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A cross-sectional study of most prevalent uropathogens in urinary tract infection in relation to gender and antibiotic sensitivity.

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Abstract: *Introduction: Knowledge of most prevalent uropathogens in a geographic area is vital for treating physicians of that area to make a better choice of empirical antibiotics. In addition, understanding how the most prevalent uropathogens are associated with gender can be helpful to implement better preventive health initiatives to decrease the prevalence of urinary tract infection (UTI) in the susceptible gender. This study primarily aims to study the most common UTI causing uropathogens' antibiotic sensitivity pattern and their association with gender.*

Methods: This cross-sectional study was done retrospectively using positive urine culture reports of 140 patients tested between January 2011 to January 2014 in a tertiary care health facility in Mumbai, India. Most common uropathogens were identified and their antibiotic sensitivity and association with gender were assessed.

Results: Most prevalent uropathogens were E. coli (47%), Klebsiella spp. (28%) and Pseudomonas spp. (20%). Nitrofurantoin's sensitivity to E. coli was significantly higher than the other two uropathogens. Females were more prone to UTI due to these uropathogens.

Conclusion: E. coli, Klebsiella spp. and Pseudomonas spp. were the most common uropathogens. Nitrofurantoin may be considered as the empiric antibiotic of choice for UTI. Prosperous sensitivity profile of Chloramphenicol against E. coli and Klebsiella spp. requires further research for its possible usage in UTI. Public health prevention methods focused primarily toward females to control the UTI prevalence due to these uropathogens needs to be considered seriously.

Keywords: *UTI, Uropathogens, Antibiotics, Susceptibility, Gender, Mumbai*

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I. Introduction

Urinary tract infection (UTI) is one of the commonest forms of health problem that presents to the outpatient departments, and also ranks on the top of the list of infections that cause nosocomial infection [1,2]. Depending on the type of treatment required, UTI can be classified into uncomplicated and complicated type [1]. About 75% to 90% of cases of uncomplicated UTI is attributed to Escherichia coli (E. coli), making it the most common uropathogens [3,4]. Whereas complicated UTI is due to the much wider spectrum of uropathogens [1].

Most of the time UTI is treated using empirical antibiotics, given before the culture and sensitivity reports of the urine samples are available [5], and this may give rise to antibiotic-resistance due to possible misuse [6]. In this era, a problem most healthcare providers face is the rising toll of uropathogens resistant to various antibiotics [7]. Knowledge about these causative microbial organisms and their antibiotic susceptibility pattern specific to a geographic location can help clinicians to choose better antibiotics for empirical treatment for their local patient-population [8].

Therefore, it is important to have an exact idea about the most common uropathogens that cause UTI in a particular community, and the antibiotics to which these bacteria are most sensitive and resistant to so that a better and appropriate choice of antibiotics can be made. In addition to that, it's also important to know the gender that is more susceptible to these uropathogens so that empirical treatment and preventive public health approaches can be directed towards that gender with greater confidence. This study was conducted on the basis of secondary data of urine culture reports obtained from the laboratory of Bhabha Atomic Research Centre

Hospital (BARCH), Mumbai, India. This study was done retrospectively, based on culture positive urine report of patients, between January 2011 and January 2014. One hundred forty urine culture positive cases were studied.

II. Methods

This study was approved by the scientific committee and ethics committee of BARCH. The study population of this study included all patients those who received treatment at BARCH, Mumbai between January 2011 to January 2014, at dispensaries, at outpatient departments and in inpatient departments and whose urine samples were reported as culture positive for bacterial growth (with counts of more than $\geq 10^5$ CFU/mL) by the laboratory of BARCH, Mumbai.

The study was done retrospectively. Data on age, sex, the result of urine culture, etiological agent, and susceptibility pattern were obtained from the laboratory records. Our sample size was 140. No intervention was done during the study, neither any patient was contacted. The data were analysed to understand the antibiotic susceptibility pattern of most prevalent uropathogens.

