

Research Article**Isolation and Sensitivity of Bacteria Caused Urinary Tract Infections at Wasit Province****Enass A. Abed**

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Abstract: One hundred - twenty samples collected from patients infected with acute and chronic urinary tract infection in different hospitals of Wasit Province. Seventy five bacterial isolates, which were then, diagnosed using the biochemical and API 20 tests. These bacteria were diagnosed as *Escherichia coli* (25%) *Klebsiella pneumoniae* (10.6%), *K. oxytoca* (4%), *Enterobacter aerogenes* (9.3%), *Pseudomonas aeruginosa* (8%), *Proteus mirabilis* (5.3%), *Serratia marcescens* (4%), *Acintobacter baumannii* (2.6%), and *Citrobacter freundii* (1.3%). The positive isolates of the gram stain represented *Staphylococcus aureus* (12%), *Staphylococcus epidermidis* (6.6%), and *Enterococcus fecalis* (2.6%). The sensitivity of the isolates of 13 antibiotics was tested. The results showed a variance as far as their resistance to these antibiotics. Imipenem is the most effective antibiotic on the studied bacteria isolates. On the other hands, bacteria isolate showed high resistance to Penicillins and Cephalosporins antibiotics represented Cefotaxime (62%), Cephalexin (74%), Amoxicillin (77%), and Piperacillin (64%).

Keywords: Bacteria, UTI, Antibiotics

INTRODUCTION

Urinary Tract Infections (UTIs) are one of the most prevalent extra intestinal bacterial infections. Nowadays, it represents one of the most common diseases encountered in medical practice affecting people of all ages from the neonate to the geriatric age group [1].

Worldwide, about 150 million people are diagnosed with UTI each year [2]. Most infections are caused by retrograde ascent of bacteria from the faecal flora via the urethra to the bladder and kidney especially in the females who have a shorter and wider urethra and is more readily transfer by microorganisms [3].

The more kinds of common gram negative bacilli (Enterobacteriaceae), such as bacteria *E. coli*, it caused alone (80-85%) of urinary tract infections, the percentage of hospital-acquired (50%), and are second *Klebsiella spp* *Proteus spp* and *Pseudomonas spp* and gram positive bacteria, such as *Staphylococcus saprophyticus* and *Enterococcus spp* as the rest of the injuries are caused at least isolation from other causes [4]. Majority of UTIs are not life threatening and do not cause any irreversible damage. However, when the kidneys are involved, there is a risk of irreparable tissue damage with an increased risk of bacteremia [5]. Nowadays, drug resistance is a huge growing problem in treating infectious diseases like malaria, tuberculosis

(TB), diarrheal diseases, urinary tract infections (UTIs) etc. As suggested by Goldman and Huskins [6] the improper and uncontrolled use of many antibiotics resulted in the occurrence of antimicrobial resistance, which became a major health problem worldwide. In the past decade, many kinds of resistant strains have been discovered. For example, methicillin resistant *Staphylococcus aureus* (MRSA) [7], multidrug resistant *Pseudomonas aeruginosa* [8] and *Serratia marcescens* [9], vancomycin resistant enterococci (VRE) [10] and extended spectrum beta lactamase (ESBL) resistant enterococci [11]. The present study aimed to diagnose the etiological agents of urinary tract infections and their sensitivities at Wasit Province.

MATERIALS AND METHODS**Sample collection**

A total of 120 midstream urine samples were collected from patients infected with acute urinary tract infection in different hospitals at Wasit province, during one month March, 2011. The samples were analyzed using the standard bacteriological media like blood agar, Mannitol salt agar and MacConkey agar and incubated at 37°C for 24-48 hours. All the bacterial isolates were characterized and identified by API system (API 20E, API Staph and API 20-strept) and studied their cultural and morphological features from the results of Gram staining and biochemical tests such as catalase, coagulase, motility, oxidase, Indole,

Methyl-Red, Voges-proskauer, citrate utilization, urease, carbohydrate oxidation/fermentation etc. described by Cowan [12].

