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Research Article

Bacterial Isolates and Drug Susceptibility Patterns of Urinary Tract Infection at Shaheed Monsur Ali Medical College

Akter S[1]* and Kabir MH[2]

[1]Department of Microbiology, Shaheed Monsur Ali Medical College, Bangladesh

[2]Department of Urology, Bangabandhu Sheikh Mujib Medical University, Bangladesh

***Corresponding author:** Sonia Akter, Department of Microbiology, Shaheed Monsur Ali Medical College, Dhaka, Bangladesh

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Abstract

This cross sectional study was conducted to isolate and identify the common bacterial causes of Urinary Tract Infections (UTIs) from urine by culture. A total 2542 urine samples were collected from clinically suspected UTIs patients. Common causative bacteria of UTI were detected by Gram staining, culture in different media, different biochemical tests. Among 324 (12.75%) had significant bacteriuria and the rate of positive culture was 11.48% (232/2020) for female and 17.62% (92/522) for male. In this study, the predominant bacterial isolates were *E. coli* 208 (64.20%) followed by *Proteus* 56 (17.28%). Other predominant bacterial isolates includes *Klebsella* species 30 (9.26), *Pseudomonas* 8 (2.47), *Stap. saprophyticus* 10 (3.09), *Staphylococcus aureus* 6 (1.85) and *Enterococci* 6 (1.85). *E. coli* as the predominant cause of UTI, 82.69% were resistance to Nalidexic and 98.08% sensitive to Amikacin and Imipenem. *Proteus* was resistant to Co-Trimoxazole in 60.7% and *P. aeruginosa* were 100% resistance to ceftriaxone. Among the Gram positive organism, *Stap. saprophyticus* and *Staphylococcus aureus* were 100% resistant to Nalidexic, *Enterococcus* were 66.67% resistant to ciprofloxacin and cefixime.

Keywords: Urinary tract infection; Bacterial isolates; Drug susceptibility; Bangladesh

Introduction

Urinary Tract Infection (UTI) is the commonest bacterial infectious disease in community practice with a high rate of morbidity and financial cost. It has been estimated that 150 million people were infected with UTI per annum worldwide, which costing global economy more than 6 billion US dollars [1]. UTIs is described as a bacteriuria with urinary symptoms [2]. Nearly about 10% of people will experience a UTI during their lifetime [3,4]. UTIs are the most common infections after upper respiratory tract infections [5]. The infections may be symptomatic or asymptomatic, and either type of infection can result in serious sequelae if left untreated [6]. Etiologic agents of UTIs are variable and usually depend on time, geographical location and age of patients. Although UTIs can be caused by any pathogenic organism from the urinary tract, the most frequent is family of Enterobacteriaceae, causing 84.3% of the UTIs [7,8] although several different microorganisms can cause UTIs, including fungi and viruses, bacteria are the major causative organisms and are responsible for more than 95% of UTI cases [9]. Treatment of UTI cases is often started empirically. Therapy is based on information determined from the antimicrobial resistance pattern of the urinary pathogens. However, because of the evolving and continuing antibiotic resistance phenomenon, regular monitoring of resistance patterns is necessary to improve guidelines for empirical antibiotic therapy [10-12]. The aim of this study was to determine the causative agents of UTIs and their susceptibility patterns to commonly used antibiotics in patients from the Dhaka.

Materials and Methods

Study design, area and period

A retrospective study was conducted from April, 2013 to March, 2014 at Shaheed Monsur Ali Medical College and Hospital, Bangladesh.

Study participants and data collection

Urine samples were collected from 2542 outpatients suspected of having a UTI, who had not received antimicrobials within the previous two months, and referred to the Central Laboratory for urine culture. Adult patients were sampled by clean catch midstream urine and children aged under 3 years were sampled using sterile urine bags.

