Original Article

Bacteria Isolated from Urinary Tract Infection among Patients and Determination of the Antibiotic Susceptibility Patterns of the Gram Negative Bacteria in Iran

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Abstract

Background: *Escherichia coli* (*E. coli*) is the most frequent infecting organism in acute infection. So, knowledge about the frequency and distribution of urinary tract infection (UTI) is important to improve infection control measures. The aim of this research was to determine the prevalence of bacteria isolated from urinary tract infection (UTI) in patients and determination of the antibiotic susceptibility patterns of the gram negative bacteria.

Materials and Methods: This descriptive study was performed in Imam Reza hospital, Tabriz (northwest of Iran) during March 2012 to February 2013. We surveyed 8153 patients, who had clinical manifestations of UTI. 5093 (62.47%) of them were female and 3060 (37.53%) of them male. Urine specimens were cultured for isolation of the microbial agents of UTI. The isolated bacteria were identified using biochemical tests. Disk diffusion susceptibility test was used to determine antimicrobial susceptibility.

Results: *E. coli* (55.38%) was the most common isolated pathogen, followed by *Enterobacter spp.* (29.61%), *Pseudomonas spp.* (4.9%), *S. aureus* (3.21%), *Enterococcus spp.* (2.3%), *fungi* (1.5%) and *Klebsiell*a (0.48%). The sensitivity rates of isolated gram negative bacteria were for Amikacin (95.7%), Nitrofurantoin (91.5%), Gentamicin (64.1%), Ceftizoxim (56.8%), Ciprofloxacin (37.6%), Cotrimoxazole (31.4%) and Nalidixic acid (23.5%).

Conclusion: This study showed that the frequency of *E. coli* and *Enterobacter spp*. increases the probability of urinary tract infection. Also this survey indicates the emergence of antibiotic resistant infections in the studied hospital. So, there is a need to improve the effectiveness of integrated infection control programs to control and manage nosocomial infections caused by highly resistant organisms.

Keywords: Prevalence of bacteria, Urinary tract infection, antibiotic susceptibility

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Introduction

Urinary tract infection (UTI) is one of the most prevalent infections. UTI is classified in to uncomplicated and complicated infections with respect to choices for treatment[1]. Among patients referred to

physicians, *Escherichia coli* (*E. coli*) is the most common etiological agent, approximately isolated from 75 to 90% of uncomplicated patients, while complicated ones show a broader bacterial spectrum as the cause of infection[1-3].

In comparison with men, UTI is reported more in women. It could be due to the proximity of genital tract and urethra/anus[4] and anatomical predisposition or urothelial mucosa adherence to mucopolysaccharides lining or other host factors[5]. Urinary tract infection may involve only the lower urinary tract or both the upper and the lower tracts. The term cystitis has been used to describe the syndrome involving dysuria, frequency, and occasionally supra pubic tenderness.

More than 95% of urinary tract infections are caused by a single bacterial species. *E. coli* is the most frequent infecting organism in acute infection[6,7]. *Enterobacter*, *Staphylococci*, *Klebsiella*, *Proteus*, *Pseudomonas*, and *Enterococci species* are more often isolated from inpatients, whereas there is a greater preponderance of *E. coli* in an outpatient population[8]. Hence, the aim of this study was to isolate pathogenic agents involving UTI and to determine their antibiotic susceptibility pattern among patients referred to Imam Reza hospital, Tabriz, Iran.

Methods

Sampling and identification of isolates: A total, 8153 patients with UTI referred to Imam Reza hospital laboratory located in Tabriz city, East Azerbaijan province, northwest of Iran, during March 2012 to February 2013. There were 5093 (62.47%) females and 3060 (37.53%) males, with an age range of 25-80 years (mean, 53.8 years). Urine samples were obtained from a midstream into standardized, sterile container and delivered to the laboratory within 2 hours. Identification was done based on culture characteristics, gram stain and routine standard biochemical tests[9-11]. Guidelines for proper specimen collection were given to all patients on a printed card[10].

Colony count of bacteria in UTI: A measured amount of urine, using calibrated loop method was inoculated on Blood agar medium (Merck, Germany) for colony count. A specimen was considered positive for UTI if a single organism was cultured at a concentration of $\geq 10^5$ cfu/ml, or when a single organism was cultured at a concentration of 10^4 cfu/ml while $\geq$5 leukocytes per high-power field were observed on microscopic examination of the urine[12]. Urine specimens were cultured for isolation of the microbial agents of UTI on Blood agar and MacConky agar media (Merck, Germany).

Antimicrobial susceptibility testing: Antibiotic susceptibility test was carried out on Mueller Hinton agar (Merck, Germany) by Kirby-Bauer's disk diffusion method according interpretive criteria recommended by the clinical and laboratory standards institute (CLSI) guidelines to the fallowing agents[13]: Amikacin (AN 30µg), Nitofurantoin (FM 300µg), Gentamicin (GM 10µg), Ceftizoxim (CT 30µg), Ciprofleoxacin (Cp 5 g), Trimethoprim-sulfamethoxazole (25µg), Nalidixic acid (NA 30µg), and Ampicillin (AM 10 µg). Antibiotic disks used in this research were supplied by Padtan Teb, Tehran, Iran[10]. Appropriate antibiotic discs were tested depending upon whether the organism was gram positive or gram negative. Interpretation of results was done based on the diameter of the zone. *E. coli* ATCC 25922 was used as quality control for antimicrobial susceptibility test.

Statistical analysis: The results were analyzed using SPSS software vs.16 and presented with 95% confident intervals (CI).

Results

8153 patients with clinical symptoms of UTI in this

Table 1: Frequency of bacterial agents isolated from *urine specimens* in this study.

Organisms	No. of isolates (%)
E. coli	1126 (55.38%)
Entrobacter spp.	602 (29.61%)
Pseudomonas aeruginosa	98 (4.9%)
Staphylococcus aureus	65 (3.21%)
Enterococcus spp.	46 (2.3%)
Coagulase-Negative staphylococci[1]	42 (2.06)
Fungi	30 (1.5%)
Klebsiella spp.	11(0.48%)
Proteus spp.	11 (0.54%)
serratia	4 (0.2%)

Table 2: Antimicrobial sensitivity pattern of bacterial agents isolated from urine specimens[a].

Antimicrobial resistance trait of isolates	No. (%)
Amikacin	1949 (95.7)
Nitofurantoin	1862(91.5)
Gentamicin	1304 (64.1)
Ceftizoxim	1155 (56.8)
Ciprofloxacin	765 (37.6)
Trimethoprimsulfamethoxazole	639 (31.4)
Nalidixic acid	478 (23.5)
Tetracycline	407 (20)
Ampicillin	40(2)

a Shown are the numbers and percentages of isolates exhibiting sensitive to the tested antimicrobials.

retrospectively study were investigated (62.47% of them were female and 37.53% were male). 2035 (24.96%) of cases had positive urine culture and 6118 (75.04%) ones did not have significant bacteriuria or bacterial count of their urine samples were very low. Ten types of microorganisms were isolated from positive urine cultures. The most common isolates were *E. coli* (55.38%), followed by *Entrobacter spp.* (29.61%), *Pseudomonas spp.* (4.9%), *S. aureus* (3.21%), (Table 1). The most common isolated uropathogens in Gram-negative bacilli and Gram- positive cocci were *E. coli* (55.3%) and *S. aureus* (3.21%) respectively.

Analysis of the antimicrobial susceptibility profile of the isolates showed that most of isolate (95.7%) were susceptible to Amikacin. This isolate showed resistance to other tested antibiotics. Of 2035 isolates, 98% of the isolates were resistant to Amoxicillin and Ampicillin (n=1995), 80% were resistant to Tetracycline (n=1628), 76.5% were resistant to Nalidixic acid (n=1557) and 62.4% were resistant to Ciprofloxacin (n=1270) (Table 2).

Discussion

Bacterial infection of the urinary tract is one of the common causes for seeking medical attention in the community[14]. Effective management of patients suffering from bacterial UTIs commonly relays on the identification of the type of organisms that caused the disease and the selection of an effective antibiotic agent to the organism in question[15]. In this study, the isolation rate of bacteria from urine was 24.96% which is comparatively lower than reports within the country and other part of the world[16,17], this might have been either due to sample size variation or the studies might have been based on retrospective survey. However, this finding is in line with studies done in Addis Ababa[18] and one from Iran which had a rate of 13.2%[19].

E. coli is the major etiological agent in causing UTI, which accounts for up to 90% of cases[20]. In this study, the most frequent UTI were Gram negatives which made up 80% of all the isolates. *E. coli* is the predominant etiology of UTI, in both outpatients and inpatients of both sexes, and this finding is in agreement with others finding too[16,17,19,21].

Resistance to antimicrobial agents has been noted since the first use of these agents and is an increasing world-wide problem[22]. This study revealed that a higher prevalence rate of resistance to the commonly prescribed antibiotic agent. The finding that 98% of *E. coli* and *K. pneumonia* isolates were resistant to Amoxicillin and Ampicillin is of great importance and implies that these antibiotics cannot be used as empirical therapy for urinary tract infection particularly in the study area. On the other hand, very low levels of resistance were detected to antibiotics such as Nitrofurantoin and Gentamicin and a comparable rate of sensitivity has been reported for these drugs in previous studies done in Ethiopia[16,18,23,24], in Kosovo[17], in Iran[19]. Nitrofurantoin represented better activity against *E. coli* isolates, but this drug would not be recommended for serious upper urinary tract infections or for those cases with systemic involvement[25]. Low resistance was observed for these drugs because they are not easily accessible and relatively expensive in price compared to others. Thus, these drugs could be considered as alternative options in the empirical treatment of UTIs.

Conclusion

In conclusion, the isolation of bacterial Urinary tract infection with a higher resistance rates for commonly used antimicrobials leaves the clinicians with very few options to choose drug used for empirical treatment of UTIs. Therefore, it is important to urge physician and other health worker in the field on the need of re-evaluation of empiric treatment of UTI.

As drug resistance among pathogens is an evolving process, so there is a need to improve the effectiveness of integrated infection control programs to control and manage nosocomial infections caused by highly resistant organisms.

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