study on the Uropathogens in the Area of Sirt

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Abstract:

A thousand Mid-stream urine samples were collected from patients having urinary tract infections visiting Ibn-Sina Hospital in the city of Sirt in the time between March 2003 to September 2003.

It has been found that more females than males were infected. The percentage of occurrence of UTI in females and males were 14.9 %, and 4.7% , respectively. Urinary tract infections were found to be high in females aged between 15 and 40 years.

Gram-negative bacteria were the main cause of these urinary tract infections, since it represented 81 % of the total number of the bacterial infections, *Escherichia coli* caused 47%, or the highest percentage of all Gram-negative bacteria. *Klebsiella spp*. were the next bacterial group with 16 %. While *Enterobacter spp, Proteus mirabilis, Serratia spp.* and *Pseudomonas spp.* came in the third place. *Candida albicans* (Fungus) represented about 11% and Gram-positive bacteria about 7% of the total number of infections.

Keyword: UTI , Bacteriuria ,Infection , Urine , Gram-neg.bacteria

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Introduction:

Urinary tract infections occur in females of various ages and increase with age. They produce considerable morbidity among those who are prone to recurrent infection (Brumfitt and Hamilton-Miller , 1990). Twenty five percent of all women of reproductive age develop a urinary tract infection at sometime in their lives, and half of those develop recurrent infections problem (Lewis *et al*., 1993).

There are several factors that are clearly associated with disruption bladder defense mechanisms and the development of urinary tract infection (UTI). In young ladies urinary tract infection is very serious during pregnancy (Wilkie *et al* .,1992). In addition to, there are problem with respect of diagnosis, subsequent impact on the pregnancy, and the effect of therapy on the fetus (Bailey. 1990).

Healthy women have an increased risk of developing UTI compared to men, perhaps in part because of the shorter length of the females urethra and the close proximity of large numbers of potentially pathogenic bacteria in the vagina and the bowel that becomes contaminated more easily (Lewis *et al.*, 1993).

Bacteriuria is much more common in females than males at all age of life except in the first year when the prevalence in boys (less than 1 percent) is four time greater than in girls (Jodal , 1987). Male infants have a higher rate of UTI than female infants ,possibly related to two factors : a higher rate of congenital genitourinary anomalies and the presence of a prepuce.

Additionally, several studies have shown that uncircumcised male infants have a significantly higher rate of UTI than those who are circumcised (Winberg,1989).

Most infections in boys and in adult men are complicated, that is the individual has an anatomical or urological abnormality , a recent catheterization , or have had urological surgery (Burbige *et al*,. 1984). Vesicoureteric reflux and urethral valves are commonly found in infant boys with bacteruiria and infection, and lead to renal scarring and chronic renal disease. The most common cause of complicated infection in men is nosocomial introduction of urinary pathogens(Cattell, 1996).

The bacteria causing UTI are transmitted by two main routes. The most common route of infection is by the ascending route. Uropathic microbes colonize the urethra and enter the bladder via the urinary stream or along the mucosal sheath. Invasive infection occurs when the microorganisms grow in urine , attach to urothelial cells, and penetrate the urothelia lining. The microorganisms may ascend to ureter and kidneys. Ascending infection is by far the most common route in females and the one most commonly associated with instrumentation in both sexes.(Calvin and Kunin , 1997)

Approach:

This paper is dealing with an investigation on the occurrence of UTIs in Sirt areas and their etiological agents (Bacteria).

Materials and methods:

Specimens collection: Mid- stream urine (MSU) specimens were collected from each adult patient.

Culture media :

Cytosine-Lactose-Electrolyte-Deficient agar (CLED) was used for urine specimens culture. It is commonly used as a selective medium for detection of lactose fermenting Gram-negative bacteria. Additional supportive media , namely; Blood agar and Chocolate agar were used to culture Gram-positive bacteria, including staphylococci and streptococci. Sabouraud agar was used for fungal growth such as *Candida albicans.*

Quantitation of bacterial isolates :

Standard loop technique was used. The number of microorganisms per milliliter recovered on urine culture can aid in the differential diagnosis of UTI, by spreading a loopful of urine over the surface of the agar plate and incubating the plate for 18 – 24 hours at 37 C aerobically. The number of Colony-Forming-Unit (CFU) is multiplied by 100 (if a 0.01 ml loop was used) to determine the number of microorganisms per milliliter in original specimen (Baily ,1990).

However, density of $(10)^5$ CFU / ml urine or more was considered as a significant Bacteriuria for pure culture (Emil and Jack ,1992). or 10.000 to 15.000 cells /ml of urine for Candida (Rosalind , 1988).