Antimicrobial susceptibility

Antimicrobial sensitivity testing of all isolates was performed on diagnostic sensitivity test plates by the Kerby Bauer method [13] following the definition of the National Committee of Clinical Laboratory Standards (NCCLS, 1999) [14]. Bacterial inoculums were prepared by suspending the freshly-grown bacteria in 25 mL sterile nutrient broth.

A sterile cotton swab was used to streak the surface of Mueller Hinton agar plates. Filter paper disks containing designated amounts of the antimicrobial drugs obtained from commercial supply firms (Himedia Labs, Mumbai, India) were used. The antimicrobial agents tested were Amoxicillin (10 µg), Cephalexin (30 µg), Cefotaxime (30 µg), Ciprofloxacin (5 µg), Norfloxacin (10 µg), Nitrofurantoin (300 µg), Amikacin (30 µg), Gentamicin (30 µg), Augmentin (Amoxicillin/clavulanic acid) (20/10 µg), Imipenem (10 µg), Trimethoprim (SXT) (5 µg), Piperacillin (100 µg), and Aztreonam (30 µg).

RESULTS AND DISCUSSION

A summary of the different microorganisms isolated during the study period was shown in Table 1. It is clear that *E. coli* was the predominant uropathogen (33%) causing UTI, followed by *Staphylococcus aureus* (12%), *Klebsiella pneumoniae* (10.6%), *Enterobacter aerogenes* (9.3%), *Pseudomonas aeruginosa* (8%), *Staphylococcus epidermidis* (6.6%), *Proteus mirabilis* (5.3%). However, *Enterococcus faecalis*, *Acinetobacter baumannii*, *Citrobacter freundii*, *K. oxytoca* and *Serratia marcescens* were the least dominant uropathogen causing UTI strains. The sensitivity of the isolates for 13 antibiotics was tested.

According to this result, Major isolates in UTI were *E. coli*, followed by *S. aureus*, *Klebsiella pneumoniae*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Proteus mirabilis* and *Staphylococcus epidermidis*. These observations were supported by several studies conducted previously. According to Goswami *et al.* [31] reports indicate *E. coli* as the most common organism (64.3%), followed by *S. aureus* (21.4%) and *Klebsiella pneumoniae* (14.3%).

The results showed a variance as far as their resistance to these antibiotics. Imipenem is the most effective antibiotic on bacteria isolates (gram negative and positive), Chart A showed percentages resistance bacteria of antibiotics. The percentages of resistance of all isolates to the antimicrobial agents were: 62% to Cefotaxime, 74% to Cephalexin, 77% to Amoxicillin (AX), 64% to Piperacillin, 50% to Aztreonam, 43% to Gentamicin, 31% to Amikacin, 24% to Ciprofloxacin, 28% to Norfloxacin, 62% Augmentin, 40%

Trimethoprim (SXT), 44% to Nitrofurantoin and 4% to Imipenem.

The present study evaluated the prevalence of microgram implicated in UTI to ascertain their antimicrobial resistance patterns. The results of resistance to antibiotics that bacterial isolates all have resistance to most antibiotics used in this study and the various rates. The findings of this study compared favorably to Al-Harhi and Al-Fifi [15] has reached an increase in resistance bacteria isolated from urinary tract infections to antibiotics.

Some antibiotics in this study showed the effectiveness of relatively high to bacteria isolated such as quinolones antibiotics Ciprofloxacin and Norfloxacin and Aminoglycosides antibiotics Gentamycin and Amikacin and Conversely, which showed a number of antibiotics weak effective, such as Amoxicillin, Cephalexin, Cefotaxime, Piperacillin and Augmentin were characterized by the rest of the antibiotics effective medium against bacteria isolates which is (Nitrofurantoin and Trimethoprim + Sulphamethoxazole). The Carbapenems Imipenem comes first antibiotics in the treatment of urinary tract infections as one of the antibiotics and wide-ranging in its impact in many bacterial species that cause UTI and have few side effects [15].

