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Prevalence and antibiogram of bacterial isolates from urinary tract infections at Dessie Health Research Laboratory, Ethiopia

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PEER REVIEW

Peer reviewer

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Comments

The area of data collection by the authors is very much useful. The urine sample ratio is more in female than male. The highest isolation rate of uropathogens was obtained in the age group between 24 to 44 years. The antibiotic resistant pattern was observed only by Kirby–Bauer disc diffusion method.

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ABSTRACT

Objective: To determine the prevalence and antimicrobial susceptibility of bacteria from suspected urinary tract infections.

Methods: A retrospective analysis of bacterial pathogens and their antimicrobial susceptibility was done on urine samples at Dessie Regional Laboratory in the period 2003 to 2010. Antimicrobial susceptibility tests were done using disc diffusion technique as per the standard of Kirby–Bauer method.

Results: The male to female ratio of the patients was 1:1.96. Of the total 1404 samples, 319 (22.7%) were culture positive. *Escherichia coli* was the dominant isolate (63.6%) followed by *Klebsiella* spp. (8.5%) and *Proteus* spp. (8.2%). The overall resistance rates to erythromycin, amoxicillin, and tetracycline were 85.6%, 88.9% and 76.7%, respectively. The three most frequently isolated bacteria had resistance rates of 80.1%–90.0% to, amoxicillin, and tetracycline and sensitivity rates of 0 to 25% to nitrofurantoin, ciprofloxacin and gentamicin. Antibiogram of isolates showed that 152 (47.85%) isolates were resistance to two and more antimicrobials.

Conclusions: In the study area resistance rates to erythromycin, amoxicillin and tetracycline were high. Since most isolates were sensitive to nitrofurantoin and gentamicin, they are considered as appropriate antimicrobials for empirical treatment urinary tract infections.

KEYWORDS

Antimicrobial resistance bacteria, Ethiopia, Uropathogens

1. Introduction

Urinary tract infection (UTI) is a term applied to a variety of clinical conditions ranging from asymptomatic presence of bacteria in the urine to severe of the kidney with sepsis[1]. UTIs are one of the most common bacterial infections in humans both in the community and hospital settings[2]. Worldwide, approximately 150 million people are diagnosed with UTIs

resulting in USD 6 billion health care expenditures[1]. UTIs are the most common bacterial infections encountered by clinicians in developing countries[3].

Most UTIs are caused by Gram–negative bacteria like *Escherichia coli* (*E. coli*), *Klebsiella* spp., *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Acinetobacter* spp., and *Serratia* spp. and Gram–positive bacteria such as *Enterococcus* spp. and *Staphylococcus* spp[3,4]. *E. coli* is responsible to most

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UTIs[5,6]. Drug resistance among bacteria causing UTI has increased since introduction to UTI chemotherapy[7–11]. The etiological agents and their susceptibility patterns of UTI vary in regions and geographical location. Besides, the etiology and drug resistance change through time[12]. Knowledge of the local bacterial etiology and susceptibility patterns is required to trace any change that might have occurred in time so that updated recommendation for optimal empirical therapy of UTI can be made[13]. In Ethiopia, a number of studies have been done on the prevalence and antimicrobial resistance patterns of UTIs[3,4,7]. However, no data have been reported from the present study area. The aim of the present study was therefore to determine the prevalence of local bacterial isolates from suspected UTI and susceptibility to the most commonly used antimicrobials.

2. Materials and methods

2.1. Study design

A retrospective analysis of culture results of urine was performed at Dessie Regional Health Research Laboratory. The sex and age of patients, the organism isolated and the antimicrobial susceptibility profiles were collected from the registration records using a standard data collection form. The data were entered into Excel for analysis.

2.2. Culture and identification

As the standard operation procedures show clean–catch midstream morning urine specimens were collected using sterile wide mouth glass container. Urine samples were plated on cystine lactose electrolyte–deficient medium, MacConkey agar and, blood agar (Oxoid, Basingstoke, UK) using calibrated wire loops and then incubated aerobically at 37 °C for 24 h. From positive cultures, uropathogens were identified according to the standard operational procedures as per the standard microbiological methods[14]. A significant bacterium was considered if urine culture yield $\geq 10^5$ CFU/mL.

