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Antimicrobial Susceptibility Patterns of Uropathogens in Different Ages and Genders in and Around Namakkal District, Tamilnadu

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Abstract: Infections are one of the main causes of morbidity and mortality in patients with urinary tract infection. Urinary tract infections (UTIs) are one of the most frequent bacterial infections in human beings. Most commonly, members of Enterobacteriaceae, particularly uropathogenic strains of *E. coli* and *Enterobacter* spp. are the primary causative organisms of UTIs in different parts of the world. It is very difficult to eradicate the urinary tract infection completely and is a challenge to the medical professionals. A sample total of 115 patients with suspected urinary tract infection were screened for the study. Urine samples were collected from private and government hospitals in and around Erode, Tamil nadu, India. Urine cultures were carried out and the isolates were identified by Gram staining and conventional biochemical methods. Antimicrobial susceptibility testing was performed by disk diffusion method according to the current National Committee for Clinical Laboratory Standards (NCCLS) guidelines. In this present study, among both sexes tested, incidence was higher in female patients (46.75%) then male patients (13.15%). The rate of infection in females was high (46.75%) and in that only a particular age group (21-30) was more prone to infections. The predominant urinary pathogens isolated from this study were *Escherichia coli* (21.95%), *Citrobacter spp.* (14.63%), *Enterobacter spp*. & *Klebsiella spp*. (12.19%), *Pseudomonas aeruginosa* and *Proteus spp.* (9.75%), *Staphylococcus aureus* (17.07%) *and Salmonella species* **(**2.43%). The incidence of Gram negative organisms was 82.9% and Gram positive organisms, 17%. Among the drugs used for testing the antimicrobial susceptibility, Amikacin, Gentamycin, Norfloxacin, and Nitrofurantoin were found to be effective in the treatment of urinary tract infection. Improvement in overall sanitary condition, proper knowledge on personal hygiene would play an important role in reducing the incidence and occurrence of urinary tract infection. Prompt referring of urine samples for microbial analysis and antibiotic sensitivity profile would make therapy more effective and reduce recurrence rate to a greater extent.

Key words: Uropathogenic · Antibiotic sensitivity Enterobacteriaceae Urinary tract infections microbial analysis

INTRODUCTION

Urinary tract infection (UTI) is the commonest and most serious disease seen in human beings. Urinary tract infection is common in females than in males because female urethra appears to be less effective in preventing the entry of bacteria. Frequency and/or urgency confirmed by the presence of bacteriuria have been seen in adult non pregnant women with apparently normal urinary tracts [1]. UTI with increased risk include infants, pregnant women and the elderly, as well as those with indwelling catheters, diabetes and underlying urologic abnormalities [2]. Among various pathogens the most common invaders are the Gram negative bacteria especially *Escherichia coli, Citrobacter species, Enterobacter aerogenes, Pseudomonas aeruginosa,* and *Proteus vulgaris,* those which are rarely found include: *Klebsiella* species, *Staphylococcus aureus* and *Salmonella* species [3].

UTIs are the leading cause of Gram-negative bacteremia in patients of all ages and are associated with a high risk of morbidity and mortality, especially in the

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elderly, and account for significant health care costs [3, 4]. Several studies has demonstrated that the geographical variability of pathogen occurrence in cases of UTI among inpatients and outpatients populations is limited by the predominance of Gram-negative species usually Enterobacteriaceae and particularly *E. coli* and *Enterobacter* spp. in various regions of the world [5, 6]. It is necessary to identify the causative agent and spectrum of its antimicrobial susceptibilities in order to treat UTI. Since this spectrum may vary among geographical locations, hospitals and also in different age groups, each institution should carefully plan its antibiotic therapy. This study was performed to find out the frequency of different urinary tract pathogens in different age groups of both sexes and the antimicrobial susceptibility patterns.

