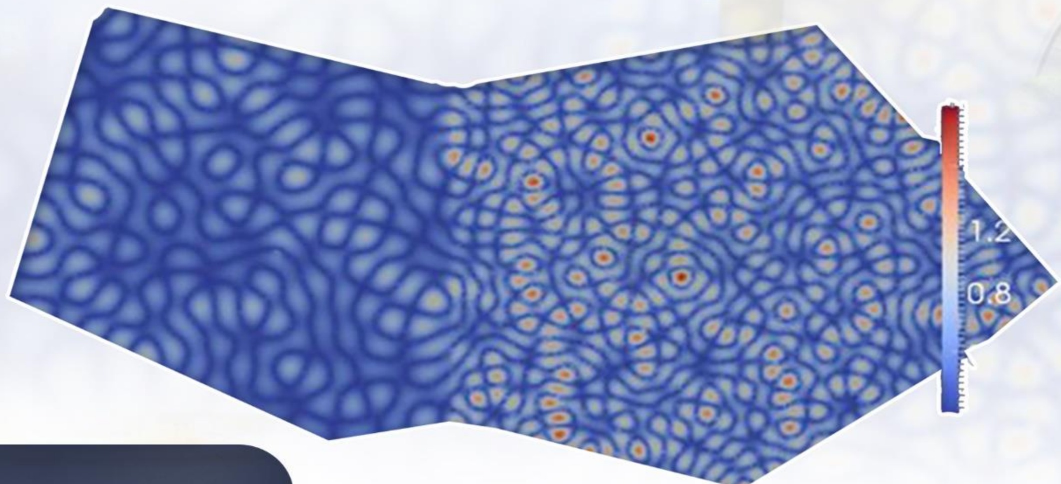
eISSN: 2789-858X

Scientific Journal for the Faculty of Science-Sirte University

DOI: 10.37375/issn.2789-858X - Indexed by Crossref, USA



Volume 2 Issue 1 April 2022

Bi-annual, Peer-Reviewed, Indexed, and Open Accessed e-Journal

SJFSSU

معامل التأثير العربي = 0.32

Legal Deposit Number@National Library (Benghazi): 990/2021

sjsfsu@su.edu.ly
journal.su.edu.ly/index.php/JSFSU

Scientific Journal for the Faculty of Science-Sirte University Vol. 2, No. 1 April (2022) 76-87

Scientific Journal for the Faculty of Science-Sirte University
Journal home page: http://journal.su.edu.ly/index.php/JSFSU/index
DOI: 10.37375/issn.2789-858X

Risk Factors for Ciprofloxacin and Gentamycin Resistance among Gram Positive and Gram Negative Bacteria Isolated from Community-Acquired Urinary Tract Infections in Benghazi City

Noor alhooda M. Alawkally[1], Sara El- warred [2], Abdlmanam Fakron[3], Hamza K. Ibrahim[4], Nessrine A. Suliman[5], Mareei A. Ali [1], Mailud El amari[6], Alreda M. Al-Awkally[7] and Mohammed A. A. Soutiyah[3]

[1]*Medical laboratory Department, Higher Institute of Science and Technology, Suluq, Libya.*

[2]*Statistic Department, Arts and Science Faculty, Benghazi University, Libya.*

[3]*Microbiology Department, Science Faculty, Omar Al-mukhtar University, Libya.*

[4]*Department of Pharmacy, Higher Institute of Medical Technology, Bani Waleed, Libya.*

[5]*Zoology Department, Science Faculty, Benghazi University, Libya.*

[6]*Health Education Department, Public Health Faculty, Benghazi University, Libya.*

[7]*Central of Pharmacy, Health Ministry, Darna, Libya.*

DOI: https://doi.org/10.37375/sjfssu.v2i1.204

ARTICLE INFO:

Received 30 December 2022.

Accepted 13 March 2022.

Published 17 April 2022.

Keywords: *Benghazi, Ciprofloxacin, Gentamycin, E. coli, Klebseilla pneumonia, Staph aureus, Urinary Tract Infection.*

A B S T R A C T

Urinary tract infections (UTIs) are a severe public health problem caused by a range of pathogens. The aims of the study were to investigate the prevalence of urinary tract infection and to see the pattern of Ciprofloxacin and Gentamycin susceptibility against uropathogens in Benghazi city. The study took place between 2021 April and October 2021. In-vitro antibacterial activity and resistance patterns of these two well-known antibiotics were studied and compared using the disk diffusion method. Laboratory reports and clinical data of patients with a positive urine culture (≥105 CFU/mL) were included in the study. Ciprofloxacin and gentamycin were tested against fourteen common bacterial pathogens, isolated from clinical samples of patients from Benghazi city Libya. A total of isolates were detected in 264 (75.4%) samples. Out of these, 75.4% were female and 24.6% were male. The majority of the study participants were in the age range of between 36-53 years (33.7%). The Increased bacterial resistance to ciprofloxacin has been shown. Most strains of *E. coli* and *Staph aureus* were resistant to ciprofloxacin and sensitive to gentamycin thereby showing that gentamycin is more effective than ciprofloxacin. This study showed that *E. coli* followed by *staph aureus* and *K. pneumonia* were the predominant uropathogen of UTIs in this geographical area. Most of the uropathogens were susceptible to Ciprofloxacin. The results showed that there is an alarming subject of resistance to Gentamicin against UTI patients in this area. Clinicians should be aware of the existing data and treat patients according to susceptibility patterns.

