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Research Article

ISOLATION OF UROPATHOGENS AND THEIR ANTIBIOTIC SUSCEPTIBILITY PATTERN AT A TERTIARY CARE HOSPITAL IN NORTHERN INDIA

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ABSTRACT

Objective: The objective of the study was to know the isolation rate and antibiotic susceptibility pattern of pathogens causing urinary tract infection (UTI).

Methods: A total of 500 urine samples of clinically suspected UTI were collected from January 2019 to June 2019. The samples were inoculated on Cystine Lactose Electrolyte-Deficient agar and incubated at 37°C for 24 h.

Results: In our study, 303 (60.6%) samples showed growth of isolates. Among 303 isolates, 31 were *Candida* spp. which was not included in the study. Out of 272 isolates, *Escherichia coli* was 31.68%, followed by *Klebsiella* spp. (27.72%), *Enterococcus faecalis* (22.44%), *Citrobacter* spp. (3.96%), *Staphylococcus aureus* (2.64%), and *Pseudomonas* spp. (1.32%). For Gram-negative and Gram-positive isolates, the most effective antibiotic was nitrofurantoin.

Conclusion: The study shows *E. coli* as the predominant pathogen that causes UTI. Nitrofurantoin and fosfomycin showed very high susceptibility to uropathogens and can be used to treat primary or uncomplicated UTI.

Keywords: Urinary tract infection, *Escherichia coli, Klebsiella* spp., *Enterococcus faecalis* nitrofurantoin, Fosfomycin.

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INTRODUCTION

Urinary tract infections (UTIs) are one of the major types of infection of the human body that involves any part of urinary tract [1]. The UTI might be symptomatic or asymptomatic, and any of which if left untreated can cause serious problems [2]. UTI can be classified into uncomplicated and complicated cases. Uncomplicated UTI usually occurs in women due to bacterial infection while complicated UTI occurs in males as well as in females and is a challenge to treat [3]. The symptoms depend on structure of the urinary tract involved, infecting organism, severity of infection, and the status of immunity of patient [3]. Common signs and symptoms include fever, dysuria, increased frequency of micturition, chills, and cloudy or foul-smelling urine [4]. Almost 95% of all UTIs are caused by bacteria which include Gram-negative bacteria (80–85%) and Gram-positive bacteria (15–20%) [1,5].

In cases of UTI, empirical treatment is started before the results of urine culture and antibiotic susceptibility testing which aids in increasing drug resistance among bacteria. The appropriate use of antibiotics not only reduces the stay of patients in the hospital but also helps the patient financially and also reduces the drug resistance. Hence, it becomes important to monitor antibiotic treatment in the management of UTI to minimize therapeutic failures [6,7].

Hence, the study was designed to summarize routine identification and antimicrobial susceptibility pattern of organisms isolated from patients suffering from UTI in the Department of Microbiology, Maharishi Markandeshwar Institute of Medical Sciences and Research, Maharishi Markandeshwar (Deemed to be University), Mullana, Ambala, India.

METHODS

A 6-month (January 2019–June 2019) cross-sectional study was conducted in the Department of Microbiology, Maharishi Markandeshwar Institute of Medical Sciences and Research, Mullana, Ambala, and a total of 500 urine samples of clinically suspected cases of UTI were processed as per standard operating procedures. The approval of the Institute's Ethical Committee was taken before the sample collection and informed consent was obtained from all the patients.

Sample collection

Clean-catch midstream urine or aspirated urine from catheter tube, suprapubic aspiration from all suspected UTI patients was collected in a wide mouth sterile container and transported immediately to the microbiology department. All samples collected were analyzed for the presence of pus cells, epithelial cells, red blood cells, cast, and crystals.

Isolation and identification of organisms

A calibrated inoculating loop was used to place 0.01 ml of urine for inoculation on Cystine Lactose Electrolyte-Deficient agar, and these plates were incubated at 37°C for 24 h. Cultures were examined and colonies were counted for consideration of significant or insignificant bacteriuria. The colony count of $>10^5$ CFU/mL was considered as significant bacteriuria as per "Kass Phenomenon" [8]. The identification of the isolated organisms was done as per standard operating procedures [9].

