

BACTERIAL PATHOGEN IN URINARY TRACT INFECTION AND ANTIBIOTIC RESISTANCE PATTEERN IN ZARAQA -JORDAN

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Abstract

This study aimed to isolate the pathogenic agents involving Urinary Tract Infections (UTI) in Zarqa city and to evaluate their antibiotic resistance pattern. Bacterial isolation and identificaction was conducted using standard methods. From 3756 urine sample collected from patients with UTI symptoms 392 (10.4 %) show positive urine cultures of which 318 (81 %) were females and 74 (19 %) were males. Among the isolated pathogen *E. coli* was the most frequent bacteria (80.8%) folloewd by *Klebsiella* spp. (11.7%), while *Staphylococcus aureus* was the least frequent bacteria (0.08%). All the isolated pathogens resist one or more antibiotic, *E. coli* and *Klebsiella* spp possessed high resentence to ampicillin (84.18 and 89.13%) respectively. One *E. coil* (0.32) isolate showed resistant against all tested antibiotics. Gram-negative bacteria were the major isolates in this study, and *E. coli* was the predominant isolated bacteria. The knowledge of antibiotic sensitivity pattern in this study will be helpful for the effective treatment of the UTI in this population.

Keywords: Urinary Tract Infection, Antibiotic Resistance, Uropathogens

Intorduction

Urinary tract infection (UTI) is a common disease that defined as the presence of infectious bacteria within the urinary system (Zelikovic *et al.*, 1992). UTI is the most widespread infectious disease after respiratory tract infection in most community. It considered as a major public health problem due to the increased costs with an estimated 150 million cases per annum

worldwide (Arjunan *et al.*, 2010). Uncomplicated UTIs are ones that diagnosed in a normal genitourinary tract with no former instrumentation, while “complicated” infections are those diagnosed in genitourinary tracts that have structural or functional abnormalities including instrumentation like indwelling urethral catheters, and are frequently asymptomatic UTI (Gonzalez & Schaeffer, 1999).

Different bacteria responsible for UTI including both gram positive and gram negative bacteria. Among the gram negative bacteria *Escherichia coli* cause approximately 75% of UTIs worldwide (Beyene & Tsegaye, 2011). *E.coli* has recognized virulence factors which aid in the persistence of bacteria in the urinary tract and induce inflammation. Such factors include the presence of pili or K antigen in bacterial capsule, fimbriae, haemolysin and colicin production and the ability to acquire iron etc.(Rushton, 1997). *Klebsiella*, *Staphylococci*, *Enterobacter*, *Proteus*, *Pseudomonas* and *Enterococci* species are more often isolated from penitents show UTIs symptoms (Bronsema *et al.*, 1993). Some reports mentioned that coagulase negative *Staphylococci* are a common cause of urinary tract infection. *Staphylococci saprophyticus* tends to cause infection in young women of a sexually active age (Schneider & Riley, 1996). There is an notable increase in the prevalence of antimicrobial resistance among pathogens that cause UTI worldwide due to extensive misuse of antibiotics in practice (Grude *et al.*, 2001)

Although there were many studies on UTIs and the resistance patterns of pathogens cause these infections conducted in Jordan, it's necessary to reevaluate the resistant pattern of these pathogens. This study is important for clinicians in order to facilitate the empiric treatment of patient with symptoms of UTIs. Moreover, the data would also help the authorities to formulate antibiotic prescription policies. The aims of this study were isolation of the pathogenic agents involving UTI in Zarqa city and determination of their antibiotic resistance pattern.

Materials and Methods

Data Collection

The microbiological and antibacterial susceptibility data of this study have been obtained from the records of clinical microbiology laboratory of Prince Hashim Hospital/Zarqa. These data were filled in a prepared data sheet during the period between January to August 2014. However, clinical microbiology laboratory of Prince Hashim Hospital use the following procedure for culturing, bacterial identification and susceptibility testing.

Urine Specimens and Primary Inoculation

The Urine samples were cultured on 5% blood agar, MacConkey, and Eosin-Methylene blue (EMB) agar using calibrated loops for semi-quantitative method (Beyene & Tsegaye, 2011) and incubated in both aerobic and anaerobic conditions for 24 hours at 37°C. Cultures without any colony at the end of 24h incubation were further incubated for 48h. Samples with colony count equal or more than 10^5 Cfu/ml were considered positive. The isolates were identified and confirmed using standard methods including: Gram staining, colonial morphology on media, growth on selective media, lactose and mannitol fermentation, H₂S production, catalase, oxidase, coagulase, indole, and citrate utilization, and urease test (Oluremi *et al.*, 2011).

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed on Mueller-Hinton agar using disk diffusion technique according to Clinical and Laboratory Standards Institute (CLSI) guidelines. The antibiotic discs and their concentrations consisted of Cefatoxime (CTX, 30µg), Cephalothin (CF, 30µg), Gentamicin (GM, 10µg), Ampicillin (AM,10µg), Nitrofurantoin (FM, 50µg), Norfloxacin (NOR, 10µg), Cotrimoxazole (SXT 25 µg), and Nalidixic acid (NA, 30µg) for Gram negative isolates, and Pencillin (PEN, 10µg), Erythromycin (E, 15µg), Gentamicin (GM, 10µg), Vancomycin (V, 30µg), Lincomycin (LM, 10 µg), Teicoplanin (TEC, 30µg) and Cloxacillin (CL, 30µg) for Gram positive isolates.

Results

During the period between January to August 2014, a total of 392 positive urine cultures of 3756 sample were studied. There were 318 (81%) females and 74 (19%) males. The data showed that the majority of uropathogen isolates were *Escherichia coli* (80.8%), *Klebsiella* spp. (11.7%), *proteus* spp. (4.1%), *Pseudomonas* spp (2.6%) and *Staphylococcus aureus* (0.08%) table (1).