2.3. Antimicrobial susceptibility tests

According to the standard operational procedures, antimicrobial susceptibility tests were done on Mueller–Hinton agar (Oxoid, Hampshire, England) using Kirby–Bauer disk diffusion method[15]. The antimicrobial agents tested were: tetracycline (30 µg), nitrofurantoin (300 µg), erythromycin (15 µg), chloramphenicol (30 µg), gentamicin (10 µg), ciprofloxacin (5 µg), cephalothin (30 µg), doxycycline (30 µg), cotrimoxazole (25 µg), ceftriaxone (30 µg) and amoxycillin (10 µg) (Oxoid, England). Resistance data were interpreted according to Clinical laboratory Standards Institute. Reference strains of *E. coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923 (*S. aureus*) were used for quality control for antimicrobial susceptibility tests[16].

2.4. Data analysis

Chi–square test was employed to compare the proportion of bacterial isolates between sex and age and comparison of antimicrobial resistances. *P*–value of less than 0.05 was considered to indicate statistically significant difference.

2.5. Ethical considerations

Ethical approval was secured from Research Ethics Committee of Bahir Dar University. Permission from Dessie Regional Health Research Laboratory was also obtained.

3. Results

During 2003 to 2010, a total of 1404 urine samples from suspected UTIs were analyzed for isolation and identification of bacteria and antimicrobial susceptibility testing. The age of the patients ranged from 1 year to 85 years, with mean age of 32.26 (SD=14.45) years. The mean ages of male and female patients were 35.1 (SD=15.8) and 30.78 (SD=12.4) years, respectively. Nine hundred thirty (62.2%) urines samples were from female and 474 (33.8%) were from male patients with male to female ratio of 1:1.96. The demographic characteristics of the patients are shown in Table 1.

Table 1
Age and sex distribution of patients with suspected UTI.

Demographic characteristics	Positive No. (%)	Negative No. (%)	Total (%)	<i>P</i> value	
Age (years)	< 4	14 (48.3)	15 (51.7)	29 (100)	<i>P</i> =0.011
	5–14	22 (21.2)	82 (78.8)	104 (100)	
	15–25	84 (23.7)	270 (76.3)	354 (100)	
	26–44	128 (20.1)	509 (79.9)	637 (100)	
	> 44	71 (25.4)	209 (74.6)	280 (100)	
Gender	Male	67 (14.1)	407 (85.9)	474 (100)	<i>P</i> <0.001
	Female	252 (27.1)	678 (72.9)	930 (100)	
	Total	319 (22.7)	1085 (77.3)	1404 (100)	

The overall prevalence of the uropathogens was 319 (22.7%). Majority of the pathogens were isolated from females with isolation rate of 27.1% and 14.1% (95% *CI*=0.14) were from males. The highest isolation rate was observed in the age group between 26 to 44 years of age (Table 1). *E. coli* was the most predominant pathogen isolated from urine samples with prevalence of 203 (63.6%). *Klebsiella* spp., *Proteus* spp., *Pseudomonas* spp., coagulate negative staphylococci (CNS), *S. aureus*, *Enterobacter* spp. and *Citrobacter* spp. accounted for 36.4% of the isolates (Table 2). Gram negative and Gram positive bacteria were responsible for 91.9% and 9.1% of the isolates, respectively.

The overall susceptibility profiles of bacterial isolates are shown in Table 3. Erythromycin had the highest overall resistance of 85.6%, followed by amoxycillin (83.9%) and tetracycline (76.7%). Nitrofurantoin, gentamicin and ciprofloxacin had overall resistance rates of 5.5%, 24.3% and

29.4%, respectively (Table 3). Species specific antimicrobial resistance rates are displayed in Table 4. *E. coli*, the most frequently isolated bacterium, showed high resistance rates (>80%) to erythromycin, amoxicillin and tetracycline. The other three most common isolates exhibited resistance rates (80%–100%) to erythromycin and amoxicillin. Majority (96.2%) of *E. coli* isolates were susceptible to nitrofurantoin with resistance rate of 3.8%. The other isolates were sensitive to gentamicin and ciprofloxacin with resistance rates of 24%–35% and 0%–40%, respectively.

