Original article

Bacteriological Profile and Antibiotic Sensitivity Pattern in Patients Attending Out Patient Departments with Urinary Tract Infections.

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Abstract

Introduction: Urinary tract infections (UTI) are the most common cause of bacterial infections in humans. A variety of organisms are associated with UTI irrespective of whether they are community or hospital acquired. **Aim and Objective:** The aim of the study was to determine the bacteriological profile and antibiotic sensitivity pattern of organisms causing UTI in patients attending OPD. **Materials and Methods:** This study was conducted in the Department of Microbiology, Sri Siddhartha Medical College, Tumakuru from Jan 2017 - Mar 2017. All suspected cases of UTI sent for urine culture were tested, bacteriological profile and antibiotic sensitivity pattern was evaluated. **Results:** A total of 392 clinically suspected cases of UTI of all ages and both sexes were studied. Among the 392 samples, 154 samples yielded significant growth, among these110 (71.42%) were of females and 44 (28.57%) were of males. Culture positive cases in the age group 21-40 years (52.59%) were the highest. *Escherichia coli* (29.22%) was the most common organism isolated followed by *Klebsiella* sps (18.18%) among gram negative bacilli and *Staphylococcus aureus* (10.38%) among gram positive cocci. *Escherichia coli* was 100% sensitive to nitrofurantoin, amikacin and imipenem. The isolation of Extended Spectrum Beta Lactamase (ESBL) among gram negative bacterial isolates was 2.5%. *Staphylococcus aureus* was sensitive to vancomycin and linezolid (100%)and all the isolates were Methicillin Sensitive Staphylococcus aureus (MSSA) **Conclusion:** UTI is most common in females and *E.coli* was the most common uropathogen isolated. Culture results and antibiogram help in the specific treatment and judicious antibiotic usage to prevent drug resistance.

Key words: Antibiotic sensitivity pattern, culture, significant bacteriuria, urinary tract infections.

Introduction

Urinary tract infections (UTI) are one of the most common bacterial infections and is the most important cause of morbidity in the general population and is the second most common cause of hospital visits.[1]

The prevalence of UTI is greater in women than men due to the anatomical predisposition or large bacterial load in the urethral mucosa or other factors like obstruction in urinary tract, sexual activity and pregnancy.[2]

Microbial invasion can occur in any part of urinary system. The severity of infection may range from asymptomatic colonization to symptomatic invasion of the urinary system.[3]

UTI is defined as the presence of more than 10^5 colony forming units (CFU) of bacteria per ml of urine for asymptomatic individual and much lower for symptomatic individual (103CFU/ml). Samples obtained by suprapubic aspiration or in-and-out catheterization and samples from patients with in-dwelling catheters, colony count of 10^2-10^3 indicates infection.[4]

Escherichia coli is the most frequently isolated organism in acute infections. *Proteus* sps, *Klebsiella* sps, *Enterobacter* sps and *Pseudomonas* sps, *Staphylococcus spp* and *Enterococcus sps* are other common organisms often isolated.[5]

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Poor patient compliance and incomplete antibiotic therapy have led to the evolution of drug resistance to antimicrobials used for the treatment of UTI.[6]

The aim of the study was to determine the bacteriological profile and antibiotic sensitivity pattern of organisms in patients with UTI attending OPDs.

Materials and methods

This cross-sectional study was carried out in the Department of Microbiology, Sri Siddhartha Medical College, Tumakuru, from Jan 2017 to Mar 2017. Informed written consent was taken from all patients. Patients who have not been on antibiotic therapy for the past 15 days were included in the study. All patients on antibiotic therapy and who did not fill the consent form were excluded. A total of 392 clinically suspected cases of UTI attending out patient department with symptoms of UTI such as fever, dysuria, increased frequency of micturition & loin pain were subjected to urine culture. Parameters like age, sex and associated diseases of patients were noted. Mid-Stream Urine (MSU) was collected in sterile wide mouth container. A calibrated sterile inoculating wire loop for the semi-quantitative method was used for the plating which has a 4.0 mm diameter designed to deliver 0.01 ml of urine. A loopful of the well mixed urine sample was inoculated into duplicate plates of Blood and Mac-Conkey agar. All plates were then incubated at 37ºC aerobically for 24 hrs. The plates were then examined macroscopically and microscopically for bacterial growth. Bacterial colonies were counted and multiplied by 100 to give an estimate of the number of bacteria per ml of urine. Growth of more than 10^5 bacterial colonies per ml of urine sample in untreated cases was considered as significant bacteruria. Isolated colonies were identified by colony morphology, gram stain, oxidase test, catalase test and standard biochemical tests.[7] Antibiotic sensitivity testing was done as per CLSI guidelines by Kirby-Bauer disc diffusion using Muller Hinton Agar (MHA).[8]

