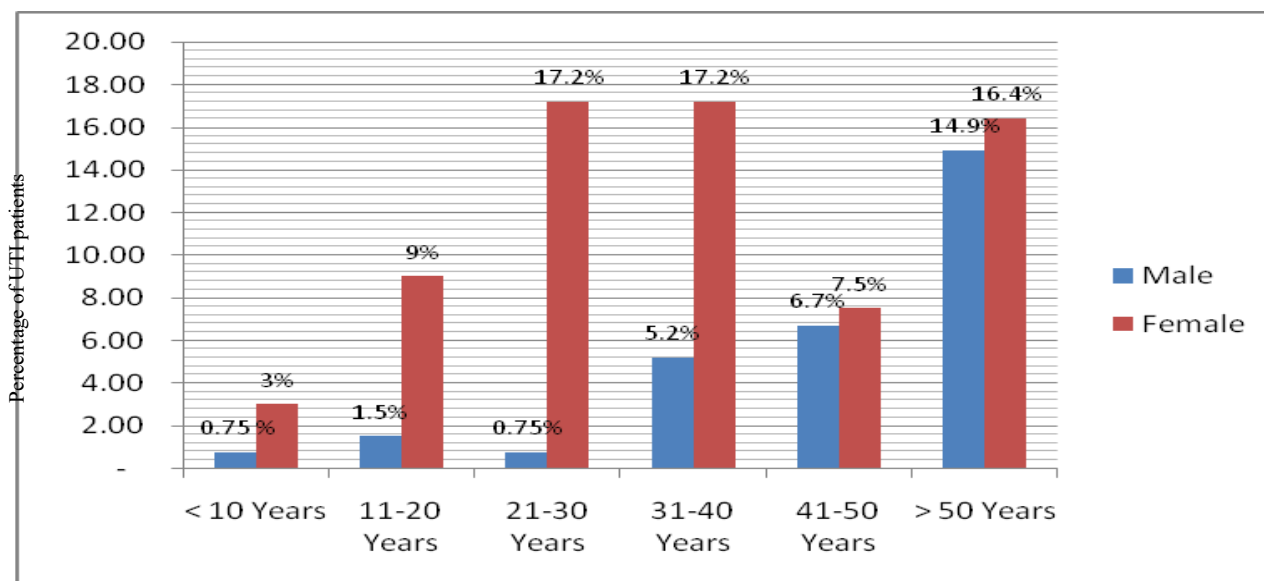


Figure 1. Percentese of male and female UTI patients in different age groups in this study
The number and percentage of each uropathogens isolated from urine samples are shown in figure 2.

