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Prevalence of Uropathogens Causing Urinary Tract Infection and their Antimicrobial Susceptibility among the Reproductive Age Group in Theni District, India

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ABSTRACT

Keywords

Urinary tract infection, Uropathogens, Antibiotic susceptibility.

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Urinary Tract Infection (UTI) is one of the most common infections especially among the population in reproductive age group. This study was designed to isolate, characterize and to determine the antimicrobial susceptibility of uropathogens causing UTI, within the age group of 25-40 yrs attending Govt. Theni Medical College Hospital. The study identified *Escherichia coli* as the most common cause of UTI with 28% proportion among the positive cultures and most isolates of them were found susceptible to colistin (92.3%), tigecycline (84.3%), meropenam (79.8%) and gentamicin (79.4%).

Introduction

Urinary Tract Infection is the presence of symptoms like increased urinary frequency, urinary urgency and pain during urination which is the most common bacterial infection, accounts for about 25% of all infections.

UTI may occur in any population with any age group; however, the infection is most common in women of reproductive age group (Karki *et al.*, 2004).

Although the *Escherichia coli* is being observed as a most common cause of UTI in all populations from different geographic regions its susceptibility pattern varied with reports (Gupta *et al.*, 1999, Moges *et al.*, 2002). It is important to note that the prevalence of antibiotic resistance among uropathogens is increasing worldwide (Bashir *et al.*, 2008). The empirical selection of antibiotic should cover the likely pathogen with its likely susceptibility based on the recent records and local epidemiological information. Therefore, this study is aimed to determine the prevalence of uropathogens and their susceptibility pattern, which will help the clinician in choosing appropriate empirical therapy for UTI.

Materials and Methods

All participants attending the Govt. Theni Medical College Hospital as either outpatient or inpatient with the symptoms of UTI within the age group of 25-40 were included in this study. The study conducted over the period of two years from January 2015 to August 2017 among the population of Theni district (Tamil Nadu, India).

Sample collection

The physician clinically examined all patients and those who suffer with the symptoms of UTI were empirically treated with the existing strategy (3^{rd} generation cephalosporin or fluoroquinolone) in our hospital.

Those who fail to respond to this empirical therapy were referred to the Microbiology Laboratory for Bacterial culture and susceptibility testing during their second visit. About 10-20 ml of mid-stream urine sample was collected in a sterile urine container from the study participants and the sample was immediately transported to the laboratory at room temperature.

Culture and identification of organism

All samples were immediately processed after receipt at the Microbiology laboratory by inoculating 10 µL of urine sample using calibrated inoculation loop on blood agar and MacConkey agar by following standard procedure. Then the culture plates were incubated at 35˚C for overnight. After incubation, the culture plates were examined for the formation of significant number (> 30 CFU) of Colony Forming Units (CFU). Then the samples found to have significant bacteriuria (10^5bacteria/mL) were further processed towards the identification of organism by Standard Biochemical Tests (Razak and Gurushantappa, 2012) and Antimicrobial Susceptibility Test by Kirby-Bauer disk diffusion method.

Antimicrobial susceptibility testing

Antimicrobial susceptibility testing was performed on Mueller–Hinton agar (HiMedia, Mumbai) by the Kirby–Bauer disk diffusion method according to Clinical and Laboratory Standards Institute (CLSI) guidelines [CLSI 2015] using *E. coli ATCC* 25922 and *Staphylococcus aureus* ATCC 25923 as quality control strains. Clinical isolates of *E. coli* or *Klebsiella* spp. were inoculated in saline and a direct colony suspension was prepared and adjusted to a 0.5 McFarland turbidity to contain 1.5 x 10^8 CFU/mL. The test strain was lawn cultured on a Mueller–Hinton agar plate, then antibiotic disks were placed and the plate was incubated at 35˚C for 16–18 h in ambient air. Following incubation, the diameter of the zone of inhibition was measured around each antibiotic disk and the data were stored in WHONET software v.5.6 (http://www.whonet.org). Cumulative antimicrobial susceptibility results were interpreted based on CLSI guidelines [CLSI, 2015]. Because of the non-availability of resistance criterion to colistin, a zone of inhibition of <11 mm was recognized as non-susceptible and was considered as resistant for this study. Furthermore, for tigecycline the US Food and Drug Administration (FDA) susceptibility criterion was used since CLSI breakpoints are not yet established.

Data analysis

The cumulative antimicrobial susceptibility testing data were retrieved from the computerized database (WHONET software ver. 5.6) and the prevalence of antimicrobial resistance was analyzed for Gram-positive and Gram-negative organisms using WHONET software. All analyzed data were presented as a percentage because of the variability in the number of tests conducted with different antibiotics.