Antibiotic susceptibility testing

Antibiotic susceptibility testing was done by Kirby–Bauer disc diffusion method using Clinical and Laboratory Standards Institute (CLSI) guidelines, 2019 [10]. The following antibiotic discs (HiMedia Laboratories, India) as per CLSI guidelines were tested: For Gram-negative bacteria: (*Escherichia coli, Klebsiella* spp., and *Citrobacter* spp.) – lomefloxacin (10 mcg), cinoxacin (100 mcg), ofloxacin (5 mcg), levofloxacin (5 mcg), ampicillin (10 mcg), carbenicillin (100 mcg), piperacillin-tazobactam (100/10 mcg), ceftriaxone (30 mcg), gentamycin (10 mcg), amikacin (30 mcg), nitrofurantoin (300 mcg), and cotrimoxazole (25 mcg). In addition to this, against *E. coli,*

fosfomycin (50 mcg) was also tested. For Gram-negative bacteria: *Pseudomonas* spp. – lomefloxacin (10 mcg), ofloxacin (5 mcg), carbenicillin (100 mcg), nitrofurantoin (300 mcg), and tetracycline (30 mcg) were only tested.

- For Gram-positive bacteria: *Staphylococcus aureus* – lomefloxacin (10 mcg), cotrimoxazole (25 mcg), nitrofurantoin (300 mcg), cefoxitin (30 mcg), and penicillin (10 unit).

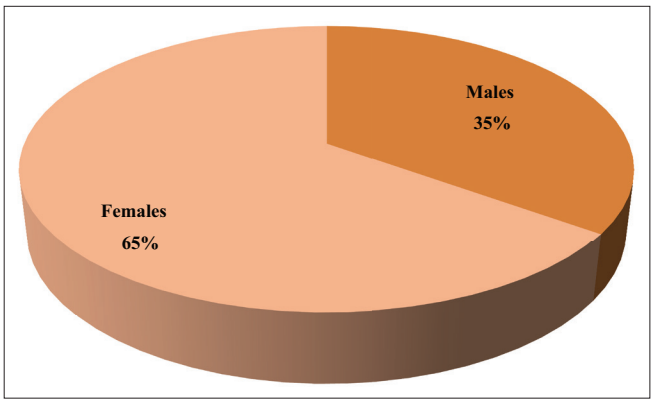

Fig. 1: Sex-wise distribution of positive samples (n=303)

Table 1: Total number of samples showing growth n (%)

Total number of samples	Number of samples showing growth	Number of sterile samples
500	303 (60.6)	197 (39.4)

Table 2: Distribution of isolates

Isolate	Number of isolates (n=303), n (%)
Gram-negative bacteria	
Escherichia coli	96 (31.68)
Klebsiella spp.	84 (27.72)
Citrobacter spp.	12 (3.96)
Pseudomonas spp.	4 (1.32)
Gram-positive bacteria	
Staphylococcus aureus	8 (2.64)
Enterococcus faecalis	68 (22.44)
Fungus	
Candida albicans	31 (10.23)

- *Enterococcus faecalis*: Ciprofloxacin (5 mcg), levofloxacin (5 mcg), penicillin (10 unit), ampicillin (10 mcg), nitrofurantoin (300 mcg), tetracycline (30 mcg), and fosfomycin (50 mcg).

RESULTS

A total of 500 samples were processed, of which 303 (60.6%) samples showed microbial growth (Table 1). Among 303 isolates, 197 (65%) were from female patients and 106 (35%) were from male patients (Fig. 1). Out of 303 isolates, 272 were bacterial isolates while 31 were *Candida* spp. Among the 272 isolates, *E. coli* (31.68%) was predominant followed by *Klebsiella* spp. (27.72%) and *E. faecalis* (22.74%) (Table 2).

E. coli was most susceptible to fosfomycin (100%) followed by nitrofurantoin (97.92%) and least susceptible to levofloxacin and ofloxacin (12.5%), respectively. *Klebsiella* spp. was most susceptible to nitrofurantoin (85.71%) followed by piperacillin-tazobactam (80.95%) and least susceptible to ampicillin (9.52%). *Citrobacter* spp. was most susceptible to nitrofurantoin (75%) followed by piperacillin-tazobactam (66.67%) and least susceptible to ampicillin (8.33%). *Pseudomonas* spp. was most susceptible to nitrofurantoin (75%) and least susceptible to carbenicillin (25%). Among Gram-positive bacteria, *E. faecalis* was most susceptible to fosfomycin (100%) and least susceptible to ciprofloxacin (23.53%) and *S. aureus* was most susceptible to nitrofurantoin (100%) and least susceptible to lomefloxacin (12.5%). Among Gram-positive and Gram-negative organisms, the highest susceptibility was shown by nitrofurantoin (Table 3).