Mid-stream urine sample collected in sterile containers from 140 suspected UTI cases at the laboratory of BARCH, Mumbai were studied. The sample size of 140 was determined using a reference to other similar studies where sample size range varied between 140 to 228 [5,6,9,10].

Samples were inoculated on MacConkey agar and Nutrient agar plates and were read after overnight incubation at 37° C. For this study, significant bacteriuria (urine culture positive) was defined at a concentration of 10^5 CFU/mL associated with microscope findings of >10 WBC per high power field [11] and was the inclusion criteria of this study. Urine samples with a count less than 10^5 CFU/ml were considered not significant for bacteriuria (culture negative) and were excluded from the study.

In the lab, urine samples were tested for the uropathogens and their antibiotic susceptibility pattern. Identification of bacterial uropathogens was made on the basis of Gram reaction, morphology, and biochemical features. Isolates were tested for antimicrobial susceptibility by the standard disk diffusion method. Mueller-Hinton agar plates were incubated for 24 hours after inoculation with organisms and placement of the disks and inhibition zones were measured.

The commercial antibiotics used for isolates included Ciprofloxacin, Norfloxacin, Sparfloxacin, Ofloxacin, Trimethoprim-sulfamethoxazole, Gentamicin, Nitrofurantoin, Ceftriaxone, Ceftizoxime, Cephalexin, Cephalexin, Cephalothin, Amoxicillin, Ampicillin, Sulbactam, Ceftazidime, Chloramphenicol, Tetracycline, Imipenem and Meropenem.

The prevalence of various uropathogens were determined along with their antibiotic susceptibility and resistance pattern by frequency and percentage calculation. Further analysis was done for the most prevalent three uropathogens in relation to gender. Besides that, this 3 uropathogens sensitivity were compared for the antibiotic to which each of these uropathogens were sensitive to. The data was evaluated using Microsoft Office Excel program and R statistical software. In this study statistical significance was considered at a p-value of less than 5%. This study has made every attempt to adhere to quality reporting guideline for observational studies, the STROBE statement [12].

III. Results

In this study the most prevalent uropathogen was E. coli accounting for 47% of the UTI cases followed by Klebsiella spp. (28%) and Pseudomonas spp. (20%) (Table 1). About a percent of the uropathogens were gram-positive organisms. Sex distribution wise majority (70%) were females and about 30% were males. Infection due to E. coli and Klebsiella spp. was more common in females, nearly 53% and 29% respectively (Table 1). Whereas Pseudomonas spp. were predominantly found in males (38%) (Table 1). The age range of patients was between 1 to 82 with the average age of 37.36. The mean age of both the genders was equal, 38. Most patients (about 75%) in the studied population were between 15 to 59 years of age (Table 2). For these three most prevalent uropathogens (i.e. E. coli, Klebsiella spp. and Pseudomonas spp.) females showed a statistically significant difference ($X^2(2) = 12.08$, p-value = 0.002) for E. coli and Klebsiella spp. in comparison to males (Table 3).

Isolates of E. coli were most susceptible to Nitrofurantoin and Chloramphenicol, 94% each. Next in the rank of sensitivity for E. coli were Sparfloxacin and Ofloxacin, attributing to 89 and 88 percentages respectively. Most Klebsiella spp. were sensitive to Chloramphenicol (95%) followed by Ofloxacin

(85%).Whereas Pseudomonas spp. demonstrated maximum sensitivity to Ofloxacin and Ciprofloxacin (accounting to 79% and 75% respectively). Antimicrobial sensitivity pattern is summarized in Table 4.

For these most common uropathogens, statistical analysis was done for each of the antibiotics to which they each showed the highest prevalence of sensitivity, to assess if the susceptibility pattern was statistically significant or not (Table 5). Nitrofurantoin's sensitivity to E. coli was statistically significantly higher than Pseudomonas spp. and Klebsiella spp. ($X^2(2) = 10.44$, $p = 0.005$). Chloramphenicol and Nitrofurantoin's sensitivity to E. coli and Klebsiella were statistically significantly higher than that of Pseudomonas spp. (Fisher's Exact Test, p-value = 0.011). However, no statistically significant difference could be achieved for Ofloxacin sensitivity among the three most common uropathogen ($X^2(2) = 0.20559$, $p = 0.902$).