Methicillin resistant Staphylococcus aureus Detection (MRSA):

Cefoxitin disc diffusion tests were performed for all Staphylococcus aureus isolates as recommended by CLSI guidelines 2018. A suspension of each isolate was made so that the turbidity was equal to a 0.5 McFarland standard and then plated onto Mueller–Hinton agar. A 30 µg cefoxitin disc was applied to each plate. After incubation at 35±1 °C for 24 hours zone sizes were measured. Isolates showing inhibition zone size ≤19 mm were considered as Methicillin resistant Staphylococcus aureus (MRSA). Standard strains used were ATCC 33591 (MRSA) and ATCC 29213 (MSSA).[8]

Extended spectrum beta lactamase Detection (ESBL):

The screening for extended spectrum beta lactamase (ESBL) was done using cefpodoxime (≤ 17mm), ceftazidime (≤ 22mm), aztreonam (≤ 27mm), cefotaxime (≤ 27mm) and ceftriaxone (≤ 25mm) If the organisms showed a zone of inhibition lower than the minimum for any antibiotic disc, ESBL was suspected. The phenotypic confirmation was done by testing the strain against ceftazidime and ceftazidime/clavulanic acid. A zone of inhibition of ≥ 5mm diameter for ceftazidime/ clavulanic acid in comparison to ceftazidime was indicative of ESBL production. *Escherichia coli* ATCC 25922 & *Klebsiella pneumoniae* ATCC 7006003 were used as negative and positive controls respectively.[8]

Statistical analysis used was descriptive statistics: All data were entered in Microsoft Excel and analyzed.

Results

A total of 392 clinically suspected cases of UTI attending OPD of all ages and both sexes were studied between January 2017 to March 2017. Among the 392 samples,154 (39.28%) samples yielded growth. Of the

Table 1: Age and sex distribution of culture positive cases.

Sl. No.	Age Group	Male (%)	Female (%)	Total
1	upto-20	02 (4.54)	07 (6.36)	**09 (5.84%)**
2	21-40	26 (59.09)	55 (50.00)	**81 (52.59%)**
3	41-60	12 (27.27)	38 (34.54)	**50 (32.46%)**
4	>60	04 (9.09)	10 (9.09)	**14 (9.09%)**
Total		**44 (28.57)**	**110 (71.42)**	**154 (100%)**

154 culture positive cases 110 (71.42%) were of females and 44 (28.57%) were of males. (Table 1)

The study shows higher number of culture positive cases among patients in age group between 21-40 years (52.59%) followed by age group 41-60 years (32.46%).

Distribution of bacterial agents causing UTI are shown in Table 2. *Escherichia coli* (29.22%) was the most common organism isolated followed by *Klebsiella* sps (18.18%), *Pseudomonas aeruginosa* (10.38%), *Proteus* sps (9.07%) and *Citrobacter* sps (8.44%) among gram negative bacilli and *Staphylococcus aureus* (10.38%), *Enterococci* (7.97%) and CONS (6.49%) among gram positive cocci.

Antibiotic sensitivity pattern of the gram negative bacillary isolates grown in culture is shown in table 3. *Escherichia coli* were highly sensitive to nitrofurantoin, amikacin and imipenem (100%), resistance was seen to ampicillin, and other antibiotics. *Klebsiella* sps also had the similar type of sensitivity pattern to ciprofloxacin, norfloxacin (89%) and nitrofurantoin (79%).

Table 2: Distribution of Bacterial agents among culture positive cases.

Sl No	Organisms	Frequency	Percentage
1	*Escherichia coli*	45	29.22%
2	*Klebsiella* sps.	28	18.18%
3	*Pseudomonas aeruginosa*	16	10.38%
4	*Staphylococcus aureus*	16	10.38%
5.	*Proteus* sps	14	9.07%
6	*Citrobacter* sps	13	8.44%
7	*Enterococcus sps.*	12	7.97%
8	CONS	10	6.49%
	TOTAL	**154**	**100**

Pseudomonas sps was sensitive to amikacin (75%), Imipenem and ciprofloxacin (81%), *Proteus* sps was sensitive to nitrofurantoin (100%), *Citrobacter* sps was 100% sensitive to imipenem, cefotaxime and ceftazidime.

Table 3: Antibiotic sensitivity pattern (%) of Gram Negative Bacilli

	E.coli (%)	Klebsiella sps (%)	Pseudomonas sps (%)	Proteus sps (%)	Citrobacter (%)
Ampicillin	03	04	00	29	38
Ciprofloxacin	95	89	81	64	85
Norfloxacin	95	89	88	50	69
Co-trimoxazole	88	80	00	79	77
Piperacillin-tazobactam	84	61	75	86	77
Nitrofurantoin	100	79	63	100	77
Gentamycin	68	75	63	71	85
Amikacin	100	71	75	71	85
Imipenem	100	80	81	93	100
Cefotaxime	98	80	75	93	100
Ceftazidime	73	80	75	86	100

Table 4: Antibiotic sensitivity pattern of Gram Positive Cocci

Sl. No.	Organism	A (%)	Cf (%)	Cx (%)	Co (%)	Nf (%)	G/HlG (%)	Amoxy (%)	Le (%)	Va (%)
1.	*Staphylococcus aureus* (16)	10 (63)	07 (44)	16 (100)	10 (63)	14 (88)	07 (44)	07 (44)	16 (100)	16 (100)
2	CONS (12)	10 (83)	09 (75)	12 (100)	07 (58)	12 (100)	07 (58)	10 (83)	12 (100)	12 (100)
3	*Enterococci* (10)	07 (70)	07 (70)	00 (00)	05 (50)	08 (80)	10 (100)	05 (50)	10 (100)	08 (80)

A-Ampicillin, Cf-Ciproflocaxin, Cx- Cefoxitin, Co- co-trimoxazole, Nf- Nitrofurantoin, G- Gentamycin, HIG-High level gentamycin, Amoxy-Amoxicillin-clavulanic acid, Le-Linezolid, Va-vancomycin.

Out of the 116 gram negative bacilli isolated 3 (2.5%) were ESBL producers. Two strains of *Escherichia coli* (4.44%) and one strain of *Klebsiella sps* (3.54%) were ESBL producers.

Among the gram-positive isolates *Staphylococcus aureus* was sensitive to vancomycin and linezolid (100%), norfloxacin (88%), ampicillin (63%), ciprofloxacin (44%). *Enterococci* were sensitive to gentamycin, linezolid (100%), norfloxacin and vancomycin (80%). All the *Staphylococcus aureus* isolated were sensitive to cefoxitin and were Methicillin Sensitive Staphylococcus Aureus (MSSA).

Discussion

The burden of urinary tract infections is on the rise in health care settings as well as community settings. It is caused by various types of pathogens including *Escherichia coli, Klebsiella sps* and *Pseudomonas* sps. Identification of the type of causative agent is helpful in effective management of patients suffering from bacterial UTI. In community and hospital settings the etiology and antimicrobial susceptibility to bacteria causing UTI has been changing over the years. Choice of treatment of UTI has changed from co-trimoxazole to quinolones over the last decade.[9]

In the present study, 392 clinically suspected cases of UTI of all age groups and from both sex attending various OPD departments of Sri Siddhartha Medical College and Hospital, were studied. Among the total 154 positive urine cultures analyzed, the prevalence of UTI was 39.28%. Other studies show a higher prevalence rate, which may be because of the short duration of study and very small sample size .[6,9,10]

Females were more commonly affected as compared to males this can be attributed to short female urethra. Bacteria from faecal matter can be easily transferred to the vagina or the urethra in ascending fashion. Frequency of recent sexual activity is the most important risk factor in young women. Biological changes due to menopause put elder women at particular risk for primary and recurrent UTI. Other factors like lifestyle, hormonal, genetic susceptibility poses more risk for females. Men commonly suffer from UTI after the age of 50 yrs when they begin to develop prostate problems. Similar observations have been made in many studies.[6,9,10]

Significant growth was seen in 154 cases. Collection of specimen before antibiotic therapy helps in good isolation of the infecting microorganism. Distribution of bacterial isolates in UTI varies from time to time and place to place. The commonest organism isolated was *Escherichia coli* followed by *Klebsiella* sps among gram negative bacilli and *Staphylococcus aureus* and *Enterococci* among gram positive cocci. Similar observations were made in many previous studies.[4, 6,11]

In our study, gram negative bacteria isolated showed good sensitivity to amikacin, nitrofurantoin, imipenem (100%) for *Escherichia coli, Klebsiella* sps and high degree of resistance to ampicillin and gentamycin.[11]

The percentage of ESBL among gram negative bacterial isolates was 2.5% which was contrary to higher isolation of ESBL in some studies.[2,11]

Gram positive cocci were sensitive to linezolid and vancomycin (100%). The results of the study are in concordance when compared with other studies.[11]

Conclusion

UTI is one of the most important causes of morbidity in the general population and is the second most common cause of hospital visits. The recognition of UTI, proper diagnosis with urine culture and starting appropriate antibiotics according to culture report plays a major role in preventing a complicated UTI. In present study, females were mostly affected and the most commonly isolated organism was *Escherichia coli* and *Klebsiella* sps. The isolated organisms were 100% sensitive to amikacin and nitrofurantoin. The isolates were also sensitive to piperacillin-tazobactam and imipenem which can be reserved as second line drug.

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