The results of the current study, most bacteria isolates were resistant to the group Penicillins such as Amoxicillin and Piperacillin was the proportion of their resistance (77%) and (64%) respectively of the total isolates, either Cephalosporins groups such as Cephalexin and Cefotaxime was the proportion of their resistance (74%) and (62%) respectively of the total isolates, working these antibiotics to inhibit the process of protein synthesis of cell wall of bacteria through the interaction with the manufacturing process layer Peptidoglycan, and perhaps reasons for this resistance to the secretion of bacterial enzyme B-lactamase which by cleavage rang B-lactam of penicillins and cephalosporins [16, 17]. The results of this study are compatible with the findings of Ahmad [18] in that most isolates were resistant to Penicillins such as Piperacillin and Amoxicillin.

The Aztreonam of B-lactam antibiotics novel, overall resistance (50%) of the total isolates, one can say that most of the isolates local under study possessed the status of resistance to B-lactam antibiotics of a group Penicillins (Amoxicillin and Piperacillin) and cephalosporins (Aztreonam, Cefotaxime) and B-lactam antibiotics novel (Aztreonam), possibly due to the fact that these antibiotics sensitive to B-lactamase enzymes released of *P. mirabilis* and *K. pneumoniae* and *E. coli*, and the rest of the isolates or to the lack of affinity of antibiotics link protein responsible for the strength of cell-wall associated proteins called Penicillins (Penicillin binding Proteins) [19].

The reason for the moderate resistance isolates under study for Aztreoname may be due to the production of extend spectrum B-lactamase enzymes (ESBLs) working alone or with B-lactamase enzymes encoded with plasmid and diffusion among the many gram negative bacteria and that work on antimicrobial resistance B-lactam antibiotics such as cefotaxime and cefazidime and Aztreoname [32]. As for Augmentin the resistance percentage (62%) of the total isolates and is combination of enzyme inhibitor (Amoxicillin + Clavulanic acid), and the reason for the resistance to the production of bacterial B-lactamase enzymes stimulating by chromosomes that not inhibition by Clavulanic acid [20]. The enzymes Clavulanic acid resistance is TEM-1 and SHV-5, as well as the presence of AmpC enzymes that are responsible for multiple resistances to antibiotics and also has an important role in resistance to these antibiotics [21].

The results of this study are compatible with the results of Subha *et al.* [22] who found that (90%) of isolates were resistant to Augmentin, and interpreted the reason for high resistance against inhibitors B-lactamase enzymes to production of higher enzyme B-lactamase enzymes which makes all the inhibitors (Sulbactam and Tazobactam and Clavulanic acid) is suitable for treatment, because the isolates that produce enzymes B-lactamase enzymes naturally be sensitive to (B-lactams antibiotics enzymes + inhibitor), while strains of multi-resistance produces enzymes B-lactamase enzymes 5 times more natural, therefore, be multi-resistance. and that there are other reasons for not working inhibitor is the lack of stability, and evidence of inhibitor during the storage period (according to manufacturer).

As for the resistance to antibiotic (Trimethoprim + Sulphamethoxazole), the ratio of a resistance (40%) of the total isolates, that the reasons for high resistance to isolates of Trimethoprim under study could be due to one of the following mechanisms [23].

A- Increase the production of an enzyme (DHFR) Dihydrofolatereductase.

B- Mutation in gene responsible for enzyme (DHFR).

C- The acquisition of bacteria of the gene (*dfr*), which encodes for the enzyme (DHFR), is resistant to the effect of antibiotic.

The resistance of bacterial isolates under study for Nitrofurantoin reached (44%), and due to its wide use, and also affects the pH of medium in effectiveness of antibiotic because its effectiveness increases when pH: 5.5 or less [24]

The resistance of bacterial isolates under study for Quinolones antibiotics which included Ciprofloxacin and Norfloxacin were proportion of resistance (24%) and (28%), respectively of the total isolates under study, that cause of resistant isolates under study for Quinolones antibiotics used could be due to a change in the target site for a link to antibiotics on enzyme, as it even in the change (GyrA), one of the structural blocks of an enzyme (DNA gyrase) [23].

