Original Article

The Culture and Sensitivity Pattern of Urinary Tract Infections in Females of Reproductive Age Group

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Objective: To determine microorganisms responsible for urinary tract infection in females of reproductive age group and their antibiotic susceptibility pattern.

Place & Duration of Study: The study was conducted Sharif Medical City Hospital, Lahore, from June 2009 - June 2010.

Study Design: Retrospective study

Materials and Methods: A total of 181 midstream urine specimens were collected in sterile containers from suspected urinary tract infected cases of women (reproductive age group) from outdoor and indoor female patients (n=181). The patients included were between the ages of 15-50 years who came through OPD or emergency and whose routine urine examination revealed numerous pus cells on microscopy. These samples were collected from June 2009 to June 2010.

Results: 70 specimens were found to be culture positive. The predominant specie was *Escherichia coli.* Other isolates were *Klebsiella pneumoniae, Streptococcus faecalis, Staphylococcus aureus, Proteus vulgaris* and *Acinetobacter.* Antimicrobial sensitivity pattern of *Escherichia coli* was also done by Kirby Bauer Disc Diffusion method using CLSI Standard Guide Lines 2010, which revealed different patterns.

Conclusion: Most common urinary tract pathogen in females (reproductive age group) is *E. coli* and amikacin is the most effective drug. It is suggested that females should be regularly checked and examined by visiting the clinicians. They should be promptly treated after the proper investigations.

Key Words: UTI, antimicrobial susceptibility, pathogens, Cysteine Lactose Electrolyte Deficient (CLED) agar.

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Introduction

Urine in the human bladder is normally sterile. The presence of bacteria in the urine is called bacteriuria.[1] Bacteriuria may be asymptomatic or show apparent symptoms of urinary tract infection.[2] It is one of the most common infections occurring in all the age groups from neonates to old age. It is more common in females as compared to males, especially females of reproductive age group (from 15-50 years).[3] This is due to anatomical predisposition, close approximation of urethra and vagina and sexually active life during these years.[4] Bacteria normally present in the colon and hence on the perineum, may enter the urethral opening from the skin around the anus and genitals. Women may be more susceptible to UTI because their urethral opening is near the source of bacteria (e.g., anus, vagina) and their urethra is shorter, providing bacteria easier access to the bladder. 50-80% women experience urinary tract infection at least once or twice in their lives.[5] UTI is also an important complication of pregnancy. When it is associated with any structural and neurological deficit of the urinary tract often leads to death.[6] The rate of urinary tract infection usually increases during pregnancy particularly due to the pressure of gravid uterus on the ureters resulting in the stasis of urine flow and due to the humoral and immunological changes during normal pregnancy. The hormonal changes in pregnancy leads to decreased bladder tone, diminished peristalsis and dilatation of renal pelvis and ureter. Women who use a diaphragm develop infections more often, and condoms with spermicidal foam may cause the growth of *E. coli* in the vagina, which may enter the urethra. UTIs are most commonly caused by

Escherichia coli.[7] Other gram negative bacteria are *Klebsiella* spp*, Enterobacter* spp*.,* and *Pseudomonas aeruginosa, Proteus* spp. Gram positive organisms include *Enterococcus* spp*, Staphylococci* and *Streptococci* and account for 5 to 15 % of the cases.[8] Despite of the fact that wide range of antibiotics is available against UTI, it remains one of the most common infections and is responsible for significant morbidity in females. The quality of life is affected in women with UTI and may have serious consequences of developing renal damage. This warrants greater attention for reproductive health needs of the women, and health education for hygienic/ safe use of family planning methods.

