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Incidence of urinary tract infection (UTI) among pregnant women in Al-Muthanna Province

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Abstract: This study reported the incidence of urinary tract infections (UTIs) among 64 pregnant women (50 patients and 14 controls) conducted in AL-Samawa Teaching Hospital of Gynecology and Pediatrics, as well as the isolation and identification of the pathogens responsible for the infection. Significant bacteriuria is defined as greater than $1^{x}10^{5}$ of the same bacteria per milliliter of urine. A total of 64 clean voided mid-stream urine samples were collected from pregnant women between the ages of 13-40 years. The results showed that the incidence of UTIs in this study population was 66%, and 33 bacterial isolates (20 single cultures and 13 mixed cultures) were identified based on colonial size, color characteristics, according to HiCrome UTI agar. The most predominant bacterium was *Staphylococcus aureus (ATCC 25923)* 12 (36.4%). This was followed by *Enterococcus faecalis (ATCC29212)* 4 (12%), *Pseudomonas aeruginosa (ATCC 27853)* 2(6), *Klebsiella pneumonia (ATCC 13883)* 1 (3%), *Escherichia coli (ATCC 25922)* 1 (3%), and a mixed culture of *Enterococcus faecalis (ATCC 29212)* and *Staphylococcus aureus (ATCC 25923)* 8(24.2%), *Escherichia coli (ATCC 25922)* and *Enterococcus faecalis (ATCC 29212)* 2(6%), *Staphylococcus aureus (ATCC 25923)* and *Klebsiella pneumonia (ATCC 13883)* 2(6%) , *Enterococcus faecalis (ATCC* 29212) and *Klebsiella pneumonia (ATCC 13883)* 1(3%) . All bacterial species exhibited 100% susceptibility to Amikacin and Imipenem whereas showed 100% resistance to Ampicillin, Erythromycin, Cephalothin, Cephalexin, Augmentin, Kanamycin, Tobramycin, and Neomycin

Introduction:

Urinary Tract Infections (UTIs) is an infection caused by the presence and growth of microorganisms anywhere in the urinary tract. It is perhaps the single most common bacterial infection of mankind (1; 2). Urinary tract includes the organs that collect and store urine and release it from the body which include: kidneys, ureters, bladder and urethra. (UTIs) are among the most common bacterial infectionsin humans, both in the community and hospital settings and have been reported in all age groups in both sexes (3). UTI has become the most common hospital-acquired infection, accounting for as many as 35% of nosocomial infections, and it is the second most common cause of bacteraemia in hospitalized patients (4; 5; 6). UTI accounts for a significant part of the work load in clinical microbiology laboratories and enteric bacteria (in particular, Escherichia coli) remain the most frequent cause of UTI, although the distribution of pathogens that cause UTI is changing (7; 6). Numerous reports have also suggested that UTI can occur in both males and females of any age, with bacterial counts as low as 100 colony forming units (CFU) per millimeter in urine (8; 2). Females are however believed to be more affected than males except at the extremes of life (2; 6). Untreated upper UTI in pregnancy carries well documented risks of morbidity, and rarely, mortality to the pregnant women (9). Recently published studies have added to the body of knowledge concerning the pathogenesis, diagnosis and management of UTIs (10).Usually, a UTI is caused by bacteria that can also live in the digestive tract, in the vagina, or around the urethra, which is at the entrance to the urinary tract. Most often these bacteria enter the urethra and travel to the bladder and kidneys. Usually, the body removes the bacteria, and shows no symptoms. The signs and symptoms include