1 Introduction

Urinary tract infections (UTI) are among the most frequent bacterial infections affecting people both in the community and in hospitals. (Laupland *et al.*, 2007) it is estimated that about 150 million people are diagnosed with UTI worldwide per annum. (Gupta *et al.*, 2001) The problem of antibiotic resistance is severe in Libya. (Khalifa *et al.*, 1993) A recent World Health Organization (WHO) report on antimicrobial resistance

Corresponding author: E-mail: abdlmanam.fakron@omu.edu.ly

(AMR) surveillance specified nine bacteria of international concern which are responsible for some of the most common infections in community and hospital settings (WHO, 2014). Escherichia coli, the pathogen most often implicated in UTIs, is one of the nine. In all six WHO regions (Africa, Americas, Eastern Mediterranean, European, South-East Asia and Western Pacific), high rates of antimicrobial resistance have been observed in this pathogen (WHO, 2014). Ciprofloxacin belongs to the group of drugs called fluoroquinolones. Ciprofloxacin the most commonly prescribed fluoroquinolone for UTIs because it is available in oral and intravenous preparations. (Schaeffer, 2007) It is well absorbed from the gastrointestinal tract after oral administration. It also has a documented safety profile, broad Gram-negative organism coverage and high urinary excretion rate. (Schaeffer, 2007) During the last decade, the resistance rate of *E. coli* to fluoroquinolones such as ciprofloxacin has increased. (Mcquiston *et al*., 2013) Gentamicin Antibiotic Class Aminoglycoside. Mechanism of Action is the Inhibition of protein biosynthesis by irreversible binding of the aminoglycoside to the bacterial ribosome 30S subunit. (Wurtz R *et al*,. 1997) It was established through many studies that gentamicin produces oxidative stress in different cell types of the body accompanied with direct gonad toxic effect. (Sweileh, 2009; Denamur *et al*., 2011; Sobel, 2014). Aims of the study were to investigate the prevalence of urinary tract infection and to see the pattern of Ciprofloxacin and Gentamycin susceptibility against uropathogens in Benghazi city.

2 Materials and Methods

2.1 Study Area

The study was conducted at the Microbiology Department of Al saleem Medical Laboratory, Benghazi between 2021 April and October 2021. A total of 264 samples were included in this study.

2.2 Sample Collections

Urine specimens were collected from patients attending the al saleem Medical Laboratory. Urine was collected from patients into a sterile clean wide-mouth container. Upon collection, immediately urine was conveyed to the Microbiology Department.

2.1 Culture

Urine specimens were cultured within one hour of specimen collection on blood agar, MacConkey agar and CLED plate. They then incubated aerobically at 37oC for 24-48 h, whereas chocolate agar cultures were incubated at a 5% CO2 candle jar. (Denamur *et al*., 2011; Sobel, 2014). A culture that grew >105 units (CFU/mL) was considered significant bacteriuria.

2.4 Identification of bacterial isolates

After obtaining the pure strains, Gram-negative rods were identified with the help of a series of biochemical tests such as coagulase, catalase, oxidase, indole production, urease production, Triple sugar iron, simmons citrate utilization, motility, mannitol Salt agar, hydrogen sulphide production, nitrate/nitrite production, methyl red and voges Proskeur. (Mcquiston *et al*., 2013; (Wurtz *et al*, 1997; Sweileh, 2009) Morphologically identical colonies of the suspected strains were taken from the agar plates and were suspended in nutrient broth and vortexed. Then, the suspensions were inoculated into the butt and slant of the biochemical testing media. The inoculated media were aerobically incubated at 37°C and after overnight incubation bacteria were identified following the standard flow chart. Gram-positive cocci were determined based on their Gram reaction in catalase and coagulase tests (Cheesebrough, 2006; Baron *et al*., 1994).

2.4.1 Gram's Stain

The smear was made from the isolate on a clean grease-free slide and allowed to be air-dried and fixed. The smear was flooded with crystal violet as a primary stain and was allowed to stain for 2 minutes and rinsed with water. A mordant (lugol's iodine) was flooded, allowed to stay for 1 minute, and rinsed with water. Decolorize rapidly (few seconds) with acetone–alcohol. Wash immediately with clean water A smear was then inundated with secondary stain (neutral red) and was allowed to stain for 2 minutes and then rinsed in water and allowed to air dry. (Oladeinde *et al*., 2011).

