Antibiotics Resistance of Isolated Bacteria from Patients with Urinary Tract Infection

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Abstract

From the period of March to thirty August 2015, 210 urine samples were collected from patients with signs and symptoms of UTIs with an age range of 20-60 years, who referred to outpatient department of AlKarama teaching hospital, Baghdad. Out of 210 samples of urine 150(71.43%) were positive culture and 50(23.81%) were negative culture while 10 (4.76%) were showed contaminated urine culture. The common isolated bacteria were *E. coli* 45 (30%), *K. pneumonia* 33(22%)as Gram negative bacteria respectively, while Gram positive bacteria was *Staphylococcus aureus* 42(28%). The other bacteria were *Enterobacter spp*.20 (13.3%), Proteus *spp*. 6(4%), and *Pseudomonas aeruginosa*4 (2.7%). Susceptibility testing were used with the following antibiotic disks: Ciprofloxacin, Amikacin, Nitrofurantion, Trimethoprim-Sulfamethoxazol, Gentamicin, Pipracillin , Nalidixic acid , Tetracycline , Imipenem , Ceftriaxone , and Cefotaxime. The percentage of susceptibility showed variable susceptibility to the antibiotics used. All bacteria showed (100%) susceptible to Imipenem while all bacteria showed (100%) resistant to Tetracycline. *Pseudomonas aeruginosa* isolated bacteria was showed multidrug resistant (MDR); seven antibiotics: Nitrofurantion, Trimethoprim-Sulfamethoxazol, Pipracillin, Nalidixic acid, Tetracycline, Ceftriaxone and Cefotaxime.

Key words: Antibiotics resistance, urinary tract infection (UTIs)

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مقاومة البكتيريا المعزولة من المرضى المصابين بالتهاب المجاري البولية للمضادات الحيوية

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الخلاصة

جمعت 210 عينة ادرار للفترة من شهر نيسان ولغاية شهر اب 2015 من المرضى الذين يعانون من اعراض وعلامات لالتهاب المجاري البولية الذين يراجعون العيادة الخارجية في مستشفى الكرامة التعليمي. تم تشخيص 45 عزلة(30%) لاشيريشيا القولونية و33 عزلة للكليبسيلا الرئوية و42 (28%) عزلة للعنقوديات الذهبية و20 (13.3%)عزلة لجنس البكتيريا المعوية و 6 (4 %) عزلات لجنس البروتيس و 4 (2.7%) عزلات للزوائف الزنجارية.اظهرت العزلات حساسية متباينة للمضادات الحيوية المستخدمة .اظهرت جميع العزلات حساسية للمضاد اميبينيم بينما اظهرت جميع العزلات مقاومة للمضاد تتراسايكلين. اظهرت بكتيريا الزوائف الزنجارية مقاومة متعددة للمضادات الحيوية :سبعة مضادات حيوية: نايتروفيوريشن، تراي ميثوبريم-سلفاميثوكسازول،ببراسلين،نالديسك اسد،تتراسايكلين ،سيفترياكسون و سيفوتاكسيم .

كلمات مفتاحية : مقاومة المضادات الحيوية ـ التهاب المجاري البولية

Introduction

Urinary tract infections (UTIs) are among the most common bacterial infections. It has been estimated that symptomatic urinary tract infections result in as many as 7 million visits to outpatient clinics, 1 million visits to emergency departments, and 100,000 hospitalizations annually [1]. Urinary tract infections have become the most common hospital-acquired infection, accounting for as many as 35% of nosocomial infections [2].Urinary tract infections are common bacterial infections with signs and symptoms or asymptomatic in many subjects who referred to the outpatient departments. Furthermore UTIs become the second cause of bacteremia in patients who stay in hospital for prolong time (nosocomial infection) [1]. Millions people worldwide were diagnosed and treated as urinary tract infections each year and women tend to get infections more than men because the urethra is shorter and closer to the anus [2]. In addition menopause become the risk of increasing urinary tract infection [3]. UTIs are mostly caused by Gram positive ,Gram negative bacteria and *Candida* species that