burning feeling during urination, frequent or intense urges to urinate, even when one have little urine to pass, pains in the back or lower abdomen, cloudy, dark, bloody, or unusual-smelling urine, fever or chills (11). Women tend to have UTIs more often than men because bacteria can reach the bladder more easily in women. This is partially due to the short and wider female urethra and its proximity to anus. Bacteria from the rectum can easily travel up the urethra and cause infections (2; 12; 6). Moreover, the main factors predisposing married women to bacteriuria are pregnancy and sexual intercourse (13). Sexual activity increases the chances of bacterial contamination of female urethra. Having intercourse may also cause UTIs in women because bacteria can be pushed into the urethra. This anatomical relationship of the female urethra to the vagina makes it liable to trauma during sexual intercourse as well as bacteria being massaged up the urethra into the bladder during pregnancy/child birth (14; 2; 6). Using a diaphragm can also lead to UTIs because diaphragms push against the urethra and make it more difficult to completely empty the bladder. The urine that stays in the bladder is more likely to allow growth of bacteria and cause infections (12; 11). However, the importance of coliform bacilli in UTI among pregnant women has long been known in developed countries (15). Health care practitioners regularly have to make decisions about prescription of antibiotics for urinary tract infections. UTI is the second most common clinical indication for empirical antimicrobial treatment in primary and secondary care, and urine samples constitute the largest single category of specimens examined in most medical microbiology laboratories (1).

UTI is challenging, not only because of the large number of infections that occur each year, but alsobecause the diagnosis of UTI is not always straight forward (6). Criteria for the diagnosis of UTI vary greatly depending on the patients and context. According to (16), there is considerable evidence of practice variation in use of diagnostic tests, interpretation of signs or symptoms and initiation of antibiotic treatment such as drug selection, dose, duration and route of administration (17). For patients with symptoms of UTI and bacteriuria the main aim of treatment is to get rid of infectious bacteria causing the symptoms. Secondary outcomes are adverse effects of treatment or recurrence of symptoms.

This study therefore focuses on the detection and incidence of UTI among pregnant women. It also aimed to isolate and identify the organisms isolated from clinical specimen.

MATERIALS AND METHODS

Study population

A total of 64 pregnant women (50 patients and 14 controls) between the ages of 13 to 40 years were included in this study which lasted from September to November 2012. All these persons were outpatients attending the antenatal clinics in AL-Samawa Teaching Hospital of Gynecology and Pediatrics.

Demographic and clinical information

Demographic and clinical information of the subjects were obtained by chart abstraction and recorded on a prepared data collection form. The study groups were also stratified by age distribution. Information was collected on the women's age, gestational age, and perceived gynaecological symptoms, health care-seeking behavior and contraceptive practices.

Urine collection

Clean catch urine samples were collected in sterile universal containers as described by 18 and 19. Eighty "clean catch" mid-stream urine (MSU) samples were collected inside sterile disposable universal bottles from pregnant women. They were instructed on how to collect samples and the need for prompt delivery to the laboratory. The samples were labeled and transported to the Medical Microbiology laboratory of AL-Muthanna University-College of medicine and were analyzed within 30 min to 1 h of collection.

Media

The media used was HiCrome UTI Agar. HiCrome UTI Agar is a differential medium recommended for presumptive identification of microorganisms mainly causing urinary tract infections.

This media was prepared according to 20; 21; 22; 23; 24 and 25.

Bacterial colonies growing on the agar after the incubation period were enumerated to determine urine samples with significant bacteriuria, this carried out as described by 26.

Diluting and plating procedure:-

1. Liquefy a bottle of nutrient agar. While it is being heated, label three 99 ml sterile water blanks A, B and C. also, label the four Petri plates 1:10,000, 1:100,000, 1:1,000,000, and 1:10,000,000. In addition, indicate with labels the amount to be pipette into each plate (0.1 ml or 1.0 ml).
2. Shake the culture of bacteria and transfer 1 ml of the organisms to blank A, using a sterile 1.1 ml pipette. After using the pipette place it in the discard canister.
3. Shake blank A 25 times in an arc of 1 foot for 7 seconds with your elbow on the table. Forceful shaking not only brings about good distribution, but it also breaks up clumps of bacteria.
4. With a different 1.1 ml pipette, transfer 1 ml from blank A to blank B.
5. Shake water blank B 25 times in same manner.
6. With another sterile pipette, transfer 0.1 ml from blank B to the 1:100,000 plate and 1.0 ml to the 1:10,000 plate. With the same pipette transfer 1.0 ml to blank C.
7. Shake blank C 25 times.
8. With another sterile pipette, transfer from blank C 0.1 ml to the 1:10,000,000 plate and 1.0 ml to the 1:1,000,000 plate.
9. After the bottle of nutrient agar has boiled for 8 minutes, cool it down in a water bath at 50℃ for at least 10 minutes.
10. Pour one-fourth of the nutrient agar (20 ml) into each of 4plates. Rotate the plates gently to get adequate mixing of medium and organisms. This step is critical, too little action will result in poor dispersion and too much action may slop inoculated medium over the edge.
11. After the medium has cooled completely, incubate at 35℃ for 48 hours, inverted.