2.4.2 Coagulase Test

Staphylococcus spp were further tested for the production of free coagulase enzyme using tube coagulase test. Coagulase test, a drop of plasma was placed on a clean dried slide. A drop of saline was placed next to the drop of plasma as a control. A portion of the isolated colonies was mixed in each drop with a loop, starting with the saline until a smooth suspension was obtained. Then, the suspension was mixed well and rocked gently for 5-10 seconds. (Eltahawy and Khalaf 1988).

2.4.3 Oxidase Test

A piece of filter paper was moisture with a substrate (1% tetramethyle -p-phenylene -diamine dihydrochloride). A wooden stick was used to remove a small portion of bacterial colony and streak across the wetted filter paper streaked area on wetted filter paper was observed for the color change to deep blue. (Oladeinde *et al*., 2011).

2.4.4 Dnase Test

Using a sterile loop, test and control organisms (ATCC 2923) were spot-inoculated and incubated at 35-37 overnight. The surface of the plate was covered with 1mol/ml hydrochloric acid solution and excess was tipped off. Clearing around each colony was observed within 5 minutes of adding the acid. (Oladeinde *et al*., 2011)

2.4.5 Analytical Profile Index (API) 20e Test

5ml ample of API Na cl, 0.85% medium, was opened. A single well- isolated colony from an isolation plate was removed using a pipette. It was carefully emulsified in 5ml ample of API Na cl 0.85% to obtain a homogeneous bacterial suspension. Using the same pipette, both tube and cupule of the test CIT, VP and GEL were full with the bacterial suspension. Anaerobiosis was created in ADH, LDC, ODC, H2S, and URE tests by overlaying mineral oil. The incubation box was closed and incubated at 36° C for 24 hours (Eltahawy and Khalaf 1988).

2.4.6 Urease Test

The surface of the urea slant agar was streaked with a portion of well-isolated colonies. The slanted cap was left on the loose and incubated at 35°C for 18-24 hours. (Oladeinde *et al*., 2011)**.**

2.4.7 Carbohydrate Utilization Test

0.1ml of a heavy saline suspension of the test organism was added to each of the four tubes containing glucose, lactose, maltose and sucrose carbohydrate disk and no to the fifth tube and was incubated at 37°C for 5 hours. It was examined at 30-minute intervals for up to 5 hours from red to yellow indicating carbohydrate utilization (Hummers *et al*., 2005).

2.4.8 Citrate Utilization Test

The surface of the Simmons citrate agar slant was streaked with a portion of a well-isolated colony. The slant cap was left on loosely and was incubated at 35°C for 18-24 hours. (Eltahawy and Khalaf 1988).

2.5 Antibiotic Susceptibility

Antibiotic susceptibility testing was done for the bacterial isolates identified from urine cultures with significant bacteriuria using the Kirby-Bauer disk diffusion method. (Bauer, 1966). The procedure for antimicrobial susceptibility testing is as follows: Briefly, 4–6 morphologically identical colonies of bacteria from pure cultures were collected with an inoculating loop and transferred into a tube containing 5 mL of nutrient broth, then mixed gently until a homogenous suspension was formed, and incubated at 37°C. Using a sterile nontoxic dry cotton swab, a sample of the standardized inoculums (turbidity was adjusted to obtain confluent growth) was taken and streaked on the entire surface of the dried Mueller–Hinton agar plate three times, turning the plate at 60° angle between each streaking to ensure even distribution. The inoculums were allowed to dry for 5–15 minutes with the lid in place. Using sterile forceps, the selected antibiotics disks were applied to the plates at a distance of 15 mm away from the edge and 24 mm apart from each other. After incubating the plates at 37°C for 24 hours, the diameters of the zone of bacterial growth inhibition around the disks were measured to the nearest millimetre. The susceptibility or resistance to the agent in each disk was determined, and the isolates were classified as sensitive (S), intermediate (I), or resistant (R) according to the standardized table. A ruler's zone of inhibition was measured in mm. (Donne *et al*., 2017; CLSI, 2014). The antibiotics tested were Gentamycin- CN (10µg). CIP Zone in diameter in mm (MIC) (R<=12, I 13-14, S>=15) and ciprofloxacin- CIP (5mcg). CN Zone in diameter in mm (MIC) (R<=15, I 16-20, S>=21).

file:///D:/F+NA%20U%202021/cip,cn,f,na/Gentamicin.

Statistical Analysis

The data was analyzed using SPSS programs version 20.

3 Results

A total of 264 (100.0%) urine samples were positive in 2021 (April-October) in the selected area.

3.1 Distribution table of Urinary Tract Infection (UTI) patients by genders

Isolates were detected in 264 (75.4%) samples. Out of these, 199 (75.4%) were female and 65 (24.6%) were male.

Table (1). Distribution table of Urinary Tract Infection (UTI) patients by genders

Gender	Frequency	Percent
Female	199	75.4
Male	65	24.6
Total	264	100.0

3.2 Distribution of the Cases by Age Group

Urinary tract infection and its association with age are presented in Table 2. Males aged 36-53 years old had a somewhat high prevalence (89/264:33.7%) of urinary tract infections. In the age group 72-89 years (n=23), the incidence of urinary tract infection is somewhat reduced to (8.7%).