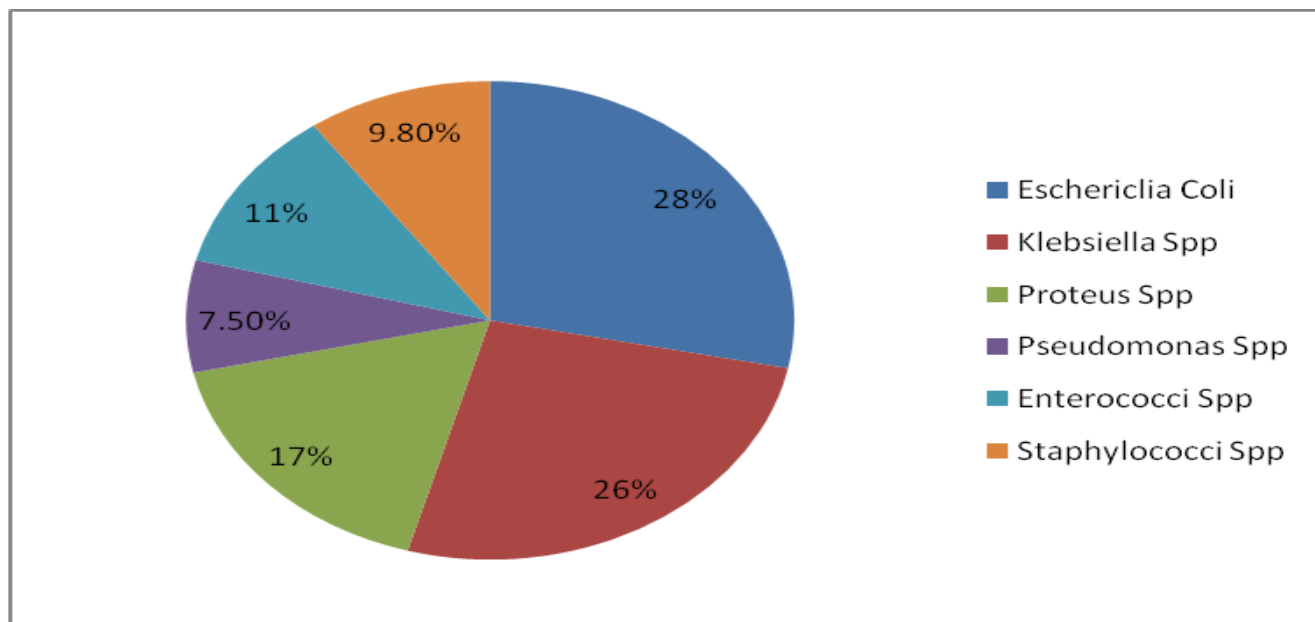


Figure 2. Comparative prevalance of bacterial species in urine sample.

Amongst the 134 isolates Escherichia coli was found to be the most frequently isolated organism 38 (28%), followed by Klebsiella pneumoniae 35 (26%), Proteus Spp 23 (17%), Enterococci Spp 15 (11%), Staphylococci 13 (9.8%) and Peudomonas Spp 10 (7.5%). Antibiotic resistance pattern of uropathogens isolated from this study was shown in Table 1.

Table 1: Antibiotic resistance pattern of individual isolates

Bacterial SPP	Antibiotic Used												
	Amoxi	AK	AZ	Cipro	Cot	CFM	Cro	Cefu	Nit	Linez	MP	Gen	Caz
Escherichia Coli	79.94	0	0	65.79	65.79	60.5	47.35	0	13.16	2.6	0	28.95	7.9
Klebsiella Spp	51.42	14.28	34.29	62.86	17.14	60.0	62.86	48.57	42.85	8.87	14.28	20.00	8.57
Proteus Spp	88.24	17.65	58.82	64.7	5.88	58.82	35.29	70.59	58.82	0	23.52	5.88	11.76
Pseudomonas Spp	80.00	20.00	80.00	90.00	0	60.0	80.00	90.00	60.00	20.00	10.00	10.00	40.00
Enterococci Spp	73.33	33.33	40	55.33	0	55.33	46.66	73.33	60	33.33	6.66	6.66	6.66
Staphylococci Spp	76.9	7.69	38.46	61.53	7.69	76.9	61.53	84.62	38.46	15.38	30.77	23.07	0

Abbreviations : Amoxi – Amoxicillin, AK – Amikacin, AZ – Azithromycin, Cipro – Ciprofloxacin, Cot – Cotrimoxazole, CFM – Cefixime, Cro – Ceftriaxone, Cefu – Cefuroxime, Nit – Nitrofurantoin, Linez – Linezolid, MP – Meropenem, Gen – Gentamycin, Caz – Ceftazidime.

Data are given as percentage of resistance.