Counting and calculations:-

After incubation period the colony counted by classical method to numerate the number of colony in each plate. Select the plates that have no fewer than 30 nor more than 300 colonies for your count. Plates with less than 30 or more than 300 colonies are statically unreliable.

Calculate the number of bacteria per ml of undiluted culture by multiply the number of colonies counted by the dilution factor (the reciprocal of the dilution) for example: if you counted 220 colonies on the plate received 1.0 ml of the 1:1000000 dilution (220×1000000 or 2.2×10^8).

Antimicrobial susceptibility test:

Ten standard commercial antibiotics (Ampicillin, Amikacin, Erythromycin, Kanamycin, Tobramycin, Imipenem, Neomycin, Cephalothin, Cephalexin and Augmentin) used for antibiotic sensitivity test by using Antibiotic diffusion test (Kirby-Bauer susceptibility test) was carried out according to (27).

Results:

Of the 64 samples examined in this study, only 33 patients (66%) were found to contain bacterial growth (significant bacteriuria) and 17 (34%) had no bacterial growth while in control 2 pregnant women have positive bacterial culture (significant bacteriuria) and 12 pregnant women contain no bacterial growth (figure 1).

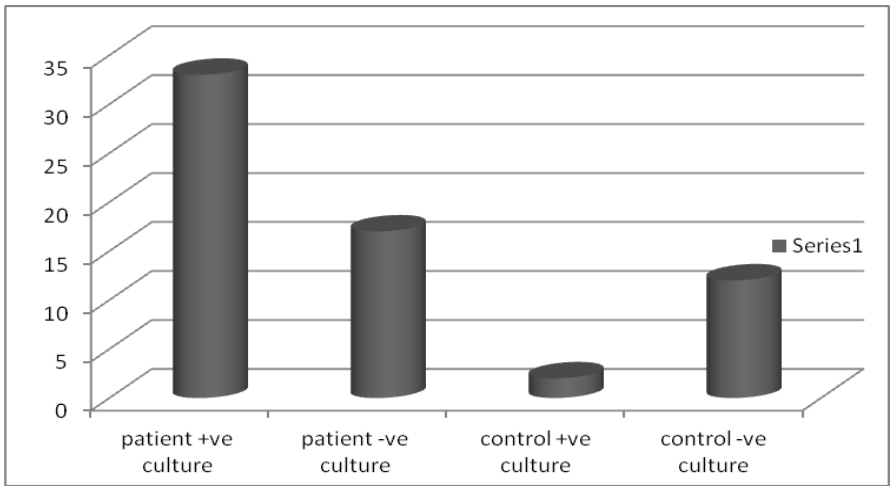

Figure (1) percentage of positive and negative urine culture for patients and control group

Urine culture revealed 51.51% Gram positive bacteria and 48.48% Gram negative bacteria as show in figure 2.

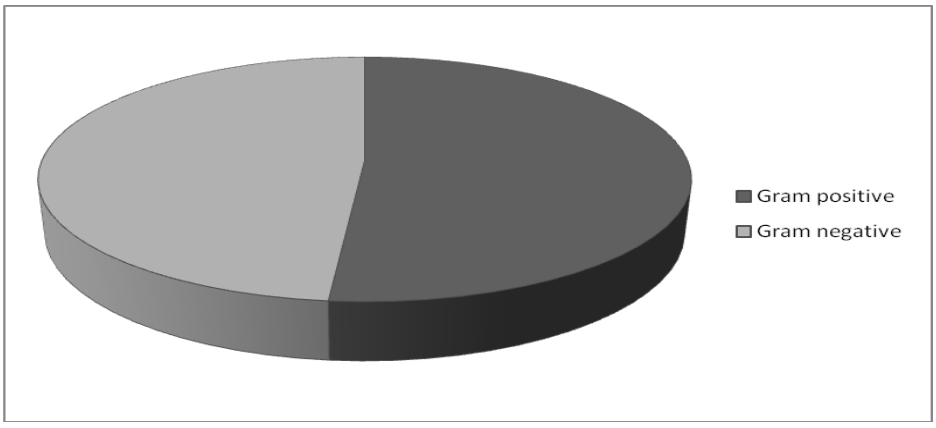

Figure (2) percentage of Gram positive and Gram negative bacteria isolated from pregnant with UTI.