Table (2). Distribution of the cases by age group.

Age	Frequency	Percent
0-17	33	12.5
18-35	85	32.2
36-53	89	33.7
54-71	34	12.9
72-89	23	8.7
Total	264	100.0

3.3 Distribution of Isolates in Clinical Specimens Collected from Patients

In the present study, Enterobacteraeae 65.1% was the most predominant 65.1% isolates. *Escherichia coli* spp (41.7%) and *Staph aureus* (15.9%) were the predominant organisms isolated from the study subjects. The other bacterial isolates include *Klebseilla pneumonia* (9.8%), *Klebsiella* spp (6.8%), *Strep agalactia* (5.7%), *Strep pyogen* (4.9%), *strep pneumonia* (4.2%), *Staph saprophyticus* and *Pseudomonas aeruginosa* (2.3%) equally, *Enterobacter* spp and *proteus* spp (1.5%) equally, *Acinetobacter* spp (1.1%), and *Citrobacter* spp (0.4%) as indicated in Table 3.

Table (3). Distributions of Isolates in Clinical Specimens Collected from Patients.

Bacteria	Frequency	Percent
***E. coli* spp**	110	41.7
Staph aureus	42	15.9
Klebsiella pneumonia	26	9.8
***Klebsiella* spp**	18	6.8
Strep agalactia	15	5.7
Strep pyogen	13	4.9
Strep pneumonia	11	4.2
Pseudomonas aeruginosa	6	2.3
Staph saprophyticus	6	2.3
***Enterococcus* spp**	5	1.9
***Enterobacter* spp**	4	1.5
***Proteus* spp**	4	1.5
***Acinetobacter* spp**	3	1.1
***Citrobacter* spp**	1	.4
Total	264	100.0

3.4 Prevalence of Urinary Tract Infection Among Tested Patients in Relation to Month

May (32.6%) was the most month in our study in which UTI cases were recorded followed by June (22.3%) and September (15.5%).

Table (4). Prevalence of urinary tract infection among tested patients in relation to the month**.**

Months	Frequency	Percent
April	39	14.8
May	86	32.6
June	59	22.3
July	8	3.0
August	30	11.4
September	41	15.5
October	1	.4
Total	264	100.0

3.5 Gender Distribution by Months

According to gender, the most UTIs was recorded in May (25.3%) followed by June (17.4%) in females. While in the males, the most UTIs were recorded in May (7.1%) followed by June and August (4.9%) equally.

Table (5). Gender distribution by months

	Gender		
Months	**Female**	**Male**	**Total**
April	30	9	39
May	67	19	86
June	46	13	59
July	6	2	8
August	17	13	30
September	33	8	41
November	0	1	1
Total	199	65	264

3.6 Prevalence of Different Uropathogens Among Male and Female Patients. `

In this study, the urinary tract infections of female patients (199) were more prone to male patients (65). In females, the most predominant uropathogen were *E. coli* 91 (34.4%) followed by *Staph aureus* 26 (9.8%) and *Klebsiella pneumonia* 23 (8.7%). In the male, the most prevalent uropathogens were *E. coli* 19 (7.1%) followed by *Staph aureus* 16 (6%) and *Strep pneumonia* 8 (3%). The study noted that] male patients were more infected by the entire isolated organism except organism *Enterococcus* spp.

Table (6). Prevalence of different uropathogens among male and female patients**.**

	Gender		
Uropathogen	**Female**	**Male**	**Total**
***E. coli* spp**	91	19	110
Staph aureus	26	16	42
Klebsiella pneumonia	23	3	26
***Klebsiella* spp**	16	2	18
Strep agalactia	10	5	15
Strep pyogen	9	4	13
Strep pneumonia	3	8	11
Pseudomonas aeruginosa	5	1	6
Staph saprophyticus	4	2	6
***Enterococcus* spp**	3	2	5
***Enterobacter* spp**	4	0	4
***Proteus* spp**	3	1	4
***Acinetobacter* spp**	1	2	3
***Citrobacter* spp**	1	0	1
Total	199	65	264

3.7 Distribution of Different Age Groups of UTI Patients by Months

36-53 was the most age group of UTI patients recorded in the present study, and it recorded the most frequent cases in May.

Table (7). Distribution of different age groups of UTI patients by months.

	Age					
Month	**0-17**	**18-35**	**36-53**	**54-71**	**72-89**	**Total**
April	10	6	9	6	8	39
May	13	28	29	12	4	86
June	5	24	15	9	6	59
July	0	4	4	0	0	8
August	1	9	16	3	1	30
September	4	14	15	4	4	41
October	0	0	1	0	0	1
Total	33	85	89	34	23	264

3.8 Distribution of Isolates by Ages

The most isolated age group from which *E. coli* was among 18-38 (12.5%), followed by 36-53 (10.9%), followed by 0-7 and 54-71 (6.4%) equally and the least isolated were at 72-18 (4.5%).

Table (8). Distribution of isolates by ages.