Results and Discussion

A total of 2190 patient samples including 575 male and 1615 female were processed during the study period. Among them 133 and 528 were identified to have significant bacteriuria by culture identification. The leading causes of UTI were identified as *Escherichia coli*, Coagulase Negative Staphylococci, *Klebsiella* spp. and *Pseudomonas aeruginosa* (Table 1; Figure 1). Notably, leading causes of UTI were unchanged between male and female. Further, significant deviation was not observed among all causes of UTI. Interestingly, enterococci, citrobacter and enterobacter were higher among male when compared with female (Figure 1). Interestingly, *Proteus* spp. and *Candida* spp. was not observed among male participants in this study. The causative organisms were grouped as Gram-positive and Gram-negative and its susceptibility pattern was grossly analyzed. The study observed linezolid, vancomycin, and doxycycline, were having superior activity against Gram-positive organisms than other antimicrobials tested. Similarly, colistin, doripenam, tigecyclin, meropenam, pipercillin-tazobactam, and amikacin were active against Gram-negative organisms. Although the fluoroquinolones were the common choice for the treatment of UTI, this study observed less than 50% efficacy against Gram-negative isolates. This could be owing to that the patients included in this study were empirically treated with the strategy in our hospital; those who fail to respond to this empirical therapy were investigated. Thus, this study observed high rate resistance not only to fluoroquinolones but also for all tested antimicrobials (Tables 2 and 3; Figs. 2 and 3).

Table.1 Organisms isolated from significant bacteriuria

Organism	**Female**		**Male**	
	No.	**Percent**	**No.**	**Percent**
Escherichia coli	154	29.17%	35	26.32%
Staphylococcus, coagulase negative	121	22.92%	30	22.56%
Non-fermenting gram negative rods	67	12.69%	16	12.03%
Klebsiella spp.	48	9.09%	14	10.53%
Edwardsiella sp.	23	4.36%	4	3.01%
Morganella sp.	22	4.17%	8	6.02%
Pseudomonas aeruginosa	19	3.60%	7	5.26%
Enterococcus sp.	18	3.41%	8	6.02%
Candida sp.	15	2.84%	0	0.00%
Citrobacter spp.	17	3.22%	6	4.51%
Staphylococcus aureus	11	2.08%	2	1.50%
Enterobacter spp.	4	0.76%	3	2.26%
Proteus spp.	4	0.76%	0	0.00%
Others	5	0.95%	0	0.00%
Total	**528**	**100**	**133**	**100**

Table.2 Antimicrobial susceptibility – Gram positive cocci (isolates from both male and female; n= 190)

Antibiotic name	Number	%R	%S	%R 95%C.I.
Penicillin G	148	91.2	8.1	86.0-95.6
Ampicillin	156	90.4	9	85.1-94.8
Oxacillin	143	60.1	22.4	51.6-68.1
Cefazolin	125	59.2	36	50.0-67.8
Cefuroxime	99	56.6	32.3	46.3-66.4
Cefotaxime	159	61	32.7	53.5-69.1
Cefepime	156	63.5	32.7	55.4-70.9
Cefoxitin	119	70.6	28.6	61.4-78.4
Gentamicin	143	26.6	67.1	19.7-34.8
Ciprofloxacin	169	65.1	18.3	57.3-72.2
Levofloxacin	169	52.7	20.7	44.9-60.4
Norfloxacin	41	75.6	17.1	59.3-87.1
Trimethoprim/Sulfamethoxazole	161	55.9	31.7	47.9-63.6
Clindamycin	169	25.4	65.1	19.2-32.8
Erythromycin	164	31.7	32.3	24.8-39.5
Nitrofurantoin	28	14.3	85.7	4.7-33.6
Linezolid	168	1.2	98.8	0.2-4.7
Vancomycin	161	1.2	97.5	0.8-6.5
Doxycycline	166	18.1	77.1	12.7-25.0
Tetracycline	134	24.6	68.7	17.8-32.9

Table.3 Antimicrobial susceptibility – Gram negative bacilli (isolates from both male and female; n= 455)

