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ORIGINAL ARTICLE

Bacteriological profile of urinary tract infections and antibiotic susceptibility of *Escherichia coli* in Algeria

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ABSTRACT

Background and Objectives: Urinary tract infections are one of the most common bacterial infections worldwide. The emergence of antibiotic-resistant bacterial strains is a serious problem and greatest challenge in public health care. The purpose of this study was to determine the prevalence of uropathogenic microorganisms and the antibiotic resistance pattern of *Escherichia coli* in Algeria.

Materials and Methods: Urine samples were collected from 760 outpatients in the hospital of Tizi-Ouzou (Algeria). From the positive cultures, 120 strains of *E. coli* were isolated and tested for their susceptibility to antibiotics by disk diffusion method on Mueller Hinton agar medium.

Results: Among the collected urine specimens, 270 (35.5%) yielded positive cultures for urinary tract infection. Females were more affected with a sex ratio F/M of 1.14. *E. coli* was the most prevalent isolated bacteria with a rate of 44.44%, followed by *Klebsiella pneumoniae* (12.21%), *Pseudomonas aeruginosa* (11.1%) and *Proteus mirabilis* (5.55%). Isolates of *E. coli* showed high level of resistance to cephalothin (85.83%), ticarcillin (82.5%), ampicillin (73.3%) and amoxicillin-clavulanic acid (58.33%). Imipenem was the most effective antimicrobial agent.

Conclusion: These results highlight the inappropriate utilization of antibiotics and suggest the need to improve prescription practices in our country.

Keywords: Urinary tract infection; *Escherichia coli*; Antibiotic resistance; Microbiota

INTRODUCTION

Urinary tract infections (UTIs) are among the most common infections in humans. Approximately, 150 million UTIs occur every year worldwide, resulting in more than 6 billion US dollars in direct healthcare costs (1). These infections are caused by the colonization of uropathogens into the urinary tract. The most common causative agents of UTI are Gram-negative bacteria represented by *Escherichia coli, Klebsiella pneumoniae*, *Proteus mirabilis*, *Enterococcus faecalis*, and *Staphylococcus saprophyticus* (2). The relative frequency of uropathogens varies depending upon age, sex, catheterization, hospitalization and previous antimicrobials exposure (3).

The high incidence of these infections requires often a rapid intervention and the adoption of empirical treatment without knowing the bacteriological profile of the patients. However, knowing the resistance profile of uropathogenic bacteria is important in choosing an adequate treatment. For the past years, drug resistance in bacteria has become a global

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health problem and an emerging threat due to misuse of antibiotics. Hence, it is necessary to be aware of the changes in the spectrum of drug resistance to ensure appropriate treatments (1, 4).

The choice of antibiotics should take into consideration local data of antibiotic resistance patterns. Thus, this work was undertaken in order to assess the most frequent pathogens involved in the appearance of UTI in Tizi-Ouzou (Algeria) and the antibiotic susceptibility of *E. coli* isolates. Even though, there are few published information concerning the etiology and resistance pattern of UTIs in some hospitals of Algeria, there are no previous investigations in the study area.

MATERIALS AND METHODS

Sample collection. Our study was conducted in the laboratory of bacteriology of Tizi-Ouzou Hospital (Algeria) over a period from February to June 2019. In total, 760 midstream urine samples were collected from outpatients presenting for a cytobacteriological examination of urine. Most of the patients presented symptoms suggesting a urinary tract infection. The samples were collected in sterile urine container and analyzed in a period not exceeding 2 hours in order to avoid the overgrowth of bacteria in the urine.

Isolation of bacteria from urine samples. In this work, the urine samples were inoculated on chromogenic medium CHROMagar (CHROMagar Company, Paris, France) for direct identification. Inoculated agar plates were incubated aerobically at 35°C for 48 h. The cultured plates were examined for growth and a sample with at least 10^5 colony forming unit per milliliter (CFU/mL) of urine was considered positive for UTI. Mixed colonies on a plate were inoculated on blood agar and nutrient agar media for the growth of discrete isolates. The identification of the isolated pathogens was based on morphological characteristics and biochemical tests. Gram staining was done for all strains and the smears were examined microscopically for the morphology and staining reactions. Biochemical analysis was performed by the automated VITEK 2 compact system (bio Mérieux, France).

Antibiotic susceptibility test. Overall, 120 strains of *E. coli* were isolated from positive cultures and tested for their susceptibility to antibiotics. The antibiogram was carried out by the disk diffusion method in Mueller Hinton agar medium (Institut Pasteur, Algeria) according to the recommendations of the CLSI guidelines (5). Several commercial antibiotic discs (Bio-Rad; Oxoid) used to treat UTIs were tested including: ampicillin, ticarcillin, cephalothin, cefoxitin, cefixime, ceftazidime, imipenem, amoxicillin-clavulanic acid, piperacillin-tazobactam, amikacin, gentamicin, tobramycin, vancomycin, ciprofloxacin, trimethoprim-sulfamethoxazole, ofloxacin, nitrofurantoin and nalidixic acid. *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 29213 and *Pseudomonas aeruginosa* ATCC 27853 were used as quality control strains.

RESULTS

Patients' characteristics. During this study, 760 urine samples obtained from outpatients with symptoms of urinary tract infection were analyzed. These patients included 500 (65.7%) adults and 260 (34.2%) children (aged less than 17 years). The percentage of male and females was 55.5% (422/760) and 44.5% (338/760), respectively. Among the studied patients, 35.5% (270/760) had a positive urine culture. A rate of 33.12% (113/338) was represented by male patients. In females, the incidence was slightly higher (37.2%, 157/422) compared with males, with a sex ratio F/M of 1.14. In children, 31.11% (81/260) of samples were positive. The frequency of infection in adults was higher with a percentage reaching 69% (345/500).

Prevalence of uropathogens among UTI positive patients. The prevalence of uropathogens isolated from mid-stream urine samples is shown in Table 1. The microbial flora was represented by bacteria (87.04%) and yeasts (12.95%). The fungal isolates obtained in this study were represented by *C. albicans*. In the case of bacteria, a predominance of *Enterobacteriaceae* (65.55%) was observed. Their frequency was higher in females (36.29%) than in males (29.62%). Other species belonging to the *Pseudomonadaceae* (11.1%), *Enterococcaceae* (4.81%), *Staphylococcaceae* (2.59%), *Streptococcaceae* (2.22%) and *Moraxellaceae* (0.74%) were also isolated. Our results showed the prevalence of Gram-negative bacteria as 77.41%. Gram-positive bacteria represented mainly by *Enterococcaceae* were found at a rate of 9.63%. The most frequently isolated species among bacteria was *Esch-*

Table 1. Number and frequency (%) of uropathogens among positive patients.

Isolated micro-organisms	Females No, (%)	Males No, (%)	Total No, (%)
E. coli	66 (24.44)	54 (20)	120 (44.44)
K. pneumoniae	17 (6.3)	16 (5.92)	33 (12.21)
Enterobacter spp.	6 (2.22)	4 (1.48)	10 (3.7)
P. mirabilis	9 (3.33)	6 (2.2)	15 (5.55)
P. aeruginosa	16 (5.92)	14 (5.18)	30 (11.1)
A. baumannii	2 (0.74)	0 (0)	2 (0.74)
S. aureus	3 (1.11)	4 (1.48)	7 (2.59)
Enterococcus spp.	4 (1.48)	8 (2.96)	12 (4.44)
Streptococcus spp.	3 (1.11)	3 (1.11)	6 (2.22)
C. albicans	18 (6.66)	17 (6.29)	35 (12.95)
Total	144 (53.33)	126 (46.64)	270 (100)

erichia coli (44.44%, n=120), followed by *Klebsiella pneumoniae* (12.21%, n=33), *Pseudomonas aeruginosa* (11.1%, n=30), *Proteus mirabilis* (5.55%, n=15), *Enterococcus* spp. (4.44%, n=12), *Enterobacter* spp. (3.7%, n=10), *Staphylococcus aureus* (2.59%, n=7), *Streptococcus* spp. (2.22%, n=6) and *Acinetobacter baumannii* (0.74%, n=2). According to patient sex, our results indicated that 24.44% (66) of *E. coli* were isolated from females compared to 20% (54) from males.

Antibiotic susceptibility test. Antibiotic resistance has become one of the major threats to healthcare in the word. During this study, 177 uropathogenic *Enterobacteriaceae* were isolated. Antibiotic susceptibility testing was performed on 120 isolates of *E. coli* using a standard method of disk diffusion method. The obtained results are summarized in Table 2. Imipenem was found to be a strong agent against *E. coli.* Indeed, none of *E. coli* isolates was resistant to this antibiotic. The tested isolates exhibited high resistance to ampicillin, ticarcillin, amoxicillin- clavulanic acid and cefalotin. Moreover, significant resistance levels (> 50%) to gentamicin and tobramycin, vancomycin, trimethoprim-sulfamethoxazole and offloxacin were observed.

Table 2. Antibiotic-resistance rate (%) of *E. coli* (n= 120) isolated from positive urine cultures against the tested antibiotics.

Antibiotics	Resistance rate n (%)
Ampicillin	88 (73.3)
Ticarcillin	99 (82.5)
Cephalothin	103 (85.83)
Cefoxitin	24 (20)
Cefixime	9 (7.5)
Ceftazidime	38 (32)
Imipenem	0 (0)
Amoxicillin-clavulanic acid	72 (58.33)
Piperacillin-tazobactam	6 (5)
Amikacin	9 (8)
Gentamicin	56 (47.5)
Tobramycin	62 (51)
Vancomycin	60 (50)
Ciprofloxacin	36 (30)
Trimethoprim-sulfamethoxazole	63 (53)
Ofloxacin	55 (46)
Nitrofurantoin	3 (2.5)
Nalidixic acid	39 (32.5)

DISCUSSION

Urinary tract infection is one of the most frequent bacterial infections in the world. Clinical features may range from asymptomatic to severe sepsis. UTIs are caused by the colonization of uropathogens into the urinary tract. Uropathogens colonize through their characteristics like by producing toxins, siderophore, and adhesins. These characteristics support them in colonization and invasion (2).

This study was undertaken to determine the distribution of microbial species isolated from patients with urinary tract infection and the antibiotic susceptibility patterns of *E. coli.*

Out of all analyzed samples, 35.53% were found to be positive for UTI. The observed rate is higher than the levels recorded previously in India (17%) (6), in Italy (20.4%) (7) and in Libya (20.4%) (8). However, it has been found to be similar to the one carried out in Uganda (35%) by Johnson et al. (9). The prevalence of positive UTI was slightly higher among females. Our results differ from those recorded by Salem et al. where the distribution of infection rates was much greater in females (81.4%) than in males (18.6%) (8). UTIs occur more often in women than in men due to anatomical factors (10).

In this work, the predominant microorganisms were represented by Gram-negative bacteria, with a frequency of 77.41%. Our observations corroborate

with previous studies carried out in several countries (6, 11, 12). *E. coli* and *K. pneumoniae* were the most isolated bacterial species. These results are consistent with those published by other authors describing *E. coli* as the predominant uropathogen. Recently, Shaki et al. (13) reported a predominance of *E. coli* and *Klebsiella* spp. in urine samples from Southern Israel. Comparable results were published by Lebanese researchers where *E. coli* was the most isolated microorganism in community-acquired UTI (10). In Libya, Salem et al. (8) indicated *Klebsiella pneumoniae* (43.6%) and *E. coli* (33%) as the two most isolated pathogens. Genus *Klebsiella* is generally associated with nosocomial infections.

The percentage of *E. coli* obtained in our study is a little low when compared with other studies in Brazil (4) and Lebanon (10) indicating rates of 60%, but it remains close in comparison with the one recorded in Ethiopia (48%) (14).

We observed high levels of resistance to ampicillin, ticarcillin and cefalotin among *E. coli*. This may be associated with the high prescribing rate of these antibiotics in the treatment of bacterial infections in Algeria. Indeed, excessive and inappropriate use of broad-spectrum antibiotics by patients is the main cause of the emergence of resistance caused by bacterial mutations (15).

Resistance to antimicrobial agents is an urgent global health challenge threatening the successful prevention and treatment of bacteria, viruses, parasites and fungal infections. The high frequency of infections caused by bacteria resistant to antibiotics makes empiric treatment more difficult. The choice of treatment depends on the microbiological spectrum and the antibiogram of each patient. Most UTIs are generally treated without bacteriological testing. Among the antibiotics used, penicillins (beta-lactams), cephalosporins, fluoroquinolones and carbapenems generally remain the initial choices. However, these broad-spectrum antibiotics should always be replaced by a narrow-spectrum and targeted antibiotic (15, 16).

The present study revealed that imipenem is an effective antibiotic against *E. coli* isolates. The lack of resistance to this antibiotic among *E. coli* isolates could be due to the low prescription in our country. Our results are consistent with a previous work conducted in Algeria where an absence of resistance was found with imipenem (17). Recently, Demir and Kazanasmaz observed low resistance rates of *E. coli* for imipenem with a value of 1.4% (18). However, strong resistance to ampicillin (88.1%) and cefuroxime (73.7%) was found. In a study carried out in Iran, the lowest levels of resistance were identified against imipenem, gentamicin and tetracycline, while no resistance to tobramycin and amikacin was recorded (19).

Relatively high rates of resistance in *E. coli* to ampicillin have already been reported in several studies. Al-Zahrani et al. (15) showed a remarkable resistance of *E. coli* strains to ampicillin (80.5%), trimethoprim-sulfamethoxazole (72.2%), aztreonam (71.4%) and cefalotin (55.9%). Sulfamethoxazole-trimethoprim is an antibiotic widely used in the treatment of UTI but also in the treatment of digestive and respiratory infections (18), which explains the sensitivity rate of *E. coli* found in our study. Usually, antibiotic resistance is significantly higher in developing countries. This can be explained by the misuse of antibiotics without a medical prescription. In fact, developed countries that have adopted stricter control policies to regulate sales have seen decreases in antimicrobial use and resistance rates (20).

CONCLUSION

Through this work, we noticed that the bacterial strains involved in the case of community urinary infections were mainly represented by *E. coli* and *K. pneumoniae*. Besides, high levels of yeasts represented by *C. albicans* were also found. Isolated *E. coli* were highly resistant to a large number of antibiotics, which makes them unsuitable in the treatment of urinary tract infections. Imipenem was completely active against all tested isolates.

In order to prevent the development of resistance to antimicrobial agents in Algeria, the overuse or misuse of antibiotics should be limited and sales control measures must be established. Further research in this field is needed to describe antibiotics' resistance in community-acquired infections and hospital-acquired UTI.

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