Age	**0-17**	**18-35**	**36-53**	**54-71**	**72-89**	**Total**
***Acinetobacter* spp**	0	1	2	0	0	3
***Citrobacter* spp**	0	0	1	0	0	1
***E.coli* spp**	18	33	29	18	12	110
***Enterobacter* spp**	0	1	3	0	0	4
***Enterococcus* spp**	0	2	2	1	0	5
***Klebsiella* spp**	2	7	4	1	4	18
Klebsiella pneumonia	6	8	5	4	3	26
Pseudomonas aeruginosa	0	1	4	0	1	6
***Proteus* spp**	0	1	3	0	0	4
Staph aureus	6	15	16	4	1	42
Staph saprophytics	1	3	1	1	0	6
Strep agalactia	0	6	6	1	2	15
Strep pneumonia	0	3	7	1	0	11
Strep pyogen	0	4	6	3	0	13
Total	33	85	89	34	23	264

3.9 Distribution of Susceptibility Pattern of Gentamicin Against Uropathogens by Months

The bacteria showed the most resistance to Gentamycin in May, while the most sensitive isolates were recorded in May.

Table (9). Susceptibility pattern of Gentamicin against uropathogens.

Month/Susceptibility	**Gentamycin**				
	Miss	**Intermediate**	**Resistant**	**Sensitive**	**Total**
April	2	5	8	24	39
May	42	8	8	28	86
June	36	3	10	10	59
July	3	0	1	4	8
August	19	1	6	4	30
September	31	4	5	1	41
October	0	0	1	0	1
Total	133	21	39	71	264

3.10 Antibiotic Sensitivity, Resistance and Intermediate Sensitivity of Bacteria Isolated from Urine Culture to Gentamycin.

The susceptibility patterns of the bacterial isolates to Gentamycin antibiotic are presented in Table 5. From the results, 26.9% of isolates were most sensitive to

Gentamycin. Percentage resistance of isolates to Gentamycin antibiotics was 14.8%. The activity of Gentamycin against the isolates was somewhat acceptable.

Table (10). Antibiotic sensitivity, resistance and intermediate sensitivity of bacteria isolated from urine culture to Gentamycin.

Gentamycin		
Susceptibility patterns	**Frequency**	**Percent**
Miss	133	50.4
Intermediate	21	8.0
Resistant	39	14.8
Sensitive	71	26.9
Total	264	100.0

3.11 Distribution of Susceptibility Pattern of Ciprofloxacin Against Uropathogens by Months

Table (11). Susceptibility pattern of Ciprofloxacin against uropathogens

Ciprofloxacin					Total
Month	**Miss**	**I**	**R**	**S**	
April	5	4	16	14	39
May	5	8	33	40	86
June	12	7	23	17	59
July	4	3	1	0	8
August	6	2	9	13	30
September	2	7	14	18	41
October	0	0	0	1	1
Total	34	31	96	103	264

Note: R: Resistant; I: Intermediate; S: Sensitive.

3.12 Antibiotic sensitivity, resistance and intermediate sensitivity of bacteria isolated from urine culture to Ciprofloxacin.

The susceptibility patterns of the bacterial isolates to Ciprofloxacin antibiotic are presented in Table 6. From the results, 39% of isolates were most sensitive to Ciprofloxacin. Percentage resistance of isolates to Ciprofloxacin antibiotics was 36.4%, the activity against the isolates was also somewhat acceptable.

Table (12). Antibiotic sensitivity, resistance and intermediate sensitivity of bacteria isolated from urine culture to Ciprofloxacin.

Ciprofloxacin		
Susceptibility	**Frequency**	**Percent**
Miss	34	12.9
Intermediate	31	11.7
Resistant	96	36.4
Sensitive	103	39.0
Total	264	100.0

3.13 Sensitivity of Gram Positive and Gram Negative Organism Groups to Ciprofloxacin

The Ciprofloxacin antibiotic has sensitive against almost all of the isolates, *E. coli* (17%), *staph aureus* (4.9%), *Klebsiella pneumonia* (4.1%). Ciprofloxacin exhibited good antibacterial activity against *Escherichia coli* more than Gentamycin, but these activities to both of them were also not 100%.

Table (13). Sensitivity of gram positive and gram negative organism groups to Ciprofloxacin

Ciprofloxacin					
Bacteria	**Miss**	**Intermediate**	**Resistant**	**Sensitive**	**Total**
Acinetobacter spp	0	0	1	2	3
***Citrobacter* spp**	0	0	1	0	1
***E. coli* spp**	17	13	35	45	110
***Enterobacter* spp**	0	0	1	3	4
***Enterococcus* spp**	2	0	1	2	5
***Klebsiella* spp**	0	2	9	7	18
Klebsiella pneumonia	2	7	6	11	26
Pseudomonas aeruginosa	0	1	1	4	6
***Proteus* spp**	0	2	0	2	4
Staph aureus	5	4	20	13	42
Staph saprophytics	2	0	2	2	6
Strep agalactia	4	0	3	8	15
Strep pneumonia	0	1	8	2	11
Strep pyogen	2	1	8	2	13
Total	34	31	96	103	264

3.14 Sensitivity of Gram Positive and Gram Negative Organism Groups to Gentamycin

The Gentamycin antibiotic has sensitive against almost all of the isolates, *E. coli* (13.2%), *staph aureus* (5.3%), *Enterobacter* spp (6%), *Klebsiella pneumonia* (1.8%). Gentamycin has antibacterial activity against *Escherichia coli* but this activity is not 100%

Table (14). Sensitivity of gram positive and gram negative organism groups to Gentamycin.