Table (1) incidence of UTI in relation to age distributions of pregnant women.

Age group (years)	No. tested (%)	No. positive (%)	No. negative (%)
13-20	13 (26)	12 (92.3)	1 (7.7)
21-25	15 (30)	12 (80)	3 (20)
26-30	12 (24)	5 (41.7)	7 (58.3)
31-35	5 (10)	3 (60)	2 (40)
36-40	5 (10)	4 (80)	1 (20)
Total	50 (100)	36 (72)	14 (28)

Table 1 shows the incidence of UTIs in relation to age of the subjects. A higher percentage of pregnant women (30%) with UTIs were found within the age brackets of 21-25 years while age groups 31-35 and 36-40 years had the least percentage (10%). The highest number of bacterial isolates was obtained from pregnant women within the age brackets of 13-20 years followed by 21-25 years. Comparatively, lower number of bacterial isolates was obtained from age groups 31-35 years.

Table (2) UTI incidence by gestational age (age of pregnancy)

Age of pregnancy (months)	No. tested	No. positive (%)
2	13	10 (76.9)
3	6	6 (100)
4	6	5 (83.4)
5	3	2 (66.7)
6	4	2 (50)
7	4	1 (25)
8	8	3 (37.5)
9	6	4 (66.7)
Total	50	33 (66)

Table 2 shows the prevalence of UTI by gestational age (age of pregnancy) as at the time of this study. This revealed that women in the 3 and 5 months of their pregnancy had the highest prevalence of 100% and 83% respectively while women in the 7 month of their pregnancy had the lowest bacteria growth.

Table (3) Incidence of UTI by trimester (period of 3- three-months of pregnancy).

Trimester (period of 3- three-months)	No. tested	No. positive (%)
First trimester (1st 3 months)	19	16 (84.2)
Second trimester (2nd 3 months)	13	9 (69.2)
Third trimester (3rd 3 months)	18	8 (44.4)
Total	50	33 (66)

Table 3 shows the incidence of UTI by trimester (a period of three months, especially one of the three-three-month periods into which human pregnancy is divided for medical purposes) as at the time of this study. This also shows that women in their 1st and 2nd trimester had a greater number of UTI cases having an incidence of 84.2 and 69.2% respectively. Women in their third trimester, though fewer in number. This shows that the incidence of UTIs among pregnant women could also be contributed by trimester**.**

Table (4) No. of bacterial isolates in pregnant women infected with UTI.

Isolates	No. of bacterial isolates (%)
Single culture	
Staphylococcus aureus ATCC 25923	**12 (36.4)**
Enterococcus faecalis ATCC 29212	**4 (12)**
Klebsiella pneumonia ATCC 13883	**2(6)**
Escherichia coli ATCC 25922	**1 (3)**
Pseudomonas aeruginosa ATCC 27853	**1 (3)**
Mixed culture	
Staphylococcus aureus ATCC 25923 and Enterococcus faecalis ATCC 29212	**8 (24.2)**
Escherichia coli ATCC 25922 and Enterococcus faecalis ATCC 29212	**2 (6)**
Staphylococcus aureus ATCC 25923 and Klebsiella pneumonia ATCC 13883	**2 (6)**
Enterococcus faecalis ATCC 29212 and Klebsiella pneumonia ATCC 13883	**1 (3)**
Total	**33 (100.0)**