Table 2

Bacterial isolates from urine samples of patients with suspected UTI.

Bacteria Isolated	Frequency	Percent
<i>E. coli</i>	203	63.6
<i>Klebsiella</i> spp.	27	8.5
<i>Proteus</i> spp.	26	8.2
<i>Pseudomonas</i> spp.	22	6.9
CNS	17	5.3
<i>S. aureus</i>	12	3.8
<i>Enterobacter</i> spp.	7	2.2
<i>Citrobacter</i> spp.	5	1.6
Total	319	100

CNS: Coagulase negative staphylococci.

Table 4

Antimicrobial susceptibility of bacterial isolates from patients with suspected UTI.

Antimicrobial agents	<i>E. coli</i>		<i>Klebsiella</i> spp.		<i>Proteus</i> spp.		<i>Pseudomonas</i> spp.		CNS		<i>S. aureus</i>		<i>Enterobacter</i> spp.		<i>Citrobacter</i> spp.	
	# T	% R	# T	% R	# T	% R	# T	% R	# T	% R	# T	% R	# T	% R	# T	% R
Gentamicin	188	24.5	25	24.0	24	25.0	20	35.0	13	7.7	12	16.7	7	14.3	3	66.7
Tetracycline	151	80.1	16	81.3	16	87.5	8	87.5	24	54.2	12	66.7	6	50.0	3	66.7
Chloramphenicol	139	37.4	20	40.0	18	61.1	11	100.0	14	35.7	8	62.5	7	14.3	4	50.0
Cotrimoxazole	140	72.1	18	61.1	19	57.9	9	77.8	11	45.5	9	77.8	4	50.0	5	80.0
Erythromycin	79	93.7	7	85.7	9	88.9	8	100.0	14	28.6	7	71.4	6	100.0	2	100.0
Ciprofloxacin	80	34.1	6	16.7	5	40.0	5	0.0	11	18.2	6	33.3	7	14.7	1	0.0
Nitrofurantoin	85	3.8	7	0.0	4	0.0	12	16.7	3	0.0	1	0.0	1	0.0	2	50.0
Cephalothin	58	56.9	9	44.4	9	77.8	5	40.0	4	25.0	1	0.0	3	66.7	–	–
Ceftriaxone	56	33.9	6	50.0	8	62.5	8	50.0	6	0.0	3	75.0	3	66.7	2	0.0
Amoxicillin	55	87.3	8	87.5	10	90.0	5	80.0	4	0.0	2	100.0	2	100.0	1	100.0
Doxycycline	41	61.0	6	50.0	5	80.0	11	72.7	–	1.0	–	–	1	0		

CNS: Coagulase negative staphylococci; # T: Number of isolates tested against each antimicrobial agent; % R: Percent of isolates resistant to the antimicrobial agent.

Table 5

Multiple antimicrobial resistance patterns of bacterial isolates from patients with suspected UTI.