E. coli showed highest resistance to amoxicillin (78.94%), ciprofloxacin (65.79%), cotrimoxazole (65.79%) and cefixime (60.5%). *Klebsiella* showed highest resistance to ciprofloxacin (62.86%), ceftriaxone (62.86%), cefixime (60%) and amoxicillin (51.42%). The *Proteus* had highest resistance to amoxicillin (88.24%), cefuroxime (70.59%), ciprofloxacin (64.7%) and cefixime (58.82%); while *Pseudomonas* had resistance to ciprofloxacin (90%), cefuroxime (90%), amoxicillin (80%) and azithromycin (80%). The *Enterococcus* had amoxicillin resistance (73.33%), cefuroxime (73.33%) and cefixime (53.33%); while *Staphylococcus* had resistance to cefuroxime (84.62%), cefixime (76.9%), amoxicillin (76.9%) and ciprofloxacin (61.53%).

Discussion

Amongst all infectious diseases urinary tract infections (UTIs) represent one of the most common disease in both developed and

developing countries causing a large number of morbidity in different age group.¹⁰ In this study, from the screened cases 134 were confirmed with UTIs, where *E. coli* was mostly the etiologic agent (28%). The finding of this study is in concordance with that of others showing *E. coli* (47.6%) and 49.4% as the predominant organism respectively.¹¹⁻¹² Among 134 UTIs patients the percentage of female (69%) were more compared to male patients. The finding of this study is in similar with that of others.¹³ The prevalence of UTIs in female was higher in the age group between 21-40 years (17.2%). The lowest percentage of female patients was in the age group <10 Years (3%). Among the female patients adult women were observed to be very much prone to UTIs. The similar finding was also shown in various earlier studies.¹⁴ Antibiotic resistance is a major clinical problem in treating infections caused by microorganisms. The resistance to the antibiotic has increased over the years. In this

study, the range of resistance of E.coli to amoxicillin was 78.94% and that for cotrimoxazole was 65.79%. This resistance were higher than those in studies from developed countries.¹⁵ On the other hand, similar rate have been found in other studies.¹⁶ Therefore these drugs should no longer be prescribed as initial empirical therapy in our region. The susceptibility pattern of E.coli to meropenem and amikacin was very significant. The resistance rate of E.coli to ceftriaxone (47.35%) and cefixime (60.5%), which is contrary to other community acquired UTI studies in Europe and USA.¹⁷ In this present study, Klebsiella Spp were shown high sensitivity to amikacin, meropenem and ceftazidime. The resistance rate to cefixime (60%), ceftriaxone (62.86%), cefuroxime (48.57%) and ciprofloxacin (62.86%). Isolates of Pseudomonas Spp were shown very high resistance to most antibiotics. The rate of resistance to amoxycillin, azithromycin, ciprofloxacin, cefixime, ceftriaxone, cefuroxime and

nitrofurantoin were 80%, 90%, 60%, 80%, 90% and 60% respectively.

Among antibiotics used in this study. Pseudomonas Spp isolates were very sensitive to amikacin, gentamycin, and meropenem. Resistance rate to amoxycillin, ciprofloxacin and cefuroxime among Proteus Spp isolates was very high (88.24%, 64.7% and 70.59% respectively). This study also showed that Enterococci Spp isolates were resistant to amoxycillin (73.33%), cefuroxime (75.33%), nitrofurantoin (60%) and cefixime (55.33%). The resistance rate to azithromycin, cefixime and ciprofoxacin were moderate (40-55%). The Staphylococci Spp isolates were shown high resisance to amoxycilin, cefuroxime, cefixime, ceftriaxone and ciprofloxacin (76.6%, 84.62%, 76.9% and 61.53% respectively). According to this study, all the isolates were shown difference in resistance between amoxycilin, azitrhomycoin, ciprofloxacin, cotrimoxazole, cefixime, ceftriaxone, cefuroxime, nitrofurantoin and gentamycin.

