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Isolation and Diagnosis of Bacteria from Women with Urinary Tract Infection and Study of Antibiotic Susceptibility

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ABSTRACT

Urinary tract infection (UTI) is an inflammation that results from the presence of a number of opportunistic bacteria that make their way to the urinary tract. When untreated, they may reach the kidneys and cause damage to them. Therefore, this study aims to isolate the negative bacteria of Gram stain, the most common bacteria in UTI, and study their susceptibility to antibiotics. The study sample consisted of (100) pregnant and non-pregnant women who suffer from frequent UTI. The current study showed that the most frequent bacteria in this type of infection obtained in this research were *E. coli* bacteria, *Klebsiella*, and *pseudomonas*. Additionally, the most effective antibiotics affecting isolated bacteria were gentamicin, Ciprofloxacin and Nitroflurontion. Therefore, these antibiotics can be used for treating such bacteria.

Keywords: Women diseases, Isolation and Diagnosis of Bacteria Urinary Tract Infection, Antibiotic Susceptibility.

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INTRODUCTION

Urinary tract infection (UTI) is one of the most prevalent infections and is classified into complicated and uncomplicated infections with respect to choices for treatment (Sherifa & Moataz, 2012). It occurs because of the spontaneous transfer of bacteria from the urethra to the bladder. Then, the bacteria may transfer from bladder to the kidneys and bloodstream causing chronic kidney infections and blood poisoning (Vasudevan, 2014). It is caused by the presence of negative bacteria, such as Gram stain like *E. coli, proteus, pseudomonas, Enterobacter and Klebsiella* (Ali, Garba & Abdallah, 2018). There are common recurrent UTIs that can cause permanent damage to the kidneys. In several cases, this results in renal hypertension and renal failure (New CH., 1992). Multi drug resistant Microorganisms are a danger to public health (Ibrahim, Bilal, & Hamid, 2012; Kothari & Sagar, 2008; Tiruneh et al., 2014).

In most UTI cases, starting treatment is required before having the microbiological results of culture. The clinician can choose the right treatment through reviewing studies that are conducted in certain areas to determine the type of bacteria that causes UTIs and patterns of their antibiotic's resistance (Hryniewicz et al., 2001). In spite of their significant role in managing many infectious clinical syndromes, the increasing usage of antibiotics by several means (e.g., non-careful prescription, improper dosing and treatment duration, and over-the-counter availability of antibiotics) has contributed to increase the antibiotic resistance to several common pathogens (Kerr, 2005). In the treatment of UTIs, antibiotic resistance is a serious health problem, especially in the developing countries. This is attributed to the high level of ignorance, poverty, and bad hygienic practices. In addition to the high prevalence of fake and illegal drugs of poor quality (Abubakar, 2009).

UTI affects women more than men (Mohammed et al., 2016; Tabasi et al., 2015). This could be because of the closeness of genital tract and urethra/anus (Schaeffer et al., 2001) and anatomical predisposition or urothelial mucosa adherence to mucopolysaccharides lining or other host factors (Akortha, 2008). It has been observed that about 20% of women have experienced a single incidence of UTI during their lifetime, and 3% of them have had more than one incidence of UTI per year (Gebre-Selassie, 1998).

During pregnancy, women are more susceptible to the infection (Van Nostrand, Junki, & Bartholdi, 2000), and hence complications may be caused due to UTI. Such complications include pyelonephritis, hypertensive disease of pregnancy, anemia, chronic renal failure, premature delivery and fetal mortality (Delzell *et al.*, 2000; Foxman, 2002). Hence, the treatment of asymptomatic and symptomatic bacteriuria can decrease the rate of these complications (Delzell *et al.*, 2000). With advancing age, men become more susceptible to UTI due to enlargement of prostate and neurogenic bladder (Liperky, 1989). Therefore, this study aims to isolate bacteria from pregnant women and non-pregnant women who suffer from frequent UTIs and to examine the resistance of isolated bacteria to antibiotics.

