Iraqi JMS

Published by Al-Nahrain College of Medicine
ISSN 1681-6579
Email: Iraqi_jms_alnahrain@yahoo.com
http://www. colmed-nahrain.edu.iq/

Antibiotic Susceptibilities of Gram Negative Aerobic Bacteria Isolated from Urinary Tract Infections in Community

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Abstract

Background Urinary tract infection (UTI) caused by bacteria that can also live in the digestive tract, in vagina, or around the urethra most often these bacteria, enter the urethra and travel to the bladder and kidneys and prostate (in men).

Objective To determine the most common gram negative aerobic bacteria caused UTI in both sex and different ages, and to study the antibiotic susceptibility in order to determine the most effective antibiotics that can cure UTI.

Methods Prospective study of 311 samples of urine has been collected from out patients complaining signs and symptoms of UTI. Isolation and Identification of causative bacteria was concluded, antibiotic susceptibility test has been done, and statistical analysis chi square had done.

Results 125 urine samples obtained from 25 male and 100 female show growths of Gram negative aerobic bacilli. No bacterial growth was defined in the rest of urine samples. Single bacterium was identified in 120 samples, while 5 samples identified as a mixed infection with two kinds of bacteria. In 55 (44%) of cases, *Echerishia Coli* was isolated; in 41 (32.8%) *Klebsiella Pneumoniae*; in 17 (13.6%) *Proteus mirabilis* plus *P. Vulgaris*; and 12 (9.6%) *Pseudomonas aeruginosa*. The percentage of resistance for *E. Coli* varies from 73% to 86%, to Ceftzidime, Ceftriaxone, and Trimethoprim Sulfamethaxozol; for *K. pneumoniae* it ranges from 71% to 100% for Amoxicillin, Pipracillin, Trimethoprim Sulfamethaxozole, and Colistin; for *P. mirabilis* plus *P. vulgaris* ranging from 66% to 100% for Cetazidime, Trimethoprim Sulfamethaxozol, and Cefotaxime; and for *P. aeruginose* ranges from 66% to 100% for Cefazidime, Colistin, Nafcillin, Gentamycin, Trimethoprim Sulfamethaxozol.

Conclusion *E. Coli* caused UTI in female higher than men in the community, and the gram negative–rods had multi antibiotic resistant.

Keyword UTI, Enterobacteriacae, Antibiotic susceptibility

Introduction

Urinary tract Infection (UTI) caused by bacteria that can also live in digestive tract, in vagina, or around the urethra, most often these bacteria, enter the urethra and ascend to the bladder and kidneys and prostate (in men), usually our body removes the bacteria and we have no symptoms [(1)]. Urinary bladder infection has become the most common urinary problem among children, according to conservative estimate, 3% of girls and 1% of boys have had a detected urinary tract infection (UTI) by the age of 11 [(2)].

Women are more likely to get UTIs than men, UTIs can be dangerous especially for older people and pregnant women, as well as for those with diabetes and those who have difficulty urination [(1,3)].

Many substances, such as soap, bubbles bath, stool or clothing can cause soreness of urethra, which makes it easier for bacteria to invade and get into the bladder and multiply [(2,4)].

Anatomic abnormality Increases the risk of bladder infections [3]. Large amount of stool sitting in colon can press up against the bladder and urethra, thus making it more difficult for the bladder to drain completely this allows bacteria to grow [4].

Microorganisms Causing UTI

The Urethra hosts a resident micro flora that colonizes its transitional epithelium, consisting of coagulase negative Staphylococci, Virdans and non hemolytic Streptococci, Lactobacilli, *Diphtheroids (Corynebacterium species)*, nonpatho*genic Neisseria* species, transient gram-negative aerobic bacilli (including Entero bacteriaceae), anaerobic cocci, *Propionibacterium* spp. anaerobic gram-negative cocci and bacilli, commensal *Mycobacterium* spp. commensal *Mycoplasma* species and occasionally Yeasts [3].

Members of the family enterobacteriaceae are among the most important human pathogens. They comprise approximately 80% of gram-negative bacteria and 50% of all isolates identified in hospital laboratories in the United States.

Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis Entrobacter Spp. and *Serratia marcescens* account for the majority of Enterobacteriaceae isolated from clinical specimens [4].

Chlamydia and mycoplasma may also cause UTIs in both men and women and may be sexually transmitted; that is why infections require treatment of both partners [5]. HIV virus may cause the UTI infection [6].

UTI are treated with antibacterial drugs. The sensitivity test is especially useful in helping the doctor to select the most effective drug. The drug most often used to treat routine uncomplicated UTI are listed below [7]:

1. Trimethoprim (Trimpex)
2. Trimethoprim sulfamethoxazole (Bactrim, Septra, Cotrim)
3. Amoxicillin (Amoxil, Trimox, Wymox)
4. Nitrofurantoin (Macrodantin, Furadantin)
5. Ampicillin (Omnipen, Polycillin, Principen, Totacillin)

A class of drugs called Quinolones includes four drugs approved in recent years four treating UTI and they include Ofloxacin (Floxin), Norfloxacin (Noroxin), Ciprofloxacin (Cipro), and trovafloxin (Trovan) [1].

The natural alternative to antibiotics is D-Mannos which is a simple sugar and close cousin of glucose can cure more than 90% of all bladder infections with 1 to 2 days and has no adverse side effects of any kind [2].

Nitric oxide (NO) has been found to possess microbicidal effects against a number of pathogens including DNA and RNA virus families [7].Oral therapy can begin from as early as two months of age and Clavulanate, Cephlexin is recommended for cystitis [15].

Methods

This study was carried out in Al-Karkh Surgery Hospital in Baghdad city, during the period 29th April 2007 to 30th June 2008. Three hundred and eleven urine samples were collected from patients who were presented to the outpatient with signs and symptoms of UTI.

Midstream urine samples were collected in sterile containers, and divided in two portions; the first portion is used for general urine examination and second portion for culture on nutrient agar, blood agar and McConkey agar plates, using sterile standard loop (1ml) then incubated at 37±2°C for 20-24 hours.

Positive urine analysis findings include leukocytes, erythrocytes, bacteria, and squamousal epithelial cells [9] chi-square analysis was done to show significant difference between groups.

Antibiotic susceptibility test done on clinically significant bacterial isolates according to standard modified "Kirby-Baur method" [9,10].

Result

The urine samples were due to patients who were in the age range of 10 to 70 years; and the patient group comprises 224 females and 87 males (Table 1).

Table 1. Age Stages of UTI patients and Bacterial isolates.

Age group	Sex	Bacterial isolates								No Bacterial growth
		Escherichia coli		*Klebsiella pneumoniae*		*P. mirabilis* + *P.vulgars*		*Pseudomonas aeruginose*		
		No.	%	No.	%	No.	%	No.	%	No.
10-20	Male	0	0	1	1.25	0	0	1	1.25	7
	Female	15	18.5	7	8.75	2	2.5	1	1.25	25
21-30	Male	0	0	1	1.25	2	2.5	0	0	9
	Female	10	12.5	7	8.75	5	6.25	2	2.5	30
31-40	Male	0	0	4	5	0	0	1	1.25	10
	Female	8	10	4	5	4	5	3	3.75	20
41-50	Male	2	2.5	2	2.5	1	1.25	1	1.25	10
	Female	10	12.5	6	7.5	1	1.25	1	1.25	20
51-60	Male	1	1.25	3	3.75	0	0	1	1.25	14
	Female	4	5	2	2.5	0	0	0	0	19
61-70	Male	2	2.5	1	1.25	0	0	1	1.25	12
	Female	3	3.75	3	3.75	2	2.5	0	0	10
Total										
Males	25	5	6.25	12	15	3	3.75	5	6.25	62
Females	100	50	62.5	29	36.25	14	17.5	7	8.75	124
all	125	55	68.75	41	51.25	17	21.25	12	15	186
%			44		32.8		13.6		9.6	