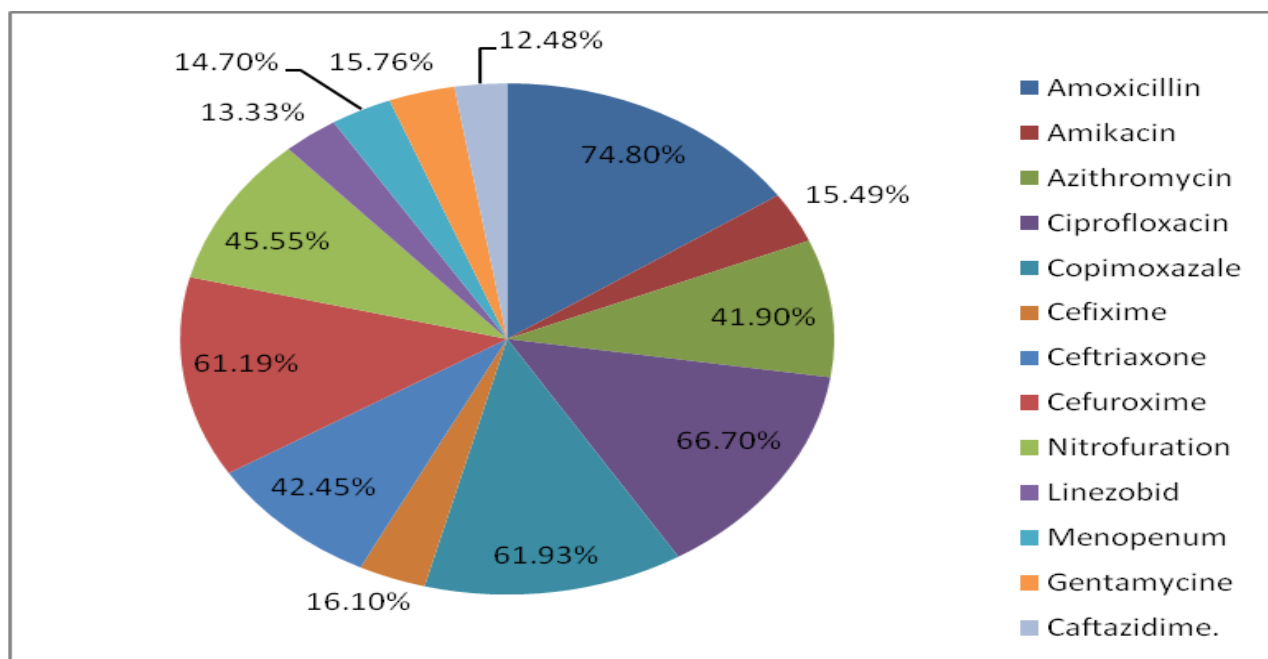


Figure 3. Comparison of antibiotic resistance profile.

Increasing resistance rate to all antibiotic used in this study with the exception of amikacin and meropenem, may be explained as uncontrolled consumption of these antibiotics during the past decade in our region.¹⁸

Finally it can be concluded that the present study demonstrate that the uropathogens isolated have shown various trends in isolation rate and resistances to various classes of antibiotics. In addition, multidrug resistant *E.coli*, *Klebsiella Spp*, *Pseudomonas Spp* can be expected to cause serious treatment problems. As the antibiotic resistance is an important factor, urine culture and antibiotics sensitivity test should be routinely performed in all UTIs patients, depending on the antibiotic susceptibility therapy should be designed of an antibacterial agent with the narrowest spectrum, least cost and few adverse effects.

Conclusion

It is quite alarming to note that almost all of the isolates included in this study were found resistant to multiple drugs (Four or more antibiotics). Antibiotic resistance has been emerged as a major problem in the management of hospitalized patients as well those with chronic conditions and adds considerably to health care cost. Our research has been confined to a particular region of Bangladesh. Larger samples sizes from other regions of Bangladesh can lead to more significant results. Therefore, it is increasingly important to study these resistant uropathogens, including mechanisms of resistance, in order to develop effective future drugs.