Of the 33 isolates obtained, Gram positive bacteria occurred more frequently than Gram negative bacteria. These include *Staphylococcus aureus (ATCC 25923*) 12 (36.4), as shown in Table 4. Gram negative bacteria accounted for (48.48%), with *Enterococcus faecalis (ATCC 29212)* 4(12%), *Klebsiella pneumonia (ATCC 13883)* 2(6%), *Escherichia coli ATCC (25922)* 1(3%), *Pseudomonas aeruginosa ATCC (27853)* 1(3%), and mixed cultures of *Staphylococcus aureus ATCC 25923 and Enterococcus faecalis ATCC 29212* accounting for 8 (24.2%), *Escherichia coli ATCC 25922 and Enterococcus faecalis ATCC 29212* accounting for 2 (6%), *Staphylococcus aureus ATCC 25923 and Klebsiella pneumonia ATCC 13883* accounting for 2 (6%), *Enterococcus faecalis ATCC 29212 and Klebsiella pneumonia ATCC 13883* accounting for 1 (3%) as shown in Table 4. It was also found that the rate of isolation of *Staphylococcus aureus ATCC 25923* was higher in specimens collected from pregnant women in all age brackets.

DISCUSSION

The incidence of UTIs in this study population was 66%. The findings in this study comparably to the 58% incidence rate reported by (28) in a similar study among pregnant women in Ondo state, but lower than a prevalence rate of 71.6% earlier reported in a similar study by (29) in non-pregnant women less than 50 years of age with acute systems of UTIs, and with (30) who recorded 77.9% among Prison inmates in Nigeria. This high incidence of UTI reported in this study may also be attributed to such factors as poor housing, poor drainage systems, lack of proper personal and environmental hygiene, genuine population susceptibility since it is that factors such as low socio-economic status, sexual intercourse, and pregnancy among others are common among Nigerian Women (31; 8; 6). The incidence of bacteriuria among women in their first trimester is 84.2%. This figure is higher than the prevalence rate of 2-9% reported by (32) in a similar study on pregnant women. This shows that symptomatic bacteriuria occurs in 17-20% of pregnancies. The findings of this study also showed that 44.4% of the women who had UTIs were in their 3rd pregnancy; 69.2% were in their 2nd pregnancy and 84.2% were in their 1st pregnancy. This showed that parity is one of the possible factors affecting the incidence and prevalence rate of UTIs among women. This study also showed that women in their 3th month (100%) and 4th month (83.4%) of their pregnancy had the higher incidence of UTI while women in their 7th month of the pregnancy had less specific bacteria growth. also reported that 10- 30% of women with bacteriuria in the first trimester develop upper UTI in the second or third trimester. Thus, pregnant women should be screened for bacteriuria by urine culture at 12 to 16 weeks of gestation. The presence of 1 x10^5 CFU of bacteria per ml of urine should be considered significant the pattern and frequency of occurrence of the bacterial isolates found in this study is different to what has been previously reported. *E. coli* is the most common pathogen among patients with uncomplicated UTIs (33). Other members of the family *Enterobacteriaceae* (such as some strains of *Klebsiella* spp.) and other organisms (such as *S. aureus*), can have similar requirements (34; 16). The most implicating organism causing urinary tract infections among these pregnant women in this study was *Staphylococcus aureus (ATCC 25923)* 12 (36.4%). This was followed by *Enterococcus faecalis (ATCC29212)* 4 (12%), *Pseudomonas aeruginosa (ATCC 27853)* 2(6), *Klebsiella pneumonia (ATCC 13883)* 1 (3%), *Escherichia coli (ATCC 25922)* 1 (3%), and a mixed culture of *Enterococcus faecalis (ATCC 29212)* and *Staphylococcus aureus (ATCC 25923)* 8(24.2%), *Escherichia coli (ATCC 25922)* and *Enterococcus faecalis (ATCC 29212)* 2(6%), *Staphylococcus aureus (ATCC 25923)* and *Klebsiella pneumonia (ATCC 13883)* 2(6) , *Enterococcus faecalis (ATCC* 29212) and *Klebsiella pneumonia (ATCC 13883)* 1(3). In a similar study by Nwanze *et al*. (2009) the commonest isolates were *Escherichia coli* (51.2%), *S. aureus* (27.3%), and *K. pneumoniae* (12.8%) respectively. This same pattern was also reported by (6). The 18.4% incidence rate reported for *K. aerogenes* in this study brings to light the fact that *Klebisella species* are achieving more prominence as aetiological agents of UTI than previously reported (35; 36; 37; 38; 6). According to (39), *S. aureus* is believed to cause cystis in mainly young sexually active females, it was also found to constitute a recognizable percentage in this study. This confirms that this organism may be achieving prominence as an aetiological agent of UTI in pregnant women. In this study, a total of 33 isolates were obtained from the 50 pregnant women. A higher percentage of the organisms found in this study were isolated mainly from pregnant women with 13-20 years age group while a higher prevalence of UTI was found in age groups 21-25 years. This confirms the usual report that the risk of UTIs increases with age. The incidence of UTIs in this study group was 66% (33 women). The higher prevalence of urinary tract infections in pregnant women might be as a result of a variety of factors; women under 50 years of age with acute symptoms such as dysuria, urgency or frequency suggesting of lower UTI or loin pain suggesting of upper UTI are extremely likely to have bacteriuria. The high incidence rate of