typically multiply at the opening of the urethra and folding up to the bladder. On the other hand bacteria much less often spread to the kidney from the blood stream [4].Recurrent signs and symptoms of the infection need to repeat the treatment and misuse the antibiotics clinically lead to an increasing in multi-drug resistance bacteria which also causing the spread of bacterial resistant strains, resulting in very high rates of morbidity rather than cost-effective related with its treatment [5]. Untreated infections may be risen to the upper urinary tract and produce fever, chills, increase burning pain on urination, and side pain. The entry of bacteria into the blood stream is associated with severe morbidity، including sepsis and death [6].The early treatment the decreasing UTI rate of morbidity. So an appropriate therapy depending on the main bacteria that involved in the urinary tract infection as well as their corresponding antimicrobial resistance pattern. This way lets controlling the increase of antibiotics resistance and the spread of resistant bacterial strains that characterize as a public health problem worldwide [7]. The aim of this study was to isolate pathogenic bacteria causing urinary tract infection and to determine the prevalence of resistance among these pathogens against antibiotics which used to treat urinary tract infections in patients referred to Al- Karama teaching hospital, Baghdad, Iraq.

Materials and Methods

Sampling

210 samples were collected from patients with signs and symptoms of urinary tract infection, females 137(65.2%) and males73(34.8%), with an age range of 20-60 years (mean, 41.8 years), who referred to outpatient department of Al-Karama teaching hospital, Baghdad, Iraq from 1 / 4 / 2015 to 1 / 9 /2015. The samples were collected using appropriate sterile screw-capped container (5 ml) by clean voided mid-stream technique, labeled and as early as possible brought to the laboratory after good advice to the patient about the collection of urine [8] .The specimens of urine were cultured on blood agar and MacConkey agar plates (Mast group Ltd. U.K.; Biomark Lab., Pune. India).After incubation for 24hrs. at 37° C, the plates were examined to identified and isolated the bacteria which were confirmed by biochemical

tests. Urinalysis was done by examination under microscope for wet mount preparation after mixing the samples by centrifuge, then Gram stain was done [9].

Antibiotics sensitivity

Antibiotics susceptibility testing was performed by the Kirby-Bauer disc diffusion method using Muller-Hinton media (Biomark Lab., Pune. India). The isolates were screened for susceptibility to the antibiotics: Ciprofloxacin (5µg), Amikacin (30µg), Nitrofurantion (300µg), Trimethoprim-Sulfamethoxazol (1.25+23.75 µg), Ggentamicin (10µg), Pipracillin (100µg), Nalidixic acid (30µg), Tetracycline (30µg), Imipenem (10µg), Ceftriaxone (30µg), and Cefotaxime (10µg) [10]. A bacterial suspension was prepared by picking up 1-2 colonies from pure cultures in to 2.5 ml of sterile distilled water. The suspension was spread on Mueller-Hinton Agar plate by sterile swabs in different directions. Antibiotic disks were placed onto the cultures medium surface by sterile pair of forceps. The culture plates were incubated at 37°C for 24 hours; then inhibition zones around the antibiotics disks were measured [11].

Results and Discussion

From 1 / 4 / 2015 to 1 / 9 /2015, a total of 210 urine samples were received and cultured. Out of 210 samples of urine 150 (71.43%) were positive culture and 50 (23.81%) were negative culture while 10 (4.76%) were showed contaminated urine culture (table 1). The distribution of isolated pathogens according to the sex was ranged between 20-60years old, there were 115 (76.7%) females and 35 (23.3%) males with a significant (p 0.05) gender difference (table 2).

Table (1): Distribution of 210 samples of urine culture and their percentage

No of samples	Positive culture	%	Negative culture	%	Contaminated culture	%
210	150	71.43	50	23.81	10	4.76

Table (2): Distribution of 150 patients according to age and sex

Age group(years)	Females No.	%	Males No.	%
20 -29	31	26.9	12	34.3
30 -39	20	17.4	7	20
40 -49	18	15.7	9	25.7
50 -59	22	19.1	5	14.3
60 -	24	20.9	2	5.7
Total	115	76.7	35	23.3