References

1. Stamm Walter E, and Norrby SR. (2001). Urinary Tract Infections: Disease Panorama and Challenges. The Journal of Infectious Diseases 183, S1- S4.
2. Dada-Adegbola HO and Muili KA 2010. Antibiotic susceptibility pattern of urinary tract pathogens in Ibadan, Nigeria. Afr. J. Med. Med. Sci., 39; 173-179.
3. Manges AR, Tabor H, Tellis P, Vincent C, and Tellier PP. (2008). Endemic and Epidemic Lineages of *Escherichia coli* that Cause Urinary Tract Infections. *Emerg Infect Dis* 14, 1575-1583.
4. Neu HC. (1992) Urinary tract infections. *Am. J. Med.* 92: 63-70.
5. Tambekar DH, Dhanorkar DV, Gulhane SR, and Dudhane MN. (2008). Prevalence And Antimicrobial Susceptibility Pattern Of Methicillin Resistant *Staphylococcus aureus*, From Healthcare And Community Associated Sources. *African Journal of Infectious Diseases* 1.
6. Mathai D, Jones RN, Pfaller MA. (2001) Epidemiology and frequency of resistance among pathogens causing urinary tract infection in 1,510 hospitalized patients: a report from the SENTRY Antimicrobial Surveillance Program (North America). *Diag Microbiol Infect Dis.* 40: 129-136.
7. Blondeau JM, Tillotson GS. (1999) Formula to help select rational antimicrobial therapy (FRAT): its application to community and hospital-acquired urinary tract infections. *Int. J. Antimicrob. Agents.* 12:145-150.
8. Cappuccino JG, Sherman N. (1996) *Microbiology, A Laboratory Manual*; 4th Ed. pp-186. The Benjamin-Cumming Publishing Company, Inc, Menlo Park, California.
9. Cernohorska L, Votava M, Determination of minimal regrowth concentration in clinical isolates of various biofilm-forming bacteria. *Folia Microbiol.* 2004; 49(1), 75-78.
10. Eliana BMG, Berezin EN, Nigro S, Nataly AS, Benini V, Toporovski J: Antibiotic resistance patterns of pediatric community-acquired urinary tract

- infections. *Braz J Infect Dis* 2008, 12(4):321–323.
11. Gul N, Mujahid TY and Ahmed S. 2004. Isolation, identification and antibiotic resistance profile of indigenous bacterial isolates from urinary tract infection patients. *Pak. J. Biol. Sci.*, 7: 736-738.
 12. Shareef SA. and Yagoub SO, 2006. Incidence and isolation of bacteria associated with nosocomial Urinary Tract Infection (UTI) in sudanese women. *Res. J. Microbiol.*, 1: 534-539.
 13. Rafique SM, Arifa Q, Mazhar and Ali Abbas Q, 2002. Prevalence patterns of community-based and nosocomial urinary tract infection caused by *Escherichia coli*. *Pak. J. Biol. Sci.*, 5: 494-496.
 14. Nahar SJ, Khanum H and Shimasaki K, 2010. Occurrence of *Escherichia coli* infection among the women of Khaka City. *ARPJ. Agric. Biol. Sci.*, 5: 68-73.
 15. Vromen M, van der Ven AJ, Knols AM, Stobberingh EE. (1999) Antimicrobial resistance patterns in urinary tract isolates from nursing home residents. Fifteen years of data reviewed. *J. Antimicrob. Chemother.* 44: 113-116.
 16. Lau SM, Peng MY, Chang FY. (2004) Resistance rates to commonly used antimicrobials among pathogens of both bacterial and non-bacterial community-acquired urinary tract infection. *J. Microbiol. Immunol. Infect.* 37: 185-191.
 17. Colodner R, Keness Y, Chazan B, Raz R. (2001) Antimicrobial susceptibility of community-acquired uropathogens in Northern Israel. *Int. J. Antimicrob. Agents.* 18:189-192.
 18. Hillier SL, Magee JT, Howard AJ, Palmer SR. (2002) How strong is the evidence that antibiotic use is risk factor for antibiotic-resistant, community -acquired urinary tract infection? *J. Antimicrob. Chemother.* 50:241–247.