An additional analysis of resistance pattern of the uropathogens was also done (Table 6). E. coli had the highest resistance to Cephalexin and Co-trimoxazole (57.58%. and 45.45% respectively). Maximum resistance for Klebsiella spp. was noted for Cephalexin (61.54%) and Co-trimoxazole (51.28%). Most Pseudomonas spp. were resistant to Co-trimoxazole (about 79 percent).

IV. Discussions

UTI is a common health problem that physicians in developing nations have to encounter frequently [13,14] hence a knowledge about the most common uropathogens and their antibiotic sensitivity pattern specific to a particular area is important for better empirical treatment. The most prevalent uropathogen in our study population was E. coli (47%), like various other studies [15-18]. The second most common uropathogen was Klebsiella spp. (28%), similar to some other studies [15,19,20] and the third most common uropathogen was Pseudomonas spp. (20% of the study population). Low prevalence was noted for the gram-positive organisms like Staph aureus, resembling the finding of various other studies [18,21,22].

Next, we studied the antibiotic susceptibility pattern of the above mentioned most prevalent uropathogens. In this study most of the E. coli infected cases were sensitive to Nitrofurantoin, about 94%. This finding is probably justified since almost 90% of Nitrofurantoin is excreted through the renal system it attains a high urinary concentration making it an ideal drug for treating UTI caused by most gram positive and gram negative uropathogens [23]. In contrast to other Indian studies, those demonstrated E. coli to be highly resistant to chloramphenicol [7,24], in this study E. coli's sensitivity to chloramphenicol was just like that of Nitrofurantoin. Following Nitrofurantoin and Chloramphenicol, E. coli was most susceptible to Sparfloxacin and Ofloxacin, almost 88% to each of the latter.

The second common pathogen in this study was Klebsiella spp. which too was most susceptible to Chloramphenicol (95%) followed by Ofloxacin (85%). In contrast to these two uropathogens Pseudomonas spp. was most susceptible to Ofloxacin (79%) and then to ciprofloxacin (75%).

Chloramphenicol deserves special mention here. While it had been reported to be highly resistant to uropathogens in other Indian studies [7,24], in this study chloramphenicol resistance was quite low for E. coli and Klebsiella spp. (6% and 8% respectively). Therefore, based on the findings of this study an empirical use of Nitrofurantoin and Chloramphenicol may be justified for the two most prevalent uropathogen E. coli and Klebsiella spp. However, the authors of this study couldn't find any strong medical literature-based evidence regarding the use of chloramphenicol in urinary tract infections in human. One study on dogs depicted Chloramphenicol’s effectiveness to treat UTI due to Staphylococcus aureus (84%), E. coli (51%) etc. [25]. Therefore, in the current era of high antibiotic resistance to urinary tract infections future research for suitability of using chloramphenicol for UTI in any form may be fruitful.

Good sensitivity to the antibiotic Gentamicin was noted in this study for all three most common uropathogens. Highest sensitivity to gentamicin was observed for E. coli, followed by Klebsiella spp. and Pseudomonas spp. accounting to 80%, 72% and 61% respectively. This was opposite to what was seen in another Indian study done by Bajaj et al. (1999) which found about 84 percent resistance of Gentamycin for E. coli, Klebsiella spp. and Pseudomonas aeruginosa [7].

Next, regarding resistance pattern of these most common uropathogens in this study E. coli demonstrated maximum resistance against Cephalexin (58%) followed by Co-trimoxazole (45%) and Tetracycline (42%). In contrast to various other studies, which reported about increasing resistance of E. coli to Ampicillin [26-28], this study demonstrated a low level of resistance to this drug (only 27%). Amazingly, the resistance pattern of Klebsiella spp. was almost similar to that of E. coli demonstrating resistance against the following antimicrobial agents in decreasing order - Cephalexin, Co-trimoxazole and Tetracycline (61%, 51%

and 43% respectively). Pseudomonas spp. demonstrated maximum resistance to Co-trimoxazole followed by Ceftizoxime and Ampicillin, each quantitating to 60%, 53% and 50% respectively.