As well as Several studies were conducted to examine UTIs and the susceptibility of bacteria to antibiotics. Moyo et al. (2010) studied bacterial isolates and patterns of medicine susceptibility of UTI among pregnant women at Muhimbili National Hospital in Tanzania. They found that the most commonly isolated bacteria were Escherichia coli, Klebsiella spp, coagulase negative Staphylococcus, Staphylococcus aureus, Proteus species and Enterococcus species. They recommended observing the resistance levels for nitrofurantoin, fluoroquinolone and cefotaxime, as well as observing the production of Extended Spectrum Beta Lactamase among cefotaxime resistant E. coli and Klebsiella spp.

Khoshbakht et al. (2012) studied antibiotic susceptibility of bacterial strains isolated from UTIs in Karaj, Iran. They aimed at determining patterns of antibiotic resistance bacteria isolated from UTIs. They found that E. coli

isolates were the predominant pathogens in UTI. In addition, antibiotics of ciprofloxacin and nitrofurantoin showed the highest activity, whereas ampicillin and sulfamethoxazole-trimethoprim showed the lowest activity. They recommended conducting studies to identify the tendency of antibiotic resistance in order to prescribe the proper antibiotics for treatment.

Shilpi et al. (2013) studied isolation of bacteria that causes UTIs and the profile of their antibiotic susceptibility at Anwer Khan Modern Medical College Hospital. They intended to examine the antibiotic susceptibility and resistance pattern of isolated urinary pathogens. They found that *Escherichia coli*, *Streptococcus feacalis*, *Pseudomonus*, *Klebsiella* species and *Staphylococcus epidermidis* were the most frequent pathogens isolated. In addition, patients with Enterobacteriaceae were sensitive to Amikacin and Nitrofurantoin by 75-100 %; however, they showed a variable sensitivity to other antibiotics that are frequently used. They recommended that Meropenem and Amikacin should be prescribed selectively in cases where there is no response to antibiotics used frequently.

Sabir et al. (2014) examined isolation and antibiotic susceptibility of E. coli from UTIs in a tertiary care hospital. They found that the most prevalent bacteria were *E. coli*, followed by *Staphylococcus aureus*, *Proteus species* and *Pseudomonas species*. *E. coli* was highly resistant to penicillin, amoxicillin and cefotaxime. However, it showed low resistance to streptomycin, kanamycin, tazocin, and amikacin. Moreover, multiple drug resistant E. coli causes most of UTIs in human.

Angoti et al. (2016) studied bacteria isolated from UTI among patients to determine the patterns of antibiotic susceptibility of the Gram-negative bacteria and their prevalence in Iran. Their study showed that the probability of UTI is increased by the incidence of *E. coli* and *Enterobacter spp*. In addition, there are antibiotic resistant infections; therefore, the efficiency of integrated infection control programs should be improved to control nosocomial infections that are caused by organisms with highly resistance.

Kulkarni et al. (2017) investigated isolation and pattern of antibiotic susceptibility of *Escherichia coli* from UTI in a Tertiary Care Hospital of North Eastern Karnataka. They intended to analyze the resistance pattern of *E. coli* that causes UTI in patients within 4 years (2012-2015). They concluded that multiple drug resistant E. coli caused UTIs in subjects of their study. The sensitivity showed a continuous decline during the study period, with Imipenem as the most effective antibiotic.

Alhamdany (2018) studied antibiotic susceptibility of bacteria isolated from patients with diabetes mellitus and frequent UTIs in Babylon Province, Iraq. He aimed to detect the types of bacteria and their antibiotic sensitivity in patients with diabetes mellitus with recurrent UTIs and compare between types of bacteria in patients with the previous admission to hospital from those who are not. He found that there was a significant difference in bacterial type between patients who previously had hospital admission and those who had not. E. coli was the main bacteria causing UTI. Amikacin showed the best sensitive drug for bacteria that cause UTI.

Mahdi et al. (2020) studied bacteria isolated from urine and their antimicrobial susceptibility. They aimed at isolating the bacteria that cause UTI and the microorganisms with the highest resistance rates to different antibiotics. They found that *Escherichia coli* was the most common pathogen, followed by *Klebsiella pneumonia*. Additionally, bacteria were most susceptible to amikacin. The highest resistance rates were observed for β-lactams, while cefixime was the most resistant antibiotic.