Table (1): Pathogen isolates from patients with UTI.

Bacteria	Gender		%		
	Male	Female	Male	Female	Total
<i>Escherichia coli</i>	54	263	13.77	67.09	80.86
<i>Klebsiella</i> spp	14	32	3.57	8.16	11.73
<i>proteus</i> spp	1	15	0.25	3.83	4.08
<i>Pseudomonas</i> spp	2	8	0.51	2.04	2.55
<i>Staphylococcus aureus</i>	2	1	0.51	0.25	0.76

The distribution of Antibiotic resistance amongst the bacterial isolates (table 2) showed that, *E. coli* which was the predominant isolate gave high resistance to Ampicillin 84.18%, Cotrimoxazole 70.89% and Nalidixic acid 66.77%. *Klebsiella* spp, the second most isolated organism, showed high resistance to Ampicillin 89.13%, Cephalothin 58.70% and Cotrimoxazole 54.34%. The Gram positive bacteria *Staphylococcus aureus* was showed high resistance to Erythromycin 100%, and Pencillin 66.67%. table (2).

Table (2): Distribution of Antibiotic resistance amongst the bacterial isolates (%)

Antibiotic	Pathogen				
	<i>Escherichia coli</i>	<i>Klebsiella</i> spp	<i>proteus</i> spp	<i>Pseudomonas</i> spp	<i>Staphylococcus aureus</i>
CTX	41.46	50	31.25	30.0	-
CF	55.06	58.70	50	50.0	-
GM	26.58	17.39	18.75	40.0	0
AM	84.18	89.13	62.5	40.0	-
FM	10.44	52.17	68.75	40.0	-
NOR	34.81	15.21	31.25	10.0	-
SXT	70.89	54.34	50	40.0	-
NA	66.77	45.65	37.5	40.0	-
PEN	-	-	-	-	66.67
E	-	-	-	-	100
LM	-	-	-	-	33.33
V	-	-	-	-	0
TEC	-	-	-	-	0
CL	-	-	-	-	33.33

The percentage of the isolates showing multidrug resistance is given in Table 3. Only one *E. coli* isolate (0.32 %) was resistance to all the antibiotics tested against gram negative bacteria, while 18.04 % of the *E. coli* isolates resist 6 of the tested antibiotics. The isolated *Klebsiella* spp resist at least one antibiotic and 19.57% of these isolates resist 2, 3, or 6 antibiotics. The gram positive bacteria *S. aureus* is the most susceptible isolated pathogen it resist only 1 and 3 antibiotics with 33.33% and 66.67 %, respectively.

Table (3): Proportion (%) of the isolates resistance to a number of antibiotics.

Bacteria	Number of antibiotic resisted								
	0	1	2	3	4	5	6	7	8
<i>Escherichia coli</i>	6.96	2.23	12.66	15.50	13.60	13.92	18.04	10.76	0.32
<i>Klebsiella</i> spp	0	10.87	19.57	19.57	13.04	8.70	19.57	8.70	0
<i>proteus</i> spp	6.25	18.75	12.50	12.50	12.50	12.50	18.75	6.25	0
<i>Pseudomonas</i> spp	10.00	30.00	20.0	20.0	10.0	10.0	0	0	0
<i>Staphylococcus aureus</i>	0	33.33	0	66.67	0	0	0	0	0

Discussion

This study was conducted to evaluate the distribution of uropathogen species isolated from patients with UTI at a Prince Hashim Hospital in Zarqa, Jordan and their antibiotic resistance pattern. Furthermore, it describes the relationship between sex and isolated pathogens. UTI is one of the most common infections worldwide, and the antibiotic resistance pattern of pathogens isolated from urine varies in different regions. In this study, only 392 (10.4 %) of the 3756 collected samples show positive urine cultures, higher results were found in other studies (Khoshbakht *et al.* 2013, Inabo & Obanibi, 2006), this may refer to the accuracy in determination of the signs of patients with UTI. The high incidence of UTI among the females 318 (81 %) than the males 74 (19 %) is due to anatomic and physical factors related to females (Al Sweih *et al.*, 2005). The sex distribution of patients in this study was consistent with those in other studies (Al-Mardeni *et al.*, 2009; Alshara, 2011).

This study showed that More than (80 %) of urinary tract infections are caused by a single bacterial species *E. coli* followed by *Klebsiella* spp. (11.7%), this results showed similarity to findings of many studies in Jordan and elsewhere that reported *E. coli* and *Klebsiella* spp as the leading UTI pathogens (Murshidi & Farah , 2002; Orrett, 2003; Al-Mardeni *et al.*, 2009; Alshara, 2011).

The present study showed that, *E.coli* 89.13% and *Klebsiella* isolate possessed a high level of resistance to AMP (84.18 % and 89.13, respectively). This is differ to previous studies that reported moderate level of resistance against this antibiotic (Sahm, *et al.*, 2001). *E. coli*, the predominant pathogen isolated from UTI patient in this study showed highest susceptibility to FM (89.56 %). This finding was differ from the results reported by Munjanath *et al.* (2011). The antibiotic resistance pattern differences among the isolated pathogen may be related to different factor related to the isolated pathogens such as molecular and physiological factors.

Conclusion

It can be concluded that Gram-negative bacteria were the most causative pathogen responsible for UTIs. The most common isolated bacteria was *E. coli*, and most effective antibiotic against this bacteria was FM, while NOR was the most effective against other gram negative isolates. This study has been conducted to reevaluate local prevalence of causative pathogens and their antibiotic resistance pattern in Zarqa/Jordan. The knowledge of antibiotic resistant pattern helps in applying appropriate empirical treatment thus reducing drug resistance in uropathogens.

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