Test statistics

	E. coli	*K. pneumoniae*	*Proteus spp.*	*Pseudomonas aeruginosa*
Chi-square [a,b]	151.675	187.328	218.521	334.583
Df	11	11	11	11
Asymp.sig	000	000	000	000

a-o cells (0%) have expected frequencies less than 5 minimum cell frequency is 25.9.
b-o cells (0%) have expected frequencies less than 5 minimum cell frequency is 28.3.
c-o cells (0%) have expected frequencies less than 5 minimum cell frequency is 24.0.
one degree of freedom 0.05, schedule value of chi square
for 11= 19.68
for 10=18.31

	E. coli	*K. pneumoniae*	*Proteus spp.*	*Pseudomonas aeruginosa*
Chi-square [a,b]	194.196	263.084	349.592	382.080
Df	3	3	3	3
Asymp.sig	000	000	000	000

a-o cells (0%) have expected frequencies less than 5. The expected minimum cell frequency is 77.8
Schedule value of chi-square 7.81

A significant growth of aerobic Gram negative bacilli as a single infection was observed in 125 sample (40%) (Table 1) and as mixed infection in 5 sample (3.2%) (Table 2); while there was

no bacterial growth in 170 (60%) of urine samples (Table 1). There were 55 (44%) infections of *E. coli*, 41 (32.8%) infection by *klebsiella pneumoniae* 17(13.6%) infection by *Proteas spp.* and 12(9.6%) infection by P*seudomonas aeruginosa.*

Table 2. Type of mixed pathogens isolated from UTI patients.

Bacteria	Frequency	% Σ single + mixed Bacterial isolates.
Pseudomonas aeruginosa+Klebsiella pneumonae	4	% 2.9
Escherichia Coli+Proteus spp.	2	% 1.48
Klebsiella Pneumoniae+Pseudomonos aeruginosa	2	% 1.48
Pseudomonas aeruginosa+Escherichia coli	1	% 0.74
Escherichia coli+Klebsiella Pneumoniae	1	% 0.74
	Σ = 10	

Statistically, there was a significant relationship between age, gender and type of infections.

The antimicrobial susceptibility test had been done, by using 16 antibiotic produced by Al-Razi Center, and BD BB™ Sensi-Disc™ Antimicrobial. The results were illustrated in table 3.

The resistant percentage to 16 antibiotics was reported as follow: for *E. Coli* was to Ceftazidine 86% then 80% to ceftriaxone, %73 to Trimethoprim sulfamethaxozol, 55% Colistin, 53% Cefotaxime, 45% Nalidixic acid, 42.8% Gentamicin, Tobramycin, 30% Nafcillin, 25% Amoxicillin, 17% Chloramphenicol, 11% Amikacin, 10% Pipracillin, 8% Nitrofurantoin, and still Kanemycin effective against all the *E. coli* strains.

Antibiotic resistant of *K. pneumoniae* was 100% Amoxicillin, 100% pipracillin, 85% Trimethoprime sulfamethaxozol, 71% Colistin, 60% Tetracyclin, 40% Ceftriaxone, Nitrofurantoin, Nalidixic acid, 33% Chloramphenical, 25% Cefotaxime and *K.pneumoniae* was sensitive 100% for kanamycin, Amikacin Nafcillin, Tobramycin.

Antibiotic resistant of P*roteus spp.* 100% Ceftazidime, 80% Trimethoprim Sulfamethoxazol, 66% Cefotaxime, 60% Nitrofurantoin, 58% Nalidixic acid, 50% Colistin, Cefriaxone Chloromphenicol 33% and *proteus spp.* still sensitive to Amikacin, Gentamicin, Nitrofurantoin, Tobramycin.

Pseudomones aeruginosa resistant to antibiotic was 100% Ceftazidime, Colistin Nafeillin, 66% Gentamycin; Trimethoprim Sulfamethaxozol, 50% Tobramycin; Chloramphenicol, Ceftazidime, 33% Nalidixic acid.