Identification of the bacterial isolates :

The morphological characteristics of bacterial colonies are important presumptive methods used for the identification of bacteria isolated. Biochemical tests were used to confirm results of identification of Enterobacteriaceae and other Gram-negative rods. The ENTEROSISTE 18 R (Liofilchem, Italy) is a rapid identification system of oxidase negative enterobacteria of particular clinical interest. The system consists of 18

biochemical substrates, the reactions of which are visible after a 18 – 24 hours of incubation at 37 Ċ. The bacterial species were identified by a numerical code deriving from the positive or negative characters of the reactions themselves.

This test contained : O-nitrophenyl-Beta-D-galactopiranosyde, Lysine, Ornithine, Arginine, Phenylalanine, Citrate, Urea, Hydrogen sulfide, Malonate, Voges-Proskauer, Indole, Glucose, Mannitol, Inositol, Sorbital, Arabinose, and Reffinose.

Additional biochemical tests such as catalase test and coagulase test were used for identification of staphylococci and streptococci.

Results:

From this study the relationship between urinary tract infection and sex was studied (the total number of samples taken was 1000), 804 episodes were non-infected cases and 196 were infected cases; the proportion of patients having bacterial or fungal UTI was 19.6 % namely 47 (4.7 %) males and 149 (14.9 %) females from all patients. UTI therefore, was higher in frequency in females than males.

Table (1): The relationship between urinary tract infections and sex of patients.

Sex	Infected cases	Percent
Females	**149**	**14.90%**
Males	**47**	**4.70%**
Total	**196**	**19.60%**

The family Enterobacteriaceae was the most common group of bacteria responsible for bacterial urinary tract infections. *Escherichia coli* was the most common cause of UTIs and accounted for approximately 47 % of all isolates, whereas *Klebsiella. spp* accounted for 16 % . *Proteus mirabilis* was responsible for 5.6 % of UTIs. *Serratia marcescens* was responsible for approximately 3.6 % of the total isolates and *Pseudomonas aeruginosa* was responsible for about 2.5 %. Generally, in this study the Gram-negative bacteria which caused UTIs made 81 % of all microorganisms isolated.

Whereas Gram-positive bacteria formed 7 % of all isolates ; Staphylococcus spp. represented 5.6 % including *Staph. aureus, Staph. epidermidis* and *Staph. Saprophyticus.*

The enterococci (*E.faecalis*) were found to be 1.5 % of all isolates.

Candidiuria due to *Candida albicans* was responsible for all fungal UTIs. *Candida albicans* accounted for about 11 % of UTIs.

Table(2) : The etiological agents causing urinary tract infections.

Type of bacteria	No. of cases	Percent
Escherichia coli	93	47.2
Klebsiella spp	32	16.2
Enterobacter spp	12	6.1
Proteus mirabilis	11	5.6
Serratia spp	7	3.6
Pseudomonas aeruginosa	5	2.5
Candida albicans	22	11.2
Staphylococcus spp, namely *Staph. aureus* *Staph. epidermidis* *Staph. Saprophyticus*	11	5.6
Enterococcus faecalis	3	1.5
Total	196	100%

Discussion :

Females had a greater susceptibility to ascending infection, as females' urethra is short and vaginal introitus may become contaminated with faecal organisms. Generally, it is assumed that the short urethra in women accounts for their increased susceptibility to urinary tract infections compared to men (Calvin and Kunin,1997) . Urinary`s flow characteristics are important in the initiation of bladder infection .Backflow of urine in the females urethra has been observed during micturition .This process will faciliate the spread of colonizing bacteria into the bladder (Inglis, 1996).

Whereas the males urethra, usually 14-20 cm in length, is substantially longer than the females urethra which presumably makes ascent to the bladder by potential uropathogens more difficult and provides a natural barrier from the colonized skin of the perineum and anus. Penetration of this urethral defense is required in order for bacteriuria to develop (Cattell,1996).

E.coli and other bacteriuria in adult women increases with age. Sexually active and pregnant women had a higher prevalence of bacteriuria than those who are not . In this study ,*E.coli* was the most common cause of urinary tract infection and accounted for approximately 47 % of the total isolated pathogens. The infecting *E.coli* strains survive the faecal flora in patient's surrounding after recolonization and invade the urinary tract.