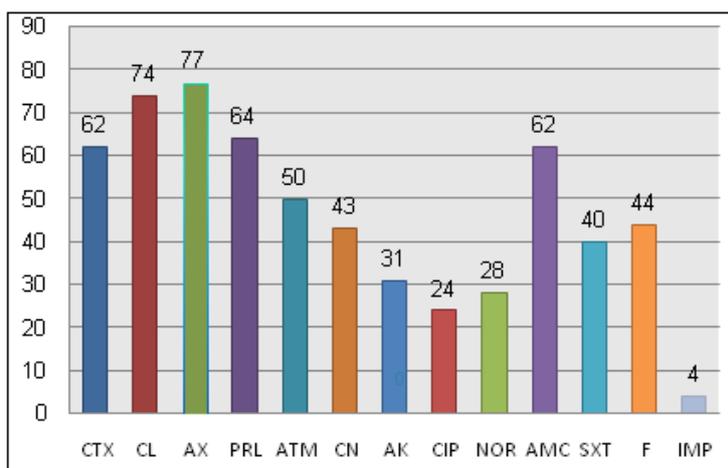
While the antimicrobial resistance group Aminoglycoside and involved in Gentamycin, Amikacin and that the ratio of their resistance (43%) and (31%), may be attributed cause of bacterial resistance to antibiotics Aminoglycoside three mechanisms: modification by enzymes modified such as Adenylating, Phosphorylating Acetylating or mutation such as chromosomal mutation in the gene coding for the target protein in under small unit ribosome 30S, causing the loss of affinity to link target protein and reduce the permeability of bacterial cell of the antibiotic [25].

As for Imipenem who belongs to the group Carbapenems showed isolates sensitive large it was rate of resistance (4%) only attributed the cause of the resistance has to developments in the mechanisms of resistance of bacteria such as the production of enzymes Carbapenemases that belong to the enzyme B-lactamases class D and B as well as *bla*OXA-23 genes which coding for resistance to this antibiotic [26, 27].

All the isolates in this study showed resistance to at least 5 different antibiotics, indicating the presence of strong selective pressures from the antibiotics in the community. Brown *et al.* [28] have reported that horizontal gene transfer is a factor in the occurrence of antibiotic resistance in clinical isolates and suggested that the high prevalence of resistance to a particular antibiotic does not always reflect antibiotic consumption as previously suggested by Nwanze *et al.* [29]. According to Mandal *et al.* [30] reports from India, *E. coli* as the commonest cause of UTI and antibiotic resistance was high among the strains, which emphasize the need for judicious use of antibiotics. Certain virulence factors like haemolysin production and presence of fimbriae in the *E. coli* may be associated with urovirulence. Moreover, these differences in sensitivity pattern of the isolates could be attributed to time difference between the two studies or environmental factors such as practices of self-medication, the drug abuse and indiscriminate misuse of antibiotics among the general population, which has favored the emergence of resistance strains.

Table 1: Percentage of UTI isolate among the pathogens

Sl. No.	Isolates	Number of Isolates	Percentage
1.	<i>Escherichia coli</i>	25%	33%
2.	<i>K. pneumoniae</i>	8%	10.6%
3.	<i>K.oxytoca</i>	3%	4%
4.	<i>Enterobacter aerogenes</i>	7%	9.3%
5.	<i>Pseudomons aeruginosa</i>	6%	8%
6.	<i>Proteus mirabilis</i>	4%	5.3%
7.	<i>Serralia marcescens</i>	3%	4%
8.	<i>Acinetobacter baumannii</i>	2%	2.6%
9.	<i>Citrobacter freundii</i>	1%	1.3%
10.	<i>Staphylococcus aureus</i>	9%	12%
11.	<i>Staphylococcus epidermdis</i>	5%	6.6%
12.	<i>Enterococcus fecalis</i>	2%	2.6%

**Fig. 1: Showed percentages Resistance bacteria of Antibiotics****REFERENCES**

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