MATERIALS AND METHODS

A total of 115 patients with suspected urinary tract infection were screened for the study. Urine samples were collected from private and government hospitals with the help of trained nursing staff. For collection early morning urine samples (mid stream) was collected and transported to the laboratory in an ice pack and analyzed within 6 hours. Following techniques were carried out for further investigation [7, 8]. Culture was done by the calibrated loop technique delivering 0.001 mL of urine and plated on Cystine-Lactose- Electrolyte Deficient (CLED) agar, MacConkey agar and Blood agar medium (Hi Media, India). The inoculated plates were incubated at 37°C for 24 h and for 48 h in negative cases. A specimen was considered positive for UTI if a single organism was cultured at a concentration of $\geq 10^5$ cfu mL^{-1}, or when a single organism was cultured at a concentration of 10^4 cfu mL^{-1} and ≥ 5 pus cell per high-power field were observed on microscopic examination of the urine [9] . Bacterial identification was based on standard culture and biochemical characteristics of isolates. Gram-negative bacteria were identified by the standard biochemical tests [2, 10]. Gram-positive microorganisms were identified with the corresponding laboratory tests: catalase, coagulase, and mannitol test for *Staphylococcus aureus* [11]. Antimicrobial susceptibility of isolates was tested by the Kirby Bauer disc diffusion method [12] according to the Clinical and Laboratory Standard Institute guidelines [13] using the following antibiotics: Amikacin, Gentamicin, Co-trimoxazole, Ampicillin, Trimethoprim, Norfloxacin, Cephalothin, Nitrofurantion and Vancomycin. Data management and statistical analysis were performed using SPSS software.

Table 1: Prevalence of UTI in the different age groups

S.NO	Age Group (Years)	UTI Positive	UTI Negative
1	1-10 (n=22)	0(0%)	22(100%)
2	11-20 (n=27)	04(14.8%)	23(85.2%)
3	21-30 (n=49)	32(65.3%)	17(34.7)
4	31-40 (n=16))	5(31.3)	11(68.7%)
5	41-50 (n=01)	0(0%)	1(100%)
Total (n=115)	41 (35.65%)	74 (64.35%)	

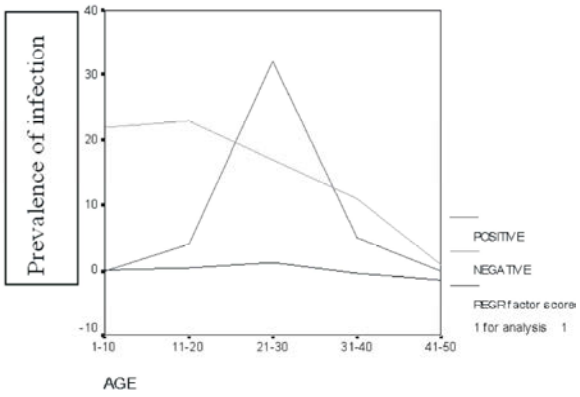

Fig. 1 : Total Survey PLOT

RESULTS

A sample total of 115 members were screened for the presence of UTI infection. The age of the members under consideration ranged between 1-50 year and the distribution was shown in Table 1 and Figure 1.

The detailed analysis of the relationship of bacterial infection with the sex and age of the patients (Table 2) was illustrated. The incidence was found to be more in female especially in the age group of 21-30 than the male patients. A total of 38 male suspected UTI samples, 5 samples revealed positive culture of pathogens. The percentage of incidence was 13.15%. Whereas in female out of 77 samples 36 were positive for culture and the percentage of incidence was 46.75%.

Eight different types of pathogens were isolated from the urine samples and their distribution in patients of different sex was shown (Table 3). Overall, more than 82.93% of isolates were Gram negative organisms. *E. coli* was the most frequently isolated uropathogen (21.95%), followed by *Citrobacter* spp., (14.63%) and least isolated was *Salmonella* spp., (2.43%) (Table 3). Percentage of Gram negative organisms was 82.9% and Gram positive organisms, 17.1%.

Nine antibiotics were tested against the isolated uropathogens among which norfloxacin, amikacin, gentamycin and nitrofurantoin were more effective than other antibiotics (Table 4). *E. coli* as the predominant

Table 2: Percentage of UTI in the different age groups of both male and female

S.No	Age Group	Male			Female			Total No. of male & female	Infected male & female	UTI percentage
		Total No. N=38	Infected No.	Percentage	Total No. N=77	Infected No.	Percentage			
1	1-10	08	0	0	14	0	0	22	0	0
2	11-20	10	0	0	17	04	23.53	27	04	14.81
3	21-30	06	02	33.33	43	30	69.77	49	32	65.31
4	31-40	13	03	23.07	03	02	66.66	16	05	31.25
5	41-50	01	0	0	0	0	0	01	0	0
Total	38	05	13.15	77	36	46.75	115	41	35.65	

Table 3: Incidence of uropathogens in different sex

S.No	Organisms	Total No. Of Isolates		Total No. of Organisms	Percentage of Organisms
		Male	Female		
1	*Escherichia coli*	02	07	09	21.95
2	*Klebsiella. Sp.*	0	05	05	12.19
3	*Pseudomonas sp.*	0	04	04	09.75
4	*Enterobacter Sp*	01	04	05	12.19
5	*Citrobacter Sp*	03	03	06	14.63
6	*Proteus Sp*	01	03	04	09.75
7	*Salmonella Sp*	0	01	01	02.43
8	*S. aureus*	0	07	07	17.07
Total		07	34	41	---

Table 4: Antimicrobial susceptibility rates of tested uropathogens

S. No.	*E. coli* (n=9)	*Klebsiella spp.(n=5)*	*Pseudomonas spp.,* (n=4)	*Enterobacter spp.,* (n=5)	*Citrobacter spp.,* (n=6)	*Proteus spp.,* (n=4)	*Salmonella spp.,* (n=1)	*Staphylococcus aureus* (n=7)
Amikacin (30µg)	8 (88.88%)	4 (80%)	4 (100%)	1 (20%)	4 (66.66%)	3 (75%)	1 (100%)	5 (71.43%)
Gentamycin (10µg)	8 (88.88%)	4 (80%)	4 (100%)	2 (40%)	3 (50%)	2 (50%)	1 (100%)	3 (42.86%)
Cotrimaxazole (10µg)	0	0	0	0	0	1 (25%)	0	0
Ampicillin (10µg)	0	0	0	0	0	3 (75%)	0	0
Trimethoprim (10µg)	9 (100%)	3 (60%)	1 (25%)	0	1 (16.66%)	4 (100%)	0	0
Norfloxin (10µg)	5 (55.55%)	2 (40%)	3 (75%)	1 (20%)	5 (83.33%)	3 (75%)	1 (100%)	6 (85.71%)
Cepalothin (30µg)	6 (66.66%)	1 (20%)	0	0	o	1 (25%)	1 (100%)	0
Nitrofurantoin (300µg)	8 (88.88%)	0	3 (75%)	2 (40%)	2 (33.33%)	2 (50%)	1 (100%)	0
Vancomyvin (30µg)	5 (55.55%)	0	0	3 (60%)	0	1 (25%)	0	4 (57.14)

cause of UTI, showed the highest percentage of resistance to ampicillin and co-trimoxazole (100) and the lowest resistance was to norfloxacin and vancomycin (55.55). *Citrobacter* spp., as the second most prevalent pathogen of UTI displayed a similar resistance pattern and was 100% resistant to ampicillin, Co-trimoxazole, vancomycin and cephalothin and susceptible to trimethoprim in 83.34% of cases. *P. aeruginosa* showed the highest antibiotic resistance rate and was significantly resistant to most of the antibiotics. *Citrobacter spp*., *Pseudomonas spp*., and *Enterobacter spp*. showed 100% resistance to Cotrimaxazole, ampicillin and cephalothin. In this study, *S. aureus* was responsible for about 17.07% of UTI cases and were resistant to nitrofurantion, ampicillin, trimethoprim, Co-trimoxazole and cephalothin in 100% (Table 4).