In addition to that, there are over 170 different cell-wall antigen (O-antigen) groups, over 80 capsular antigens (K- antigen) groups and over 50 flagellar (H –antigen) groups in different *E.coli* strains (Cavin and Kunin ,1997),it would be more expected because of their abundance in the faeces . This is an indication of epidemic virulence .P-fimbbria and some strains have hemolysin

and (O-antigen) groups are often associated more with pyelonephritis than with cystitis and all of these factors are urovirulent ones. (Donnenberg and Welch ,1986)

Whereas *Klebsiella spp* accounted for 16.2 % ,Proteus mirabilis was responsible for 5 % of UTIs. Eventually the *Serratia spp* were responsible for approximately 3% of the total isolates. Those organisms have virulence factors that are similar to those of *E.coli* . Type 1 fimbriae are found in a wide range of Gram-negative bacteria. (Abraham *et al*.,1988)

Tarkkanen (1992) investigated the virulence factors in 39 urinary *Klebsiella* isolates. All pathogenic strains were found to be encapsulated, and all of these strains agglutinated human erythrocytes and reacted with a type 3 fimbria-found in these strains (Cattell,1996).

Production of urease is thought to be the most important virulence factor in Proteus spp isolated from urinary infections. Proteus infection is one of the factors associated with the production of renal stone.(Mushar *et al* ., 1975).

A number of other bacteria produce urease including *Staph. saprophyticus* and certain strains of *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*.

In general, the enteric bacteria are the main cause of UTIs transmitted by faecal contamination.

Gram-positive bacteria were represented in Staphylococcus spp and Enterococcues faecalis. Staphylococci encounted for 6 % of the total isolates, whereas the *E.faecalis* was 1.5 % of all isolates. The Gram-positive bacteria had the ability to adhere to uroepithelial cells, including *Staph. epidermidis*, *Staph. saprophyticus* and *E. faecalis* (Schmidt *et al.*, 1988).Staph aureus is the most important predisposing factor in the urinary tract where related to

indwelling catheters, obstruction and surgery (Arbi and Renneberg ,1984). *Staph. saprophyticus* was responsible for urethritis and cystitis in sexually active women. Whereas *Staph. epidermidis* is the normal skin flora and was as a contaminant in urine, and also be responsible for mid UTI after urological instrumental operations. Enterococci are common contaminants in the urine because they are present in normal faeces(Maskael,1988).

Pseudomonas aeruginosa was one of the most common nosocomial pathogens (it was responsible for approximately 2.5 % of the total isolates) and the risk factor associated with it, is colonization and infection in hospitalized patients; also common with indwelling catheters.

Candida albicans accounted for about 11 % of UTIs. Candida frequently colonizes the urine, after antibiotic use; in diabetics, in association with indwelling catheter or in patients who have previous instrumentation of urinary tract. The presence of C. *albicans* is mostly due to colonization but, not to a true infection. There are evidences that, constituents of normal commensal flora may assume pathogenic roles under some circumstances particularly in immunosuppression. (Maskael,1988).

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الملخص العربي :

خضع لهذه الدراسة ما مجموعه 1000 عينة جمعت كلها من مرضى المسالك البولية المترددون على مستشفى ابن سينا بمدينة سرت وأخذت العينات من وسط عملية التبول (Mid Stream Urine) .

ولوحظ من خلال هذه الدراسة أن أمراض الجهاز البولي من أكثر الأمراض انتشارا فهي تؤثر على كلا الجنسين وفي مختلف الفئات العمرية. كما تبين أن إصابة الإناث بالتهابات المسالك البولية كانت أعلى بكثير من إصابة الذكور ، حيث شكلت الإناث المصابات ما نسبته 14.9 % أما الذكور فكانت 4.7 %.

و وجد أن أعلى معدل للإصابة لدى الإناث هو خلال فترة الخصوبة لديهن والممتدة مابين 15- 40 سنة. بينما يتجانس معدل الإصابة لدى الذكور على امتداد الفترات العمرية المختلفة .

والجدير بالذكر أن البكتيريا السالبة الجرام كانت المسبب الرئيسي لإصابات الجهاز البولي حيث شكلت البكتيريا السالبة التابعة لعائلة Enterobactreiaceacae ما نسبته 81 % من مجموع الحالات المصابة.

وتعتبر بكتيريا *Escherichia coli* المسبب الرئيسي لأغلب إصابات الجهاز البولي حيث شكلت ما نسبته 47% من مجموع المصابين الكلي وهي أعلى نسبة إصابة ببكتيريا سالبة الجرام , يليها بكتريا Klebsiella spp التي شكلت 16 % , بينما كانت بكتيريا Enterobacter spp , Pseudomonas spp, Serratia and *Proteus mirabilis* في المرتبة الثالثة.

Candida albicans هي فطر يسبب التهابات الجهاز البولي والتي شكلت حوالي 11 % , أما البكتيريا موجبة الجرام فشكلت حوالي 7 % من المجموع الكلي للمصابين.