	Gentamycin				
Bacteria	**Miss**	**Intermediate**	**Resistant**	**Sensitive**	**Total**
***Acinetobacter* spp**	2	1	0	0	3
***Citrobacter* spp**	1	0	0	0	1
***E. coli* spp**	51	9	15	35	110
***Enterobacter* spp**	1	0	1	2	4
***Enterococcus* spp**	1	0	2	2	5
***Klebsiella* spp**	12	2	2	2	18
Klebsiella pneumonia	15	2	4	5	26
Pseudomonas aeruginosa	3	1	1	1	6
***Proteus* spp**	3	0	1	0	4
Staph aureus	20	2	6	14	42
Staph saprophytics	2	0	1	3	6
Strep agalactia	7	0	5	3	15
Strep pneumonia	8	1	1	1	11
Strep pyogen	7	3	0	3	13
Total	133	21	39	71	264

3.15 Distribution of Isolates by Months

The highest isolation of *E. coli* was in May, followed by June.

Table (15). Distribution of isolates by months**.**

	Month							
Bacteria	**April**	**may**	**June**	**July**	**August**	**September**	**October**	**Total**
Acinetobacter spp	1	0	0	0	0	2	0	3
***Citrobacter* spp**	0	1	0	0	0	0	0	1
***E. coli* spp**	20	41	30	0	5	14	0	110
***Enterobacter* spp**	0	2	0	0	0	2	0	4
***Enterococcus* spp**	0	0	4	0	0	1	0	5
***Klebsiella* spp**	1	6	7	0	2	2	0	18
Klebsiella pneumonia	4	8	5	1	3	5	0	26
Pseudomonas aeruginosa	2	3	0	0	0	1	0	6
Proteus spp	0	1	0	0	1	2	0	4
Staph aureus	5	12	7	3	10	4	1	42
Staph saprophytics	0	4	0	0	2	0	0	6
Strep agalactia	2	3	1	1	5	3	0	15
Strep pneumonia	1	3	4	1	0	2	0	11
Strep pyogen	3	2	1	2	2	3	0	13
Total	39	86	59	8	30	41	1	264

3.16 Distribution of Susceptibility Patterns of Isolates to Gentamycin by Sex.

In this study, a total of 264 isolates from urine specimens were tested in vitro by the disk diffusion test to determine the susceptibility of these bacteria to Ciprofloxacin and Gentamycin.

Isolates that were resistant to Gentamycin were observed in females more than males.

Table (16). Distribution of susceptibility patterns of isolates to Gentamycin by sex.

Gentamycin					
Susceptibility pattern	**Miss**	**Intermediate**	**Resistant**	**Sensitive**	**Total**
Female	100	18	27	54	199
Male	33	3	12	17	65
Total	133	21	39	71	264

3.17 Distribution of Susceptibility Patterns of Isolates to Ciprofloxacin by Sex.

Isolates were resistant to Ciprofloxacin also was observed in females more than males.

Table (17). Distribution of susceptibility patterns of isolates to Gentamycin by sex.

Ciprofloxacin					
Susceptibility patterns	**Miss**	**Intermediate**	**Resistant**	**Sensitive**	**Total**
Female	30	22	66	81	199
Male	4	9	30	22	65
Total	34	31	96	103	264

4 Discussion

Urinary tract infections are primarily caused by gram-negative bacteria, but gram-positive pathogens may also be involved. More than 95% of uncomplicated UTIs are monobacterial. The most common pathogen for basic UTIs is *E. coli* (75%–95%), followed by *Klebsiella pneumoniae*, *Staphylococcus saprophyticus*, *Enterococcus faecalis*, group B streptococci, and *Proteus mirabilis*. Sobel, 2014).

This study aimed to determine the causative bacterial agent of urinary tract infection among different groups in Alsaleem Medical Laboratory, Benghazi. Bacterial pathogens were isolated from 75.4% of the requested urine culture. The overall prevalence of UTI was 75.4% in this study. This was similar to the prevalence of UTI reported from Das RN *et al* isolation rate was 71.6%, (Das *et al*., 2006) and Latika *et al* 76.29%. Other studies done in Karnataka, western India, and South India reported 71.72%, 76.2 and 71.72%, respectively. (Latika *et al*., 2015; Razak and Gurushantappa 2012).

The selection of empiric antibiotics for UTIs should be based on the severity of the infection and local susceptibility patterns. When antibiotics are indicated, short courses are effective for uncomplicated UTIs, especially cystitis, and otherwise healthy women.