47.5% reported in this study should be of great concern, as not only do UTI pose a threat to health, but they also impose an economic and social burden due to the stigma associated with these infections. The findings of this study revealed that the important infecting organisms were found to be the commensals of perianal and vaginal regions. This call for increase in personal hygiene (6).This study has highlighted the need to raise awareness of UTIs and to expand services for prevention and treatment for pregnant women. To do this effectively, however, it may be necessary to improve the quality of health care provided at the community level. Since UTI may be symptomatic and asymptomatic in most cases, it is therefore suggested that routine screening of patients with unexplained sources of fever be done for UTI and the appropriate antimicrobials administered after sensitivity tests have been carried out in order to prevent the cases becoming symptomatic later with resultant renal damage.

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نسبة حدوث خمج المجاري البولية بين النساء الحوامل في محافظة المثنى

تيسير عبدالاله كاظم / ماجستير احياء مجهرية /كلية الطب /جامعة المثنى

الكلمات المفتاحية (التهاب المجاري البولية؛ النساء الحوامل؛ تجرثم البول؛ احداث؛ وسط التهاب المجاري البولية عالي الصبغة)

الخلاصة:

تهدف هذه الدراسة الى بيان نسبة حدوث اخماج المجاري البولية بين 64 امراة حامل (50 مريضة و14 مجاميع سيطرة) ادخلوا الى مستشفى النسائية والاطفال التعليمي في السماوة بالاضافة الى عزل وتشخيص المسببات الجرثومية لاخماج المجاري البولية. معنوية تجرثم البول تعرف على انها عزل اكثر من 1×10^5 من نفس الجرثومة في المليلتر الواحد من البول. تم جمع 64 عينة بول مأخوذة من وسط مجرى البول من النساء الحوامل من اعمار تتراوح بين 13-40 سنة. اوضحت النتائج ان نسبة حدوث اخماج المجاري البولية بين النساء الحوامل كانت 66% .تم عزل 33 جرثومة (20 عينة عزلت منها جرثومة منفردة و13 عينة عزلت منها اكثر من جرثومة) حيث تم تشخيص العزلات بالاعتماد على لون وحجم المستعمرات وذلك وفقاً الى وسط اخماج المجاري البولية عالي الصبغة. اكثر الجراثيم التي تم عزلها كانت *المكورات العنقودية*12 (36.4%) واقل منها *البكتريا المعوية* 4(12%) ثم *الزوائف الزنجاية*2 (6) والكليبسيلة الرئوية (3%) 1 .اما العينات التي عزلت منها اكثر من جرثومة فكانت *البكتيريا المعوية* مع *المكورات العنقودية*8 (24.2%) بالاضافة الى *الاشريشيا القولونية* مع *البكتيريا المعوية*2 (6%) *والمكورات العنقودية* مع *الكليبسيلة الرئوية*2 (6%) واخيراً *البكتيريا المعوية* مع *الكليبسيلة الرئوية*1 (3%). كل الجراثيم المعزولة كانت 100 % حساسة للاميكاسين والاميبينيم بينما كانت 100 % مقاومة للامبيسيلين ، الاريثرومايسين ، سيفالوثين ،سيفالوكسين، اوكمنتين , كانامايسين ،توبرامايسينى ونيو مايسين.