In this study the common isolated bacteria were *E.coli* 45 (30%), *K. pneumonia* 33(22%)as Gram negative bacteria respectively ,while Gram positive bacteria was *Staphylococcus aureus* 42(28%).The other bacteria were *Enterobacter spp*.20 (13.3%), *Proteus spp*. 6(4%), and *Pseudomonas aeruginosa* 4 (2.7%) , (Gram positive bacteria : 28% ; Gram negative bacteria: 72%) (Figure 1) .
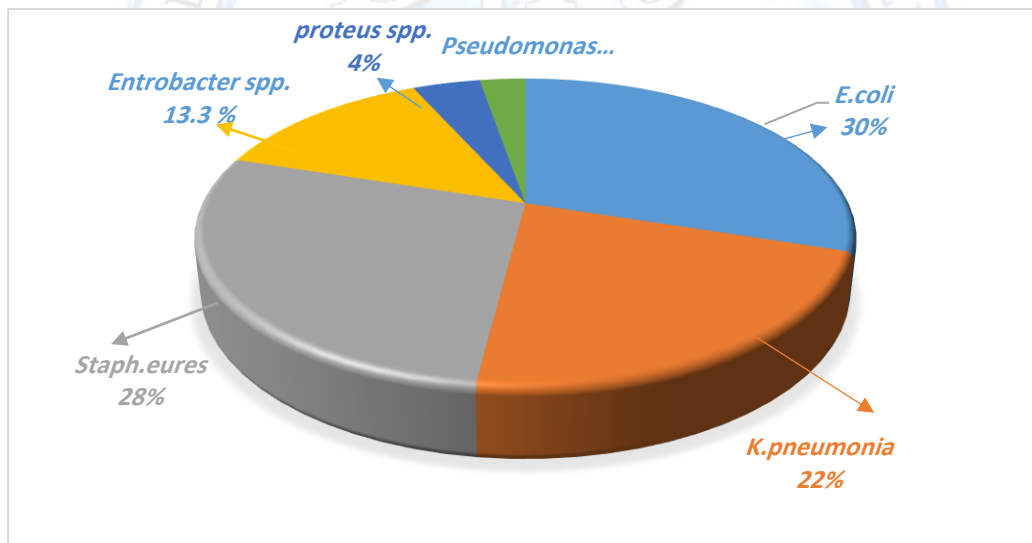

Figure (1): Distribution of the bacteria isolated from urine samples.

All the isolated bacteria were tested for their antibiotic susceptibility against the selected (11) antibiotics table (3) using Kirby-Bauer disk diffusion method by measuring the diameter of inhibition zones around antibiotic discs and compered with NCCLS, [12]. In this study the isolated bacteria showed variable susceptibility to the antibiotics were used in table (3).All bacteria showed (100%) susceptible to Imipenem while all bacteria showed (100%) resistant

to Tetracycline. Furthermore Pseudomonas *aeruginosa* isolated bacteria was showed multidrug resistant (MDR) to Nitrofurantion, Trimethoprim-Sulfamethoxazol, Pipracillin, Nalidixic acid, Tetracycline, Ceftriaxone and Cefotaxime. The highest susceptibility for *E. coli* and *Klebsiella spp* were shown by:Ciprofloxacin, Amikacin (86.7%,84.8%,88.9%,87.8%), Gentamicin (82.2%,78.8,%), Ceftriaxone,Cefotaxime (82.2%,72.7%,84.4%,75.8%) whileTrimethoprim Sulfamethoxazol (68.9%, 81.8%) respectively. The highest susceptibility for *Staphylococcus aureus* were showed by: Ciprofloxacin (76.2%), Amikacin (85.7%), Nitrofurantion (78.6%), Gentamicin (80.9%), Ceftriaxone (71.4%) and Cefotaxime 76.2%). The highest susceptibility for *Enterobacter spp* and *Proteus spp* were showed by: Amikacin (70, 66.7%), Nalidixic acid (65, 83.3%), Ceftriaxone (70%, 83.3%) and Cefotaxime (65%, 66.7%) figure (2) .Urinary tract infections were the common bacterial infections in human which produced by a variety of enteropathogenic bacteria [13,14].In this study there was a statistically predominance of females with urinary tract infection 115 (76.7%) of the (150) positive culture. There were prevalence of infection between females because of the genitourinary abnormalities among males and females, which deal with similar study by Mahsa *et al.,*[15] .There were variable differences between the distribution of bacteria that causes urinary tract infection in this study; Gram-positive (28%) and Gram- negative (72%), this finding was deal with the studies done by many researchers in the world, due to many factors; hygiene methods, education programs, environmental conditions , antibiotic consumption and misuse of antibiotics[16]. This study was showed that *E. coli* the commonest bacteria (30%) among the Gram- negative bacteria followed by *Klebsiella* spp. (22%) while *Staphylococcus aureus* (28%) among the Gram-positive bacteria. These results were deal with another studies like Orret and Davis [17].In this study all isolated bacteria were showed (100%) resistant to Tetracycline, this result was similar to the study reported by Rahem *et al.,*[18], *Pseudomonas aeruginosa* isolated bacteria was showed multidrug resistant . That multidrug resistant may be refereed to several mechanisms including multidrug efflux systems, enzyme production, outer membrane protein (porin) loss and target mutations [19].