Resistance of antibiotics is yet another serious problem. This is due to overuse as well as misuse of antibiotics that resistance of antibiotics is increasing day by day.[9] The micro organisms responsible for the UTIs are mostly bacteria. The treatment of choice for complicated UTI is antibiotics. The antibiotics commonly used to treat UTIs are broad spectrum cephalosporins, floroquinolones and aminoglycosides. Cephalosporins are cell wall inhibitors and are used commonly for treating infections caused by gram negative organisms. These include cephradine, cefaclor, cefotaxime and ceftazidime. Floroquinolones act by inhibiting the activity of DNA gyrase and topoisomerase, enzymes essential for the DNA replication and include ciprofloxacin, ofloxacin, enoxacin and sparfloxacin. The aminoglycosides include gentamicin, kanamycin, and amikacin. They inhibit the bacterial protein synthesis.[10,11] The choice of antimicrobials should depend on the micro organism isolated and its antibiotic susceptibility pattern to certain antibiotics. Hence, the knowledge on the frequency of the causative organisms and their sensitivity pattern requires reappraisal from time to time for the effective treatment. This retrospective study reports the pattern of uropathogens, isolation of *E. coli* and its antibiotic sensitivity pattern in females of reproductive age group in a tertiary care hospital.

Materials and Methods

Bacterial isolates: The study was conducted at the Microbiology Department, Sharif Medical City, Lahore**.** A total of 181 midstream urine specimens were collected in sterile containers from suspected urinary tract infected cases of women (reproductive age group) from outdoor and indoor female patients (n=181). The patients included were between the ages of 15-50 years who came through OPD or emergency and whose routine urine examination revealed numerous pus cells on microscopy. These samples were collected from June 2009 to June 2010. The specimens were processed immediately in the laboratory for physical and chemical examination, microscopy for cells and culture of the specimen. With standard calibrated loop, 1µl of urine was inoculated on Cysteine Lactose Electrolyte Deficient (CLED) agar and blood agar and incubated aerobically at 37^0C for 18-24hrs, within 30 minutes of collection. Inoculation from well-mixed specimen was performed first, followed by other procedures. After the incubation, if the CFU was more than 10^5, it was considered significant bacteriuria. Such urine samples were further processed for identification and antibiotic susceptibility pattern was determined. If the CFU was less than 10^5 it was considered as negative for culture or non significant bacteriuria. Mixed growth of two or more organisms especially with gram positive bacilli, *Lactobacilli, Gardnerella vaginalis*, diphtherias were considered to be urinary contamination. Repetion of urine examination was advised with early morning fresh urine specimen.[2]

Identification of uropathogens: Identification of the isolated bacterial pathogens was done on the basis of gram staining, morphology and biochemical characters. [TSI agar, indole reaction, citrate, urease, MR, VP and motility agar (Oxoid B D)].

Antimicrobial Susceptibility Testing: For the positive cultures, antibiotic sensitivity discs were put on the Muller Hinton agar plates. The plates were incubated at 37^0C for 18-24 hours. The results of sensitivity plates were read after 24 hours. Negative cultures were reincubated for another 24 hours and report was given as no growth at the end of 48 hours of incubation.[2] Antimicrobial sensitivity of the isolated pathogens was determined by using Kirby Bauer Disc Diffusion method according to CLSI Clinical and Laboratory Standards Institute 2010).[12,13] The antibiotics tested were amikacin, nitrofurantoin, co-trimoxazole, imipenam, cefipime, cephalothin, cefotaxime, cefalexin, ceftazidime, levofloxacin, tobramycin, norfloxacin, cefuroxime and pipedemic acid.

Results

A total 181 urine samples were analyzed for isolation and identification of bacterial isolates. Out of which 70 (38.6%) samples were found to have significant bacteriuria and remaining 111 samples were found to have either non significant bacteriuria or very low bacterial count or sterile urine. In the present study, out of 70 isolated pathogens the most common isolate was *Escherichia coli* (70%), followed by *Klebsiella pneumonia* (14%), *Streptococcus faecalis* (5.7%), *Acinetobacter* (4.2%), *Staphylococcus aureus* (2.8%), *Candida* (1.4%), *Pseudomonas* (1.4%) and *Proteus* (1.4%). (Table I)

Antimicrobial sensitivity testing of the *Escherichia coli* was done by using Kirby Bauer Disc Diffusion method, according CLSI Clinical and Laboratory Standards Institute 2010). [13-15] At the end of incubation period, the diameter of the zones of inhibition around each disc was measured with vernier calipers on the back of plate, with reflected light against a dark non-reflected background. Fourteen antibiotics commonly used were tested against the isolated *E. coli*. Amikacin was found to be the most effective drug (97.61%) followed by nitrofurantoin (90%). (Table II)

Table I: Distribution of isolated urinary tract pathogens, their percentage (n=70).