DISCUSSION

In spite of the availability and use of the antimicrobial drugs, UTIs caused by bacteria have been showing increasing trends in recent years. Much of the increase has been related to emerging antibiotic resistance in urinary tract pathogens [14]. The common uropathogens identified in adult patients with UTIs include enteric gram-negative bacteria, with *E. coli* being the most common. The remainders of infections are caused by coagulase-negative *Staphylococcus saprophyticus* (10-20%), while *Proteus mirabilis, Klebsiella*, and *Enterococcus* account for less than 5% [15-17]. Other aerobic Gram-negative bacteria of the Enterobacteriaceae family include *Citrobacter, Enterobacter, Serratia,* and *Salmonella* [18, 19].

In our study the incidence of infection was highest in the age group of 21-30 followed by 31-40. This finding correlates with the reports of earlier workers which include [20], Most UTIs are caused by bacteria and *E. coli* is the most common uropathogen detected in above 22% cases. *Citrobacter* spp. was isolated in 14.7% of cases and other occasional pathogens include *S. aureus*, *Salmonella* spp., *P. mirabilis* and *Klebsiella aerogens* (2- 12%). These results agree with Taneja *et al*. [21] who investigated a total of 1974 clean catch midstream urine samples from which significant bacteriuria was found in 558 samples (28.3%). Common uropathogens isolated were *Escherichia coli* (21.95%), *Klebsiella* spp. (12.19%), *Proteus spp.,*(9.75%), *P. aeruginosa* (9,75%) and *S. aureus* (17.07%) whereas Tambekar *et al*. [22] investigated a total of 174 urine samples from which, 68 are found to have significant bacteriuria with *E. coli* (59%), followed by *Pseudomonas* a*eruginosa* (15%), *K. pneumoniae* (10%), *P. mirabilis* (9%), *S.aureus* (6%) and *C. freundii* (1%). The urinary tract infections were found to occur most frequently in female (63%) than male (37%). On the other hand Amin *et al*. [23] reported that the urine cultures bacteriologically positive were found in 68 and 32% of the examined females and males subjects, respectively. Wazait *et al*. [24] reported that still there is a high incidence of *Escherichia coli* (47.30%) in urine samples. It is interesting to note that only few investigators have reported the presence and significance of *Citrobacter spp*. in UTI [25, 26].

Recurrence is an inevitable consequence of UTI. Frequent and abrupt emergence of drug resistant strains is attributed as a major reason for this [27]. In this present study, Amikacin Gentamycin and norfloxacin followed by Nitrofurantoin were found to be effective in the treatment of UTI. Treatment of gentamicin-resistant *Klebsiella* urinary tract infections with cephradine have been reported [28]. For some drugs like norfloxacin, though it has been used in the treatment of UTI, the isolates from urine samples were considered to be resistant to them. The increase in resistance may be due to improper treatment and indiscriminate use of antibiotics [29]. Recurrence of urinary tract infection under treatment has been reported [30]**.** Most of the isolates had exhibited resistance to common antibiotics like ampicillin, tetracycline and carbenicillin [31]. UTI recovered bacteria showed the highest degree of resistance to ampicillin, co-trimaxazole and cephalothin.

In conclusion, the present study showed that the incidence of urinary tract infection was high in the age group of 21- 30. Eight different uropathogens were identified among them *E. coli* was considered as dominant pathogen. The organisms showed resistance to older urinary antimicrobial agents such as ampicillin and cotrimoxazole; this indicates that increased consumption of a particular antibiotic can be a pathway to its resistance. Higher prevalence of UTIs was observed in female population. In this present study, Amikacin, Gentamycin and Norfloxacin were found to be effective in the treatment of UTI.

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