The common of patients with UTIs were females in the current study. This is expected and is likely the result of the anatomy of the female urinary tract compared to their male counterparts, particularly the shorter female urethra and closer proximity to the anus. (Hummers *et al*., 2005).

This finding is in line with previous in multiple countries, (Oladeinde *et al*., 2011; Bitew *et al*., 2017; Alanazi *et al*., 2018; Al Yousef *et al*., 2016) where *E. coli* was identified as the primary causative bacterium of UTIs, followed by *Staph aureus* and *Klebseilla pneumonia*, (Al Yousef *et al*., 2016; Al-Harthi and Al-Fifi 2008; Carlos *et al*., 2007; Abir *et al*., 2021) which correlates with findings from another study in Iran which revealed that uropathogens with a predominance of *Escherichia coli* (38%) and *Staphylococcus* spp (35%). (Mihankhah *et al*., 2017).

As we expected, *Escherichia coli* was by far the most frequent pathogen isolated in the medical centers evaluated and is probably the most frequent pathogen causing UTIs in Latin American hospitals. (Abir *et al*., 2021; Sader *et al*., 1999).

Antibiotic resistance is common in developing countries such as Libya, where drugs are available freely without prescription. An antibiotic stewardship program could provide educational programs and cascade the reporting of antibiotic susceptibility results as effective strategies to improve antibiotic prescribing behavior. The drug susceptibility profile of Gram-negative and Gram-positive bacteria tested in the present study was variable. Ciprofloxacin is the most commonly used antibacterial drug in treating community-acquired UTIs. (Hryniewicz *et al*., 2001) Gram negative organisms, particularly *E. coli* are commonly associated with UTI in children in developing countries. (Carlos *et al*., 2007; Jeena *et al*., 1995; Jeena *et al*., 1996; Kala and Jacobs 1992; Rabasa and Shattima 2005).

Our investigator was showed, 6.4% of *E. coli* was isolated from patients their ages between 0-17. More than half of the *Escherichia coli* and *K. pneumonia* isolates were resistant to ciprofloxacin followed by gentamycin in the current study. *E. coli* and *Klebsiella pneumonia* were resistance to the fluoroquinolones (Ciprofloxacin) were observed for 13.2 % - 3.4 % respectively, while they were resistant to the Aminoglycosid (gentamycin) was observed for 5.6% - 1.5%. *Escherichia coli* are also a significant cause of other kinds of nosocomial infections. (Sader *et al*., 1998; Pfaller *et al*., 1998) In addition to *Escherichia coli*, fluoroquinolone resistance was also high among other species, and cross-resistance to newer compounds was very common. The increasing fluoroquinolone resistance documented in this study may, due to the extensive use of fluoroquinolones, ultimately risk and the use of this important class of antibiotics in the region. However, further epidemiologic studies are necessary to improve our understanding of this problem.

The fluoroquinolones (Ciprofloxacin) was effective against many strains more than aminoglycosides (gentamicin), with 14.8%. Similar findings were seen in studies by (Abir *et al*., 2021; Butler *et al*., 2015), who concluded that the organisms exhibited utmost resistance (80.00%) against Ciprofloxacin.

This study matches results reported by (Khalifa *et al*., 1993) who revealed that Ciprofloxacin was the most effective antimicrobial agent. Therefore, we observed a higher frequency of ciprofloxacin-sensitive in *E. coli* (17%) when compared to *Klebsiella pneumonia* (4.1%). (Falagas *et al*., 2010).

Additionally, 13.2% of the *E. coli* was resistant to ciprofloxacin. Although with a different methodology, the present study demonstrated similar *E. coli* resistance rates compared to the Hummers-Pradier clinical study. (Carlos *et al*., 2007; Hummers *et al*., 2005).

The results of this study agreed with other studies that dictated that uropathogens are always predictable and *E. coli* are the leading causes, besides other common Gram negative organisms as *Klebsiella*, Enterobacter, Proteus and *Citrobacter* species (Sahm *et al*., 2001). All isolated bacteria in this study belonged to Enterobacteriaceae that can live in the digestive tract, rectum, and vagina or around the urethra. Infection occurs when these bacteria enter the normally sterile urinary system and multiply (Patterson and Andriole 1987). Similarly, Enterobacteriaceae is the predominant (78.7%) isolates, of which *E. coli* was the most (64.0%) common organisms followed by *Klebsiella* species (17.9%) (Mohammed *et al*., 2016; Thakur *et al*., 2013) and *P. aeruginosa* (2.3%), *Acinetobacter baumannii* and *Proteus* species are very often isolated in hospitals (Rampure, 2013)

The gram-negative bacteria were the most common isolates in the current study, obtained in the present study was different in rates with other reports from different areas. (Mohammed *et al*., 2016; Guermazi *et al*., 2018; Salim *et al*., 2017; Mostafa *et al*., 2016) Gram-positive bacteria were *Staphylococcus aureus* 42 (15.9%) followed by *Strep agalactia* 15 (5.7%) of the isolate's strains.