Isolation and identification of organisms

Samples for urine culture were tested within an hour of sampling. All samples were inoculated on blood agar as well as MacConkey agar and incubated at 37°C for 24 hours, and for 48 hours in negative cases. A specimen was considered positive for UTI if a single organism was cultured at a concentration of 10^5 cfu/ml, or when a single organism was cultured at a concentration of 10^4 cfu/ml and 5 leukocytes per high-power field were observed on microscopic examination of the urine. Bacterial identification was based on standard culture and biochemical characteristics of isolates. Gram-negative bacteria were identified by standard biochemical tests [13,14]. Gram-positive microorganisms were identified with the corresponding laboratory tests: catalase, coagulase and esculin agar (for enterococci) [15].

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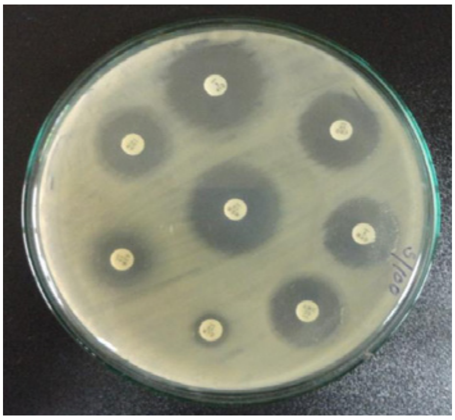

Figure 1: Antibiotic susceptibility pattern in Mueller Hinton agar media by disc diffusion method.

Antimicrobial susceptibility testing

Antimicrobial susceptibility tests were done on Mueller-Hinton agar (Oxoid, England) using disk diffusion method (Figure 1) [16]. Gram positive and gram-negative bacteria sensitivity of isolates to commonly used antimicrobials amikacin (30µg), gentamicin (10µg), ciprofloxacin (5µg), nitrofurantoin (300µg), nalidexic (30µg), cefixime (5µg), cefotaxime (30µg), imipenem (10µg), amoxiclav (30µg), ceftriaxone (30µg) and co-trimoxazole (25µg) were investigated (Oxoid, England). The drug susceptibility pattern was interpreted according to Clinical and Laboratory Standards Institute (CLSI, 2014) (formerly known as National Committee for Clinical Laboratory Standards/NCCLS). Reference strains of *E. coli* ATCC 25922 and *S. aureus* ATCC 25923 were used for quality control for antimicrobial susceptibility tests [17].

Statistical analysis

Discrete variables were expressed as percentages and proportions were compared using the Chi-square test [18].

Results

Isolation and identification of bacteria over a 12-month period, 2542 urine samples from outpatients were analyzed, of which 324 (12.75%) had significant bacteriuria. The rate of positive culture was 11.48% (232/2020) for female subjects and 17.62% (92/522) for male subjects (Table 1).

Gram negative bacilli were responsible for 93.21% of cases followed by Gram-positive cocci, responsible for 6.79% of cases. In this study, the predominant bacterial isolates were *E. coli* 208 (64.20%) followed by *Proteus* 56 (17.28%). Other predominant bacterial isolates includes *Klebsella species*, *Pseudomonas*, *S. saprophyticus*, *Staphylococcus aureus* and *Enterococci*. Majority (74 (22.84%)) of the bacterial isolates were found in 20-29 years age group (Table 2).

Table 1: Result of culture of urine culture in different gender.

Gender	Culture		Total n (%)
	Positive n (%)	Negative n (%)	
Female	232 (71.60)	1788 (80.61)	2020 (79.46)
Male	92 (28.40)	430 (19.39)	522 (20.54)
Total	324 (100)	2218 (100)	2542 (100)

n = number, % = percentage.

The rates of resistance of isolates to a panel of antibiotics, including penicillins, cephalosporins, quinolones, amino glycosides, and Trimethoprim-sulfamethoxazole, which are routinely used to treat UTI infections, are shown in (Table 3). *E. coli* as the predominant cause of UTI, showed the highest percentage of resistance to Nalidexic (82.69%) and the lowest resistance to amikacin (1.92%). *Proteus* as the second most prevalent pathogen of UTI was resistant to Co-Trimoxazole in 60.7% of cases and susceptible to amikacin in 78.57% of cases. *P. aeruginosa* showed 100% resistance to ceftriaxone and 75% to Co-Trimoxazole. In this study, staphylococci were responsible for about 4.94% of UTI cases; among these, *S. saprophyticus* was the most common species isolated. *S. saprophyticus* and *Staphylococcus aureus* were 100% resistant to Nalidexic, Enterococcus were 66.67% resistant to ciprofloxacin and cefixime.