Bacteria	Antibiogram							
	R0	R1	R2	R3	R4	R5	R6	R8
<i>E. coli</i> (n=203)	15 (7.4)	32 (15.8)	53 (16.2)	45 (22.2)	43 (21.1)	11 (5.4)	3 (1.5)	1 (0.5)
<i>Klebsiella</i> spp. (n=27)	3 (11.1)	7 (25.9)	8 (29.6)	1 (3.7)	6 (22.2)	2 (7.4)	0	0
<i>Proteus</i> spp. (n=26)	0	4 (15.4)	10 (38.5)	5 (19.2)	3 (11.5)	2 (7.7)	2 (28.6)	0
<i>Pseudomonas</i> spp. (n=22)	1 (4.5)	7 (31.8)	3 (13.6)	6 (27.3)	4 (18.2)	1 (4.5)	0	0
CNS (n=17)	3 (42.9)	2 (28.6)	6 (85.7)	4 (57.1)	1 (14.3)	1 (14.3)	0	0
<i>S. aureus</i> (n=12)	0	3 (25.0)	4 (33.3)	2 (16.7)	1 (8.3)	0	1 (8.3)	1 (8.3)
<i>Enterobacter</i> spp. (n=7)	0	2 (28.6)	1 (14.3)	2 (28.6)	0	2 (28.6)	0	0
<i>Citrobacter</i> spp. (n=5)	1 (20.0)	0	2 (40.0)	2 (40.0)	0	1 (20.0)	0	0
Total isolates (n=319)	23 (7.2)	57 (17.9)	87 (27.3)	67 (21.0)	58 (18.2)	21 (6.6)	5 (1.6)	1 (0.3)

R0: Sensitive to all antimicrobials tested; R1, R2, R3, R4, R5, R6, R8: Resistant to one, two, three, four, five, six, eight antimicrobials, respectively.

Table 3

Overall antimicrobial susceptibility profiles of bacteria isolates from patients suspected of UTIs.

Antimicrobial agents	No. of Antimicrobials tested	Susceptibility patterns		
		Resistant No. (%)	Intermediate No. (%)	Sensitive No. (%)
Gentamicin	292	71 (24.3)	7 (2.4)	214 (73.3)
Tetracycline	236	181 (76.7)	5 (2.1)	50 (21.2)
Chloramphenicol	221	95 (43.0)	3 (1.4)	123 (55.7)
Cotrimoxazole	215	148 (68.8)	4 (1.9)	63 (29.3)
Erythromycin	132	113 (85.6)	1 (0.8)	18 (13.6)
Ciprofloxacin	126	37 (29.4)	3 (2.4)	86 (68.3)
Nitrofurantoin	110	6 (5.5)	–	104 (95.5)
Cephalothin	89	49 (55.1)	5 (5.6)	35 (39.3)
Ceftriaxone	108	52 (48.1)	2 (1.9)	54 (50.0)
Amoxicillin	87	73 (83.9)	–	14 (16.1)
Doxycycline	65	40 (61.5)	2 (3.1)	23 (35.4)
P-value		0.0001		

In this study, the overall resistance rates to two and more antimicrobials was 74.9% and only 23 (7.2 %) were sensitive to all antimicrobials tested. The resistances to two and more antimicrobial agents were 50.7%, 63.0%, 84.6% and 63.6.0% to *E. coli*, *Klebsiella*, *Proteus* and *Pseudomonas*, respectively (Table 5).

4. Discussion

UTIs are one of the most common diseases diagnosed worldwide. Availability of new antimicrobials has improved the management of UTIs. However, the management of UTI infections has been jeopardized by increase in emergence of antimicrobial drug resistance.

The overall isolation rate of uropathogens in this study was 22.7% which is relatively lower than the rates reported from Ethiopia and Nigeria[17,18]. However the rate was higher than other studies[3,5,6]. *E. coli* was the most predominant bacterium isolated from urine, followed by *Klebsiella* spp., *Proteus* spp., *Pseudomonas* spp., CNS, *S. aureus*, *Enterobacter* spp. and *Citrobacter* spp. The isolation rates of

E. coli and other pathogens in this study were comparable to the rates documented previously^[6,12]. However, the rates were generally lower than other reports^[9,10]. Gram negative bacteria were more responsible for UTI than Gram positive bacteria and this finding is in agreement with the findings of previous studies^[4,5]. Difference in identification methods are known to influence the relative prevalence of bacteria which makes comparison difficult^[13]. Bacterial etiologies of UTI can show geographic variations and may even vary over time within a population^[6,18].

Statistically significant difference was observed between genders as majority of the pathogens were isolated from females ($P < 0.001$). Studies conducted all over the world have reported the differences in the prevalence rates between females and males^[4,9]. Physiological and anatomical differences are accounted for the differences in males and females. This is because of the fact that compared to females, the drier environment in the urethra prevents the optimal growth of bacteria. The antimicrobial activity of prostate secretions and longer distance between the anus and urethra meatus are among the factors responsible for the differences in prevalence between the two genders^[19]. The anatomical relationship of the female's urethra and vagina makes it liable to trauma during sexual intercourse as well as bacteria been massaged up the urethra into the bladder during pregnancy and child birth^[20]. Statistically significant association was observed for prevalence of uropathogens among age groups ($P = 0.011$) where uropathogens were more prevalence in reproductive age groups than others. This finding was in agreement with results of a study done in India^[21].

The most frequently isolated bacterial isolates were found to be highly resistant to erythromycin, amoxicillin and tetracycline but sensitive to nitrofurantoin. This result is similar to the result documented in Ethiopia and Nigeria^[21,22]. *Klebsiella* spp., *Proteus* spp. and *Pseudomonas* spp. were found to be resistant to amoxicillin, erythromycin and tetracycline but sensitive to gentamicin and ciprofloxacin. Statistically significant resistance rate was demonstrated to be amoxicillin, erythromycin and tetracycline ($P < 0.001$). These rates are higher than those reported from Ethiopia^[5,6,17] and other countries^[10,23]. Increasing drug resistance to these and other antimicrobials has been documented from previous studies^[10]. Nitrofurantoin was found to be effective against *E. coli*. Gentamicin and ciprofloxacin were effective against other isolates. High rates of sensitivity to nitrofurantoin^[24], ciprofloxacin^[21,24] and gentamicin^[25] have been documented from earlier studies. In this study, resistance to two and more antimicrobial agents was 74.9%. A previous study in Ethiopia has demonstrated a comparable result^[26].

This retrospective study is based on the results of routine microbiological tests carried out from 2003 and 2010. Due to the nature of the retrospective analysis we couldn't trace

patients' clinical settings. Thus the study did not consider such features as inpatient and outpatients, catheterized and non-catheterized patients.

This study showed that the prevalence of UTI was high in all age groups. The most frequently isolate bacterium was sensitive to nitrofurantoin and the other isolates were sensitive to gentamicin. Nitrofurantoin, gentamicin and ciprofloxacin are considered as appropriate antimicrobials for empirical treatment of UTI in the area. Periodic monitoring of etiology and drug susceptibility is recommended.

Conflict of interest statement

We declare that we have no conflict of interest.

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Comments

Background

UTI is one of the most common human bacterial infections around the world both in community and hospital settings. The different forms of antibacterial agents are used to treat the UTIs. In this focus, the continuous 7 years (2003 to 2010) study was done to analyse the data on uropathogens and its antibacterial activity at Dessie Regional Laboratory, Ethiopia.

Research frontiers

The present research work focuses the prevalence and antibacterial susceptibility of eight bacterial pathogens from suspected UTIs against 11 selected antimicrobial agents at Dessie Health Research Laboratory, Ethiopia.

Related reports

In this research data, the reference strains are used as *E. coli* (ATCC 25922) and *S. aureus* (ATCC 25923) based on Clinical and Laboratory Standards Institute.

Innovations and breakthroughs

The information on the uropathogens and its antibacterial activity from different age groups of male and female are

very effective. Because, the collection of data from Dessie Regional Laboratory for the long period of time (2003 to 2010) is very much informative.

Applications

The data on the isolation of uropathogens from different age groups of individuals and antibacterial activity of selected antibiotics are very much useful for the further research in this field. The specific age group may be selected for the collection of urine samples in the future study.

Peer review

The area of data collection by the authors is very much useful. The urine sample ratio is more in female than male. The highest isolation rate of uropathogens was obtained in the age group between 26 to 44 years. The antibiotic resistant pattern was observed only by Kirby–Bauer disc diffusion method.

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