Table 3. Antibiotic Susceptibility

Antibiotic	*E. coli*			*K. Pneumoniae*			*Proteus spp.*			*Pseudomonas aeruginosa*		
	R	R%	S	R	R%	S	R	R%	S	R	R%	S
Amikacin	2	11%	16	0	0%	4	0	0	2	0	0	5
Amoxicillin	9	25%	27	10	100%	0	0	0	0	0	0	0
Coftriaxone	8	80%	2	4	40%	6	5	50%	5	0	0	5
Ceftazidime	13	86%	2	0	0	0	4	100%	0	5	100%	0
Cefotaxime (Claforan)	8	53%	7	1	25%	4	2	66%	1	0	0	0
Chloramphenicol	4	17%	19	5	33%	10	3	33%	6	1	50%	1
Colistin	16	55%	13	10	71%	4	3	50%	3	3	100%	0
Gentamicin	3	42.2%	4	0	0	2	0	0	2	2	66%	1
Nalidixic acid (Nigram)	25	45%	30	10	40%	15	7	58%	5	3	33%	6
Nafcillin	4	30%	9	0	0	6	0	0	0	3	100%	0
Nitrofurantion	2	8%	23	6	40%	9	0	0	3	1	0	0
Kanamycin	0	0	10	0	0	5	0	0	0	0	0	0
Pipracillin	10	10%	0	5	100%	0						
Tobramycin	6	42.8%	8	0	0	2	0	0	3	1	50%	1
Trimethoprim sulfamethaxozol (TMP-SMX)	19	73%	7	17	85%	3	4	80%	1	2	66%	1

R=Resistance, S=Sensitive

Discussion

The results of present study showed that the urine cultures reveal no growth of bacteria even in the presence of signs and symptoms of UTI this may be due to other pathogens rather than gram positive bacteria, like fungi, viruses [6].

This result is agreement with [11] except some deviation because of the hospitalized in patients in urological surgery wards, where the nosocomial infections exactly caused by *K. pneumniae* and *P. aeruginosa* [11,12].

P. aeruginosa is important cause of urinary tract infections in patients with urinary catheters. This organism is able to colonize the surface of catheter, forming abiofilm that interferes with activities of antimicrobial agents and host defense mechanism [13]. This result agrees slightly with another study on Urinary tract infections in patients with renal stones [14] which were, the highest percentage of pathogens to *E. coli*, then *Proteus spp., Klebsiella pneumoniae, Pseudomonas aeruginosa*, respectively [14].

Also this result is in agreement with another study had done in France about the UTI in community [15] which showed that the most recovered pathogens were *E. coli* (71%), *Proteus mirabilis* 9%, *Klebsiella pneumonia* 6%, *Pseudomonas aerugenosa* 2%, [15].

And this results goes slightly with a study on community acquired UT1 which was *E. coli* 73% *K. pneumoniae* 6.7% ; *P.aeruginosa* 2.2%, *Proteus spp*. 2.2% [16].

Resistance to antibiotic is most often observed in the hospital setting. Unfortunately, there has been a major worldwide increase within the community in *E. coli* resistance to standard antibiotics, the (Eco. Sens. Project) has been designed to investigate resistant UTI bacteria in 17 European countries [12].

In our study we found that 38% of *E. coli* was resistant to the same antibiotic used in the European study and the difference may due to Ecology and Community circumstances.

The survey of sensitivity to antibiotic agent in Japan, clearly indicated trend for increasing resistance to fluoroqinolones among

enterococci and *P. aeruginosa* isolated from UTI [18] and this agreement with our study.In a European study [12] founded that *E. coli* Resistance to Tmp-smx was 14.1%; Nitrofurantoin and gentamycin 3% and this did not agree with our study.

E. coli is the most common bacteria in UTI. The rates of resistance to common UTI antibiotic vary; however, depending on regions and institution with the highest rate of resistance are those in which antibiotics are heavily prescribed.In European study, for example, the resistance rates were highest in Portugal and Spain and lowest in the Nordic Countries [12]. UTI was higher in females than males and *E. coli* was the commonest pathogen while *K. pneumoiae* infect hospitalized males more than females.

Multi antibiotic resistance transfer between aerobic rods enteric bacteria in community rather than hospital. Multiple organisms are often seen in UTI causes mixed infections which have multi antibiotic resistance.

Acknowledgment

Awfully thanks to the staff of Bacterial Laboratory of Al-Karkh Surgical Hospital for their cooperation in this work.

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Received 16th Sep. 2010, Accepted 30th Jan. 2011