Antibiotic name	Number	%R	%S	%R 95%C.I.
Ampicillin	396	94.4	3	91.5-96.4
Piperacillin	314	76.1	10.8	70.9-80.6
Amoxicillin/Clavulanic acid	398	84.4	10.8	80.4-87.7
Piperacillin/Tazobactam	416	18.8	67.1	15.2-23.0
Cefazolin	339	86.4	8.8	82.2-89.8
Cefuroxime	248	76.6	17.7	70.7-81.6
Ceftazidime	347	63.1	26.8	57.8-68.1
Cefotaxime	372	75.3	20.4	70.5-79.5
Cefepime	372	39.8	47.8	34.8-45.0
Cefoxitin	336	46.7	47.3	41.3-52.2
Aztreonam	389	60.7	26.5	55.6-65.6
Doripenem	234	17.9	76.9	13.3-23.6
Ertapenem	386	29.3	65.5	24.9-34.2
Meropenem	367	16.1	79.8	12.6-20.4
Amikacin	428	15.7	79.4	12.5-19.6
Gentamicin	361	50.1	45.2	44.8-55.4
Nalidixic acid	346	83.8	8.1	79.4-87.4
Ciprofloxacin	399	65.7	29.3	60.8-70.3
Gemifloxacin	328	58.8	31.7	53.2-64.1
Levofloxacin	398	58	31.4	53.0-62.9
Norfloxacin	293	66.6	30	60.8-71.9
Ofloxacin	373	61.1	31.9	55.9-66.0
Trimethoprim/Sulfamethoxazole	394	63.7	32.7	58.7-68.4
Colistin	416	7.7	92.3	5.4-10.8
Nitrofurantoin	290	36.2	56.2	30.7-42.1
Doxycycline	401	29.4	55.4	25.0-34.2
Tetracycline	345	49.9	44.3	44.5-55.3
Tigecycline	337	11.3	84.3	8.2-15.3

Fig.1 Significant bacteriuria: Organisms isolated - Female vs. Male

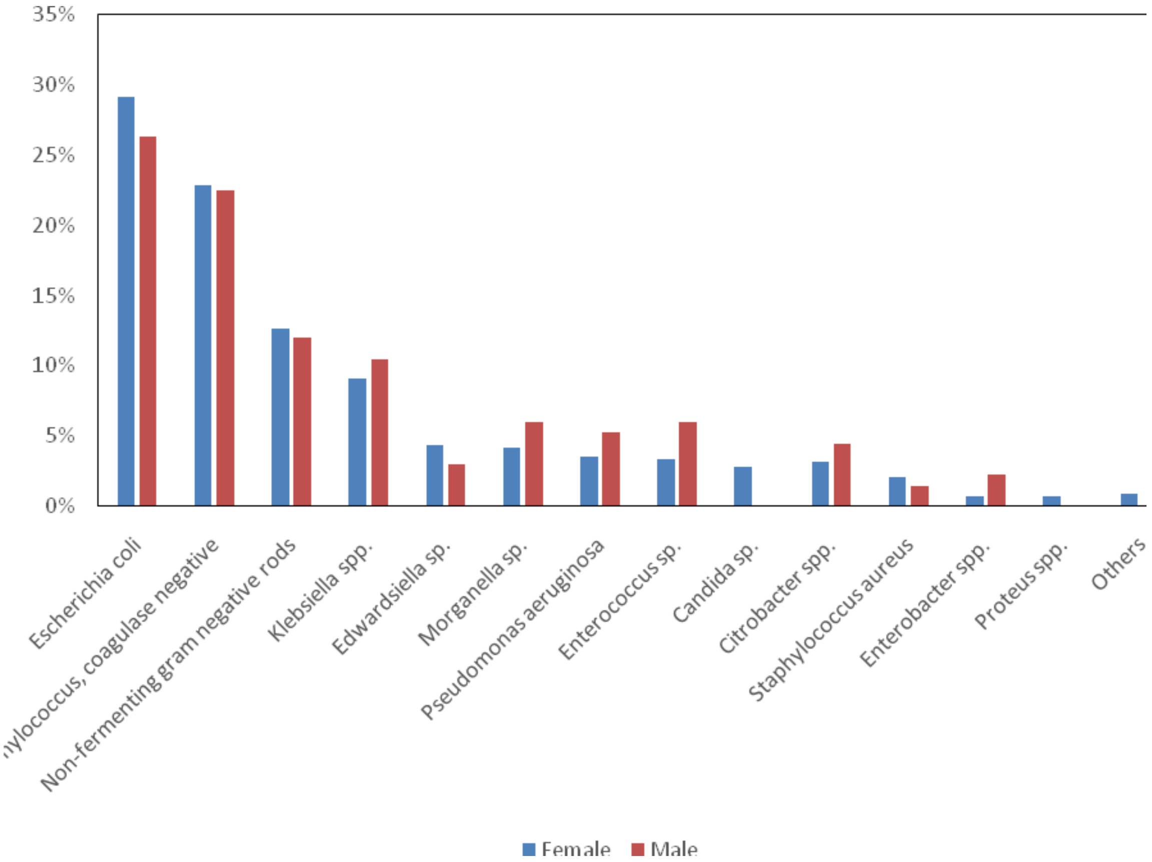


Fig.2 Susceptibility pattern of Gram positive cocci (n=190)