S. no	Organism isolated	Total	Prevalence %age
1.	*Escherichia coli*	48	70
2.	*Klebsiella pneumoniae*	10	14
3.	*Streptococcus faecalis*	4	5.7
4.	*Acinetobacter*	3	4.2
5.	*Staphylococcus aureus*	2	2.8
6.	*Candida*	1	1.4
7.	*Pseudomonas aeruginosa*	1	1.4
8.	*Proteus*	1	1.4

Table II: Antibiotics sensitivity pattern of urinary tract E. coli (n=48) used in the study

Antibiotics	Sensitivity (%)
Amikacin	97.61
Nitrofurantoin	90
Co-trimoxazole	81
Imipenam	75
Cefipime	66.66
Cephalothin	52.38
Cefotaxime	48
Cefalexin	47.05
Ceftazidime	46.66
levofloxacin	37.14
Tobramycin	37.03
Norfloxacin	34
Cefuroxime	3.12
Pipedemic acid	2

Discussion

Urinary tract infection is one of the most common types of infectious diseases encountered in the practice of medicine these days. A total of 181 urine specimens of females aged 15-50 years, suspected for urinary tract infection were processed for culture. Seventy were found to be culture positive. According to our study E. coli was the commonest cause of urinary tract infection (70%) followed by Klebsiella pneumoniae (14%), Streptococcus faecalis (5.7%), Acinetobacter (4.2%), Staphylococcus aureus (2.8%), Candida (1.4%), Pseudomonas aeruginosa (1.4%) and Proteus (1.4%). It was different from other studies which reported E. coli 24% [3] and 66%, [10] the most frequent uropathogen. This was similar to other studies where it was the most frequent pathogen causing UTI, as in a study, 2010 Mohammad MT where 62.6% cultures grew E coli. [11] and 66% E. coli in a study, 2010 Mohammad Naeem et al in Islamabad.[10] These results were also similar with a study conducted by Dilnawaz S et al 2005, which reflects that first two common organisms were E coli and Klebsiella pneumoniae. Third prevalent organism in our study was Streptococcus faecalis while in the above mentioned study it was Pseudomonas.[12]

In the context of antibiotic sensitivity, our results were comparable with a study by Mohammad MT et al 2010, which revealed that imipenem (94.20%) was most effective drug against urinary E coli followed by amikacin (93.11%).[11] We found that amikacin (97.61%) was most effective antibacterial against urinary E. coli followed by nitrofurantoin (90%) and co-trimoxazole (81%) and then imepenem (75%). According to our results ceftazidime was 46.66% sensitive, cefotaxime was 48% sensitive and pipedemic acid was only 2% effective. According to above mentioned study, ceftazidime was 90.57% effective, cefotaxime was 19.20% and pipedemic acid was 13.40% effective.[11]

The sensitivity of E. coli to cefipime, cephalothin, cephalexin and ceftazidime was found to be 66.66%, 52.38%, 47.05% and 46.66% respectively. A possible reason for this might be presence of extended spectrum beta lactamases (ESBL) in these strains. This problem is increasing day by day in our community.

Another study by Kebira et al in Kenya in 2009 revealed that amikacin and ofloxacin, were 100% sensitive to E. coli isolated from urine specimens, nitrofurantoin is 77% susceptible while norfloxacin, ciprofloxacin and ceftazidime were 95% sensitive.[3]

In a study by Uwaezuoke in Nigeria in 2006, nitrofurantoin and gentamicin were found to be 80% effective against strains of *E. coli.* [6]

Antibiotics are still effective as treatment of urinary tract infections.[16]

The selection of antibiotic against any urinary tract pathogen depends on the antibiotic resistance pattern, its pharmacokinetic properties, dose quantity and timings, its effect on gastrointestinal tract and vaginal flora allergies and adverse effects caused by that drug.[7]

Antimicrobial resistance is a big problem cause of great concern through out the world. Knowledge of the antibacterial resistance among uropathogens is essential to provide appropriate cost effective therapy.

Conclusion

Most common urinary tract pathogen in females (reproductive age group) is E. coli and amikacin is the most effective drug. It is suggested that females should be regularly checked and examined by visiting the clinicians. They should be promptly treated after the proper investigations.

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