Gender wise, a female preponderance of UTI was noted in this study. More than 70% of the study population of this study who acquired UTI were females, like the data depicted in certain other studies [21,29,30]. The high female predisposition of UTI in females may be due to female anatomy of short urethra and proximity of urethra to the anus [10]. This finding might be helpful for planning UTI preventive approaches in the women of BARCH community like the use of probiotics, cranberry juice, intravaginal administration of oestrogen [31-35].

Interestingly, E. coli and Klebsiella spp. were more common in female patients (nearly 79 and 72 percent respectively) in comparison to male patients (about 21% and 28% respectively). The reverse was true for Pseudomonas spp., male acquired the infection almost 14% more than females.

Finally, the limitations of this study. This study population may not reflect the exact UTI load of the community studied here since many patients who receive antibiotics empirically might not be referred to the lab for a urine culture test possibly due to the improvement of their symptoms following the use of antibiotics. Besides that, there might be potential confounders that might have increased the chances of those who tested positive for the urine culture test like predisposing chronic illnesses (e.g. diabetes, chronic renal failure etc.), unhygienic sanitary habits, medication history (drugs that increase susceptibility to UTI by means of compromising immunity), history of smoking or drinking, sexual activity, urinary tract instrumentation, catheter per urethra, indwelling catheters etc. A regression analysis of possible confounders could have reduced these potential biases, which was beyond the scope of this study as data for this study was available for laboratory-based urine culture reports and few related demographic data. In future researchers may consider prospective double-blinded cohort study for minimizing the bias in such similar studies. Furthermore, this study limited its exploration primarily to the most prevalent three uropathogens and their antibiotic sensitivity pattern to the most common antibiotic to which these were susceptible to, limiting the scope of knowledge exploration in this regard.

V. Tables

Table 1- Uropathogens and sex distribution of the study population

	Male		Female		Total	
Organism	No.	%	No.	%	No.	%
E. Coli	14	33%	52	53%	66	47%
Klebsiella spp.	11	26%	28	29%	39	28%
Pseudomonas spp.	16	38%	12	12%	28	20%
Proteus mirabilis	1	2%	4	4%	5	4%
Proteus vulgaris	0	0%	1	1%	1	1%
Staph aureus	0	0%	1	1%	1	1%
Total	42	30%	98	70%	140	

Table 2 – Age distribution of the study population

Age groups	Frequency	Percentage
Age of 1-14	22	16%
Age of 15 - 59	105	75%
60 years and above	13	9%
Total	140	

Table 3 – Gender wise relation with 3 most prevalent uropathogens

	Uropathogens						
	E. coli		Pseudomonas spp.		Klebsiella spp.		Pearson's Chi-squared test
	No.	%	No.	%	No.	%	X-squared = 12.08, df = 2, p-value = 0.002381
Female	52	79	12	43	28	72	
Male	14	21	16	57	11	28	
Total	66		28		39		

Table 4 - Sensitivity pattern of uropathogens of the study population

	Nitrofurantoin		Chloramphenicol		Sparfloxacin		Tetracycline		Ceftriaxone		Co-trimoxazole		Gentamycin		Cephalexin		Ampicillin		Ciprofloxacin		Ofloxacin		Norfloxacin		NI		Rifampicin		Ceftriaxone	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
E. Coli	62	94	62	94	59	89	37	56	41	62	36	55	53	80	27	41	49	74	47	71	58	88	39	59	1	2	0	0	41	62
Pseudomonas spp.	17	61	20	71	20	71	14	50	12	43	11	39	17	61	5	18	13	46	21	75	22	79	12	43	1	4	4	14	12	43
Klebsiella spp.	28	72	37	95	29	74	20	51	25	64	18	46	28	72	13	33	25	64	29	74	33	85	22	56	0	0	0	0	25	64
Proteus mirabilis	1	20	3	60	5	100	1	20	1	20	3	60	4	80	4	80	4	80	5	100	5	100	3	60	0	0	0	0	5	100
Proteus vulgaris	0	0	1	100	1	100	0	0	0	0	1	100	1	100	1	100	1	100	1	100	1	100	1	100	0	0	0	0	0	0
Staph aureus	1	100	1	100	1	100	0	0	1	100	1	100	1	100	1	100	1	100	1	100	1	100	1	100	0	0	0	0	1	100