Table (3): Antibiotics susceptibility among the bacterial isolates.

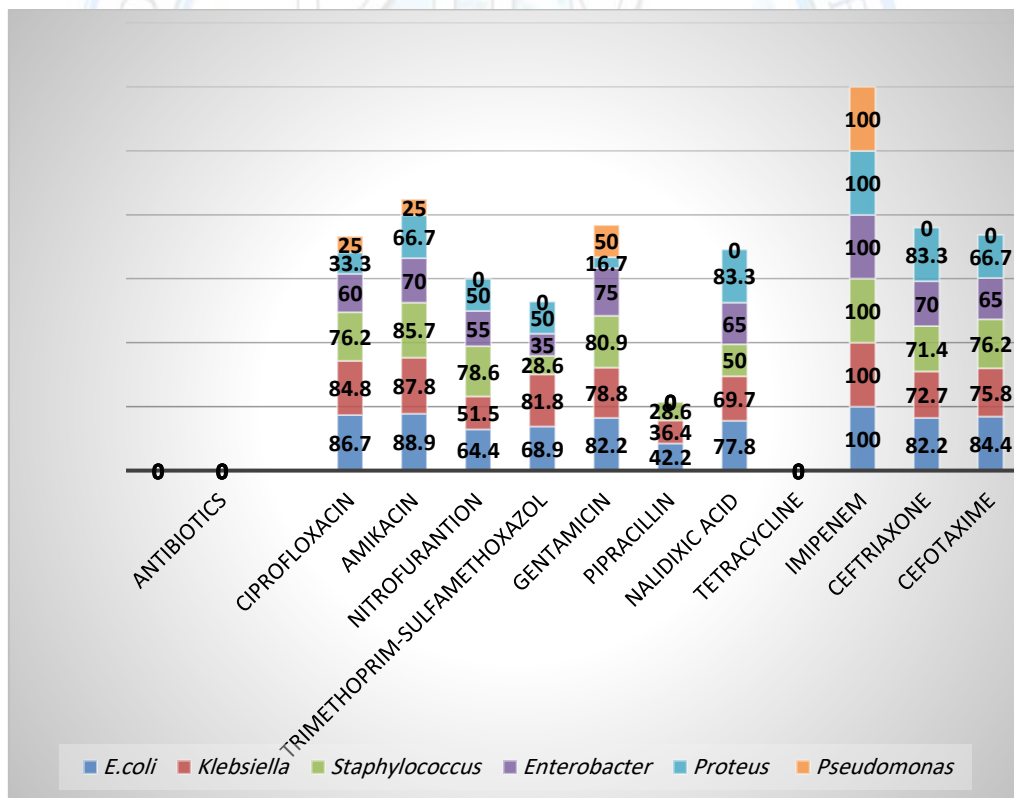| Isolates / antibiotics	E. coli No.45 %	Klebsiella spp No.33 %	Staphylococcus aureus No.42 %	Enterobacter spp No.20 %	Proteus spp No.6 %	Pseudomonas aeruginosa No.4 %
Ciprofloxacin	86.7	84.8	76.2	60	33.3	25
Amikacin	88.9	87.8	85.7	70	66.7	25
Nitrofurantion	64.4	51.5	78.6	55	50	R
Trimethoprim-Sulfamethoxazol	68.9	81.8	28.6	35	50	R
Gentamicin	82.2	78.8	80.9	75	16.7	50
Pipracillin	42.2	36.4	28.6	R	R	R
Nalidixic acid	77.8	69.7	50	65	83.3	R
Tetracycline	R	R	R	R	R	R
Imipenem	100	100	100	100	100	100
Ceftriaxone	82.2	72.7	71.4	70	83.3	R
Cefotaxime	84.4	75.8	76.2	65	66.7	R

Figure (2): antibiotics susceptibility among the bacterial isolates

Conclusion

In conclusion, all bacteria showed (100%) susceptible to Imipenem while all bacteria showed (100%) resistant to Tetracycline. Also *Pseudomonas aeruginosa* showed multidrug resistant (MDR) to Nitrofurantion, Trimethoprim-Sulfamethoxazol, Pipracillin, Nalidixic acid, Tetracycline, Ceftriaxone and Cefotaxime. The highest susceptibility for *E. coli* and *Klebsiella spp* the antibiotics Ciprofloxacin, Amikacin, Gentamicin, Ceftriaxone, Cefotaxime and Trimethoprim-Sulfamethoxazol respectively.

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