Discussion

This study provides valuable data to compare and monitor the status of antimicrobial resistance among uropathogens to improve efficient empirical treatment. In this study, of 2542 patients from who urine samples were taken, only 12.75% had a urinary tract infection. This is possibly because most of the urine samples were collected from patients had been referred by general practitioners not specialist physicians. These results indicate that urine culture is necessary for a definitive diagnosis of UTI, and that empirical therapy should only be done by specialist physicians in cases where it is necessary.

Our study showed a high prevalence of UTI in females (71.60%) than in males (28.40%) which correlate with other findings which revealed that the frequency of UTI is greater in females as compared to males [19-23]. The reason behind this high prevalence of UTI in females may be due to close proximity of the urethral meats to the anus, shorter urethra, sexual intercourse, incontinence, and bad toilet [24,25].

Although the prevalence of pathogens in different parts of the world is somewhat similar, antimicrobial resistance patterns reported from different regions are significantly different and antimicrobial resistance is increasing. In this study, the Gram negative bacilli constituted 90.47% of the total bacterial isolates while Gram positive cocci constituted 9.26%. This was in agreement with earlier report which states that Gram-positive cocci had a comparatively low contribution in causing UTIs [26]. Out of the Gram negative uropathogens isolated in this study, *E. coli* isolates (70.75%) were the dominant bacterial species isolated from urine cultures which was in agreement with previous works [26,27]. Higher incidence of Gram negative bacteria, related to *E. coli*, in causing UTI has many factors which are responsible for their attachment to the uroepithelium. In addition, they are able to colonize in the urogenital mucosa with adhesions, pili and fimbriae [28].

Worldwide data shows that there is an increasing resistance among UTI pathogens to conventional drugs. Resistance has emerged even to newer, more potent antimicrobial agents [29]. Among the Gram negative organism, the highest percentages of resistance were found for *E. coli* were 82.69% to nalidixic acid and 70% to Trimethoprim-sulfamethoxazole, whereas the highest percentages of susceptibility were seen for amikacin (98.08%); these results are

Table 2: Species of bacteria isolated from urine samples in different age group (n= 324).

			Isolated	Bacteria	No (%)			
Age in years	**Positive Cases**	***E. coli***	***Kleb. Spp.***	***Proteus***	***Pseudo monas***	***Stap. saprophyticus***	***Stap. aureus***	***Enterococcus***
0-9	18 (5.56)	12 (66.67)	0 (0.00)	6 (33.33)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
10-19	44 (13.58)	30 (68.18)	4 (9.09)	10 (22.73)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
20-29	74 (22.84)	40 (54.05)	4 (5.41)	16 (21.62)	4 (5.41)	6 (8.11)	2 (2.70)	2 (2.70)
30-39	46 (14.20)	30 (65.22)	2 (4.35)	10 (21.74)	2 (4.35)	2 (4.35)	0 (0.00)	0 (0.00)
40-49	50 (15.43)	32 (64)	10 (20)	0 (0.00)	2 (4)	2 (4)	2 (4)	2 (4)
50-60	48 (14.81)	36 (75.00)	6 (12.5)	4 (8.33)	0 (0.00)	0 (0.00)	2 (4.17)	0 (0.00)
>60	44 (13.58)	28 (63.64)	4 (9.09)	10 (22.72)	0 (0.00)	0 (0.00)	0 (0.00)	2 (4.55)
Total	324 (100)	208 (64.20)	30 (9.26)	56 (17.28)	8 (2.47)	10 (3.09)	6 (1.85)	6 (1.85)

E. coli = Escherichia coli, *Kleb. Spp = Klebsiella* species, *Stap. saprophyticus = Staphylococcus saprophyticus*, *Stap.aureus = Staphylococcus aureus*.

Table 3: Antimicrobial resistance pattern of bacterial isolates from urine samples of study participants.