Table 5 Comparative analysis of 3 most common uropathogens with respect to Nitrofurantoin, Chloramphenicol and Ofloxacin

		Nitrofurantoin sensitive		Nitrofurantoin resistant		Pearson's Chi-squared test	Chloramphenicol sensitive		Chloramphenicol resistant		Fisher's Exact Test for Count Data	Ofloxacin sensitive		Ofloxacin resistant		Pearson's Chi-squared test
		No.	%	N	%		No.	%	No.	%		No.	%	No.	%	
Uropathogens	E. Coli	62	58%	4	21%	X-squared= 10.444, df =2, p-value= 0.005	62	52	4	27	p-value= 0.011	58	51	15	47	X-squared= 0.20559, df=2, p-value= 0.902
	Pseudomonas spp	17	16%	8	42%		20	17	8	53		22	20	7	22	
	Klebsiella spp	28	26%	7	37%		37	31	3	20		33	29	10	31	
Total		107		19			119		15			113		32		

Table 6- Resistance pattern of uropathogens of the study population

Organism	Ofloxacin		Ciprofloxacin		Norfloxacin		Chloramphenicol		Cephalexin		Tetracycline		Ampicillin		Co-trimoxazole		Ceftriaxone		Sparfloxacin		Imipenem		Meropenem		Ceftazidime		NI		Cefoperazone		Nitrofurantoin		Gentamycin		Cefotaxime	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No	%	No.	%	No.	%	No	%	No.	%	No.	%	No	%	No.	%	No.	%	No.	%	No.	%
E. Coli	15	23	19	29	25	38	4	6	38	58	28	42	18	27	30	45	26	39	7	11	0	0	0	0	1	2	19	29	0	0	4	6	10	15	0	0
Pseudomonas spp.	7	25	7	25	13	46	8	29	13	46	13	46	14	50	17	61	15	54	5	18	1	4	0	0	1	4	3	11	1	4	8	29	9	32	1	4
Klebsiella spp.	10	26	10	26	17	44	3	8	24	62	17	44	13	33	20	51	13	33	9	23	3	8	2	5	1	3	10	26	0	0	7	18	8	21	0	0
Proteus mirabilis	0	0	0	0	2	40	2	40	1	20	4	80	0	0	2	40	0	0	0	0	1	20	1	20	0	0	1	20	0	0	4	80	0	0	0	0
Proteus vulgaris	0	0	0	0	0	0	0	0	0	0	1	100	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	100	0	0	0	0
Staph aureus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	100	0	0	0	0	0	0	0	0

VI. Conclusion

Most common uropathogens responsible for UTI in this study's patient population were E. coli, followed by Klebsiella spp. and Pseudomonas spp. UTI was more common in females than males. Females more commonly acquired the infection by E. coli and Klebsiella spp., whereas Pseudomonas spp. infection predominated in male patients. These findings along with the significant association between the most prevalent uropathogens and female gender emphasise the consideration of preventive public health approaches (like probiotics, cranberry juice, intravaginal administration of oestrogen etc.) in females, to decrease the prevalence of these uropathogens among females.

Nitrofurantoin may be used as the first line drug to treat UTI empirically. Good sensitivity results of Chloramphenicol to the common uropathogens (like E. coli and Klebsiella spp.) suggests the need for future research about usefulness of Chloramphenicol in UTI treatment. This is particularly important in the current time when the world is facing high antibiotic resistance to the most prevalent uropathogens.

Conflict of interest:There is no conflict of interest.

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