MATERIALS AND METHODS

Sample selection

This study consisted of (100) samples collected from pregnant and non-pregnant women (50 for each). Women's age ranged between 20-40 years. All of the women were visiting the outpatient clinics in Al-Hilla, Babylon city, Iraq.

Urine collection

Urine samples were collected in sterile sealed cans. Then, they were transferred to the laboratory to undergo microscopic and biochemical examinations. After that, they were transplanted into different selective media to diagnose bacteria.

Microscopic test

The glass slides of the sediment produced from the centrifugation of urine samples were prepared. The slides were stained with Gram stain for the initial examination of the bacteria present in the urine.

Isolation and diagnosis of different bacterial species

A series of Ten-fold serial dilutions was prepared for urine samples. After that, the samples were implanted on an optional culture media to differentiate between bacterial species. Then, the bacteria were diagnosed by conducting biochemical tests.

Antibiotic Susceptibility

Pure bacteria were planted on the center of Muller Hinton agar using cotton swabs. Then, 3 tablets of antibiotics were added per dish. The diameter of the inhibition zone was measured by comparison with the clsi tables for the year 2019 to measure the resistance of bacteria to life antibodies (Cip: Ciprofloxacin, Im: Impinem, Gm: Gentamicin, Fm: Nitroflurontion, Am: Ampicillin, and Trimethoprim/ Sulphamethoxazol SXT).

RESULT AND DISCUSSION

This section presents and discusses the most important results obtained from this study. Hence, table (1) represents the numbers and types of bacteria negative to Gram stain that were isolated and analyzed from pregnant and non-pregnant women.

Table 1: Prevalence of isolated from urine samples of UTI patients

	Total samples	*E. coli*	*Klebseilla*	*Pseudomonas*
Pregnant women	50	30	12	25
Non-pregnant women	50	35	10	22

Based on results shown in table (1), it is clear that the most recurrent bacteria were *E. coli,* followed by *Pseudomonas* and finally *Klebseilla* in both pregnant and non-pregnant women. Knowing that the number of positive samples for all types of bacteria was more apparent in pregnant women than in non-pregnant

women. This result is nearly consistent with that found by Ali et al. (2018) who concluded that the most pronounced bacteria isolated from urine samples for patients with UTI were E. coli bacteria followed by staphylococcus iris, then Klebsiella and finally Pseudomonas. In addition, Dinah et al. (2019) indicated that the most recurrent bacteria were E. coli, followed by Klebsiella, and finally the Pseudomonas. They also stated that urine specimens should be cultured to the appropriate culture media and tested for antibiotic sensitivity for all bacteria isolated from UTI, for determining appropriate treatment and thus avoiding the spread of antibiotic resistance.

In this study, the bacteria isolated were more sensitive to the antibiotics of gentamicin, Ciprofloxacin and Nitroflurontion. However, ampicillin did not affect the bacterial growth, that is, all bacterial isolates were resistant to it. The bacterial response to the remaining antibiotics (Impinem and Trimethoprim/ Sulphamethoxazol) was low. Moreover, life antibiotics did not fall into the category of antibiotics that can be used in the treatment of UTI of bacteria isolated in this study. This result corresponds to that found by Jassim et al. (2013) who indicated that the best treatment for UTI are gentamicin, Ciprofloxacin and Nitroflurontion. Furthermore, this study found that isolated bacteria from patients with UTI and those who suffer from chronic kidney infections are more resistant to antibiotics than bacteria isolated from patients without chronic kidney infections. This may be attributed to the excessive use of antibiotics (Majeed & Aljanaby, 2019).

CONCLUSION

It is concluded that the most recurrent bacteria were *E. coli,* followed by *Pseudomonas* and finally *Klebseilla* in both pregnant and non-pregnant women. The bacteria isolated were more sensitive to the antibiotics of gentamicin, Ciprofloxacin and Nitroflurontion. Furthermore, isolated bacteria from patients with UTI and those who suffer from chronic kidney infections are more resistant to antibiotics than bacteria isolated from patients without chronic kidney infections. Finally, the bacteria that causes UTI became more resistant to antibiotics due to the misuse and failure to consult a doctor before starting the treatment.

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