Antimicrobial agents	**Bacterial isolates**						
	E.coli	***Klebsiella***	***Proteus***	***Pseudomonas***	***S.saprophyticus***	***S.aureus***	***Enterococcus***
	n= 208	**n= 30**	**n= 56**	**n= 8**	**n= 10**	**n= 6**	**n= 6**
Amikacin	4 (1.92)	6 (20)	12 (21.43)	2 (25)	6 (60)	2 (33.33)	2 (33.33)
Gentamycin	52 (25)	14 (46.67)	14 (25)	2 (25)	0 (0)	4 (66.67)	4 (66.67)
Ciprofloxacin	122 (58.65)	8 (26.67)	20 (35.71)	2 (25)	2 (20)	6 (100)	4 (66.67)
Nitrofurantoin	42 (20.19)	8 (26.67)	14 (25)	6 (75)	0 (0)	0 (0)	4 (66.67)
Nalidexic	172 (82.69)	2 (6.67)	11 (19.64)	6 (75)	10 (100)	6 (100)	2 (33.33)
Cefixime	146 (70.19)	20 (66.67)	37 (66.07)	0 (0)	8 (80)	4 (66.67)	4 (66.67)
Cefotaxim	128 (61.54)	12 (40)	36 (64.29)	6 (75)	8 (80)	4 (66.67)	2 (33.33)
Imipenem	4 (1.92)	2 (6.67)	1 (1.79)	2 (25)	0 (0)	0 (0)	0 (0)
Amoxiclav	72 (34.62)	18 (60)	20 (35.71)	6 (75)	4 (40)	0 (0)	4 (66.67)
Ceftriaxone	124 (59.62)	12 (40)	28 (50)	8(100)	0 (0)	2 (33.33)	2(33.33)
Co-Trimoxazole	146 (70.19)	16 (53.33)	34 (60.71)	6 (75)	6 (60)	4 (66.67)	2 (33.33)

basically in agreement with other studies carried out around the world [30-32]. *Proteus* and *Klebsiella* shows 60.71% and 53.33% resistance to Trimethoprim-sulfamethoxazole. High microbial resistance rate to antibiotics was especially the case for *P. aeruginosa*, which was 100% resistant to ceftriaxone and 75% resistant to amoxyclav and Trimethoprim-sulfamethoxazole; this resistance is higher than that found in other reports [32,33]. This significantly higher bacterial resistance to antibiotics in our region in comparison with other countries seems to be due to a higher rate of antibiotic usage by families, even in the absence of a prescription, and to the high percentage of younger population, since UTIs are more common in the early years of life.

Among the Gram positive organism, *S. saprophyticus* was the prevalent pathogen in our study and was responsible for 3.09% of UTIs in women. *S. saprophyticus* is a pathogen during the period of sexual activity in women. Although sexual transmission has not been defined as the main transmission route of this pathogen, frequent or recent sexual activity is a major risk factor. The division time for this pathogen is longer than that for other UTI pathogens; therefore a lower colony count (100-1000) is of clinical worth [34]. *S. aureus* is an important uropathogen and was responsible for 1.85% of UTI cases in our study. It has been emphasized that any amount of this bacterium should be subjected to antibiogram test [35].

The regional variations of resistance to antibiotics may be explained in part by different local antibiotic practices [36,37]. The influence of excessive and/or inappropriate antibiotic use on the development of antibiotic-resistant strains, particularly broad spectrum agents prescribed empirically, has been demonstrated. Reducing the number of prescriptions of a particular antibiotic can lead to a decrease in resistance rates [38,39]. Transmission of resistant isolates between people and/or by consumption of foods originated from animals that have received antibiotics and greater mobility of individuals worldwide has also contributed to the expansion of antibiotic resistance [40,41].

Conclusion

In conclusion, because the pattern of sensitivity of bacteria to antibiotics varies over time and in different geographical regions, antibiotic treatment of infections should be based on local experience of sensitivity and resistance patterns. In this study, amikacin and nitrofurantoin were found to be the most appropriate antibiotics, for the empirical therapy of UTIs.

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