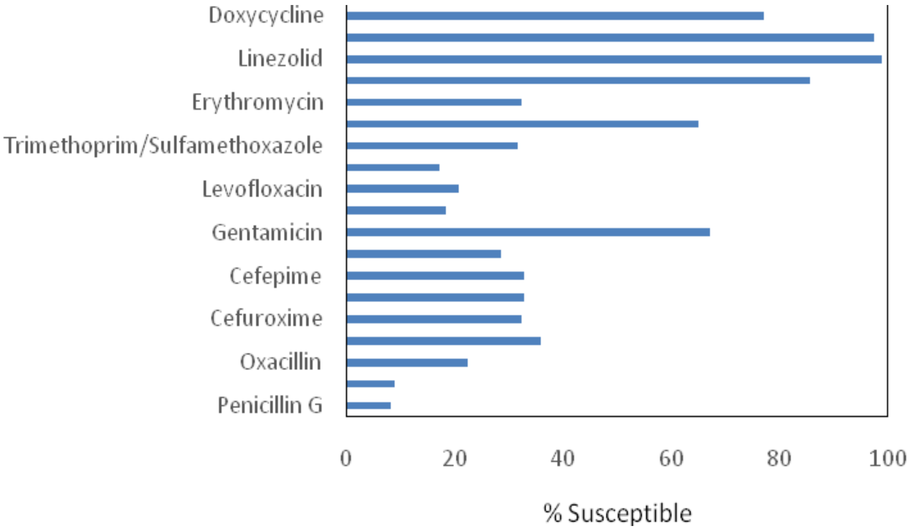
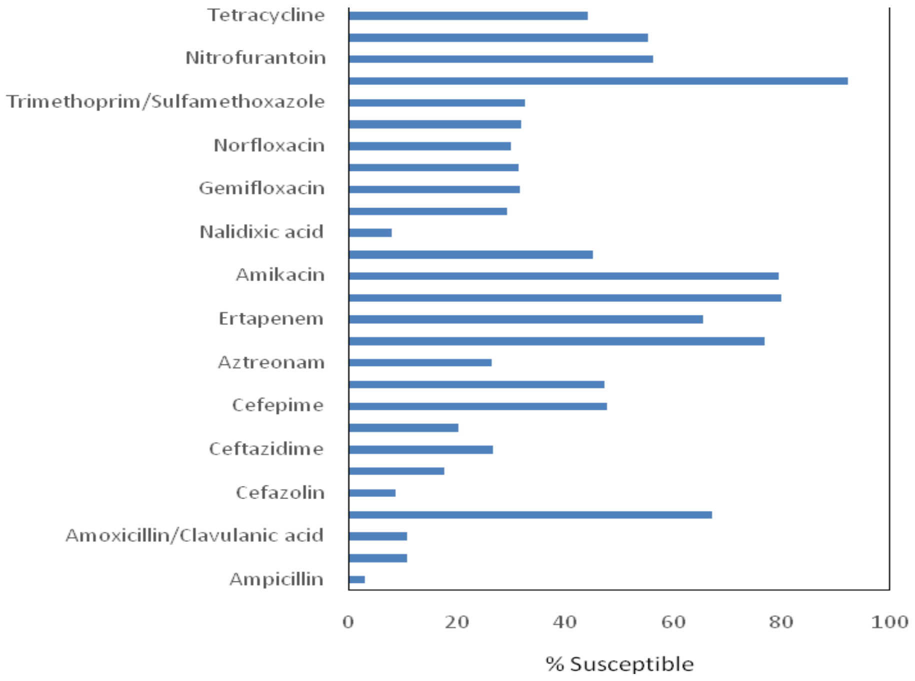


Fig.3 Susceptibility pattern of Gram negative bacilli (n= 455)



Our study results on the prevalence of pathogens were corroborated well with the earlier reports from India (Prakasam *et al.*, 2012) as well as from abroad (Moges *et al.*, 2002). The susceptibility data shows high prevalence of resistance when compared with other countries especially in Gram-negative organisms (Uwaezuoke *et al.*, 2014). However, when compared with reports from India, the prevalence of resistance was similar to earlier reports (Sekar *et al.*, 2016). This study shows that the predominant pathogen of UTI is *Escherichia coli*. This organism has acquired resistance to most of the commonly used antimicrobials may be due to indiscriminate usage of antibiotics. Periodic surveillance should be done to assess the prevalence of antimicrobial resistance as well as the emergence of newer types/mechanisms of resistance. These cumulative antimicrobial susceptibility data would help clinician to choose appropriate empirical antimicrobial form time to time.

The study observed *E. coli* and Coagulase Negative *Staphylococci* as the leading causes of UTI respectively among Gram-negative and Gram-positive organisms. The study observed nitrofurantoin or doxycycline could be used as empirical therapy for unresponsive cases with fluoroquinolones.

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