DISCUSSION

In this study, a total of 500 samples were collected from January 2019 to June 2019 from clinically suspected cases of UTI. In this study, overall isolation rate was 60.6% which was higher than studies conducted by Patel *et al.* (45.69%) [1]. This study also showed that females (65%) were more prone to UTI than males (35%) which is supported by the study of Maji *et al.* [11] showing a higher prevalence of UTI in females (54.68) as compared to males (45.31%) (Table 1 and Fig. 1). Among all pathogenic isolates, *E. coli* (31.68%) was predominant followed by *Klebsiella* spp. (27.72%) and *E. faecalis* (22.44%) which is supported by the study of Basavaraj and Jyothi [12], who showed *E. coli* (45%) as most predominant uropathogen.

E. coli, the predominant pathogen in our study showed high susceptibility to fosfomycin (100%), nitrofurantoin (97.92%), and amikacin (87.5%), respectively, which is supported by the studies conducted by Lawhale and Naikwade [13]. In our study, *Klebsiella* spp. isolated was highly sensitive to nitrofurantoin (85.71%) and piperacillin-tazobactam (80.95%) which is in the concordance of the studies conducted by

Table 3: Antibiotic susceptibility rate of isolated uropathogens, n=272

Antibiotic	*Escherichia coli* (n=96), n (%)	*Klebsiella* spp. (n=84), n (%)	*Citrobacter* spp. (n=12), n (%)	*Pseudomonas* spp. (n=4), n (%)	*Enterococcus faecalis* (n=68), n (%)	*Staphylococcus* spp. (n=8), n (%)
Lomefloxacin	24 (25)	16 (19.04)	4 (33.33)	2 (50)	-	1 (12.5)
Ciprofloxacin	-	-	-	-	16 (23.53)	-
Cinoxacin	20 (20.83)	12 (14.29)	5 (41.67)	-	-	-
Ofloxacin	12 (12.5)	16 (19.05)	4 (33.33)	2 (50)	-	-
Levofloxacin	12 (12.5)	16 (19.05)	6 (50)	-	16 (23.53)	-
Penicillin	-	-	-	-	32 (47.06)	2 (25)
Ampicillin	12 (12.5)	8 (9.52)	1 (8.33)	-	56 (82.35)	-
Carbenicillin	24 (25)	16 (19.05)	4 (33.33)	1 (25)	-	-
Piperacillin -tazobactam	52 (54.16)	68 (80.95)	8 (66.67)	-	-	-
Ceftriaxone	36 (37.5)	12 (14.29)	4 (33.33)	-	-	-
Gentamycin	68 (70.83)	60 (71.43)	6 (50)	-	-	-
Amikacin	84 (87.5)	56 (66.67)	4 (33.33)	-	-	-
Nitrofurantoin	94 (97.92)	72 (85.71)	9 (75)	3 (75)	67 (98.53)	8 (100)
Cotrimoxazole	16 (16.67)	12 (14.29)	3 (25)	-	-	5 (62.5)
Fosfomycin	96 (100)	-	-	-	68 (100)	-
Tetracycline	-	-	-	2 (50)	20 (29.41)	-

Lawhale and Naikwade and Dipak *et al*. [13,14]. *E. faecalis* was the third major organism isolated showing high susceptibility to fosfomycin and nitrofurantoin which is supported by the studies conducted by Lawhale and Naikwade and Biswas *et al*. [13,15]. The other organisms isolated also showed high sensitivity to nitrofurantoin which is also supported by the studies of Lawhale and Naikwade, Dipak *et al.,* and Biswas *et al.,* respectively [13-15]. Overall nitrofurantoin (88.69%) was the most effective antibiotic due to its broad-spectrum activity which is in the concordance of the studies conducted by George *et al.* (84.15%) [16].

CONCLUSION

The most predominant pathogen in present study was *Escherichia coli* because of mannose resistant fimbriae which helps in colonization of uropathogenic strains onto uroepithelial cells followed by *Klebsiella spp*. and *Enterococcus faecalis*. As per this study, Nitrofurantoin was the most effective antibiotic and can be used for emperical treatment of UTI but culture and sensitivity should be performed for control of anti-microbial resistance.

AUTHORS' CONTRIBUTIONS

Study conception and design: Harit Kumar and Varsha A Singh.

Acquisition of data: Harit Kumar and Shavi Nagpal.

Analysis and interpretation of data: Harit Kumar and Dipankar Biswas.

Drafting of manuscript: Harit Kumar and Varsha A Singh.

Critical revision: Varsha A Singh and Shavi Nagpal.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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