Gentamicin is an antibiotic widely used in Australian hospitals. It is known to lawyers due to its damage to the apparatus inside the kidney (nephrotoxicity). (Sweileh, 2009) In general, the broad-spectrum ciprofloxacin was the antibiotic with the highest activities, followed by Gentamycin in the present study.

This study detected the dominance of *Escherichia coli* spp (41.7%) and *Klebsiella pneumonia* (9.8%) (Table 2), which was almost identical compared with other research in Libya and other countries. In Northwest Libya, Abujnah *et al*. have found a predominance of *Escherichia coli* (56%) and *Klebsiella pneumonia* (19%). *Escherichia coli* spp (41.7%) followed by *Klebseilla pneumonia* (9.8%). In another study in Messalata, Libya, Mahammed *et al*, have reported the predominance of Escherichia coli (56%) and Klebsiella pneumonia (17%). (Mohammed *et al*., 2016) In Southern Tunisia, the authors have found *Escherichia coli* (68%) and *Klebsiella pneumonia* (13%) as predominance uropathogens among patients of UTIs. (Guermazi *et al*., 2018) A study in Iran has reported uropathogens with a predominance of *Escherichia coli* (38%) and *Staphylococcus* spp (35%). (Mihankhah *et al*., 2017) *Staph aureus* (15.9%) and *Strep agalactia* (5.7%) were the most dominant Gram positive uropathogen isolated in our study. Unlike in other studies which isolated coagulase-negative Staphylococcus and Enterococcus as the most dominant Gram-positive uropathogen. (Ayoyi *et al*., 2017; Okonko *et al*., 2009).

Our study also showed a high prevalence of UTI in females than males 199 (75.4%) and 65 (24.6%), respectively. This correlates with findings from other studies that revealed that UTI frequency is more significant in females than males. (Gilbert *et al*., 2018; Prakash and Saxena 2013).

This result also agrees with what was previously reported by Mahmoud and colleagues in 2016. Many other researchers have also reported similar findings. (Butler *et al*., 2015; Foxman, 2014) The reason behind this high prevalence of UTI in females is due to the proximity of the urethral meatus to the anus, shorter urethra, sexual intercourse, incontinence, and bad toilet. (Nili and Asasi 2002).

The drug susceptibility profile of Gram-negative and Gram-positive bacteria tested in the present study was variable. For instance, increased bacterial resistance to ciprofloxacin has been shown. This study is opposite to the results reported by (Guermazi *et al*., 2018) who revealed that Ciprofloxacin was the most effective antimicrobial agent. Therefore, we observed a higher frequency of ciprofloxacin-resistant in *E. coli* (57.14%) when compared to *K. pneumoniae* (16.67%).

5 Conclusions

In conclusion, the problem of antibiotic resistance is very serious in Libya and appears to be on the rise. These results showed that there is a high prevalence of occurrence of urinary tract infections among patients in a selected area of Benghazi city. This study finding showed that *E. coli* was the predominant uropathogen was isolated from most samples. Most of the uropathogens were susceptible to Ciprofloxacin and seem somewhat appropriate for the empirical treatments of community-acquired UTI. The results showed that there is an alarming subject of resistance of Gentamicin against UTI patients in this area. Clinicians should be aware of the existing data and treat patients according to susceptibility patterns. To reduce the risk of a UTI, it's best to wipe genitals from front to back after using the bathroom, drink plenty of fluids, urinate before and after sex and get antibiotics.

Acknowledgements

We would like to thank all medical staff of the Microbiology Department of Al saleem Medical Laboratory for their helpful assistance and cooperation.

Conflict of Interest: The authors declare that there are no conflicts of interest.

References

Abir, B. A; Hamida, E; Asma, E; Alsharif.. P. (2021). revalence and Antibiotics Susceptibility Pattern of Urine Bacterial Isolates from Tripoli Medical Center (TMC), Tripoli, Libya. IBEROAMERICAN JOURNAL OF MEDICINE 03 221-226

Al Yousef, S. A; Younis, S; Farrag, E; Moussa, H; Bayoumi, F. S; Ali, A. M. (2016). Clinical and laboratory profile of urinary tract infections associated with extended spectrum β-lactamase Producing Escherichia coli and Klebsiella pneumoniae. Ann Clin Lab Sci. 46(4):393–400.

Alanazi, M. Q; Alqahtani, F. Y; Aleanizy, F. S. (2018). An evaluation of E. coli in urinary tract infection in emergency department at KAMC in Riyadh, Saudi Arabia: retrospective study. Ann Clin Microbiol Antimicrob. 17(1):3. doi:10.1186/s12941-018-0255-z

Al-Harthi ,A. A; Al-Fifi, S. H. (2008). Antibiotic resistance pattern and empirical therapy for urinary tract infections in children. Saudi Med J. 29 (6):854–858.

Ayoyi, A. O; Kikuvi, G; Bii, C; Kariuki, S.(2017). Prevalence, aetiology and antibiotic sensitivity profile of asymptomatic bacteriuria isolates from pregnant women in selected antenatal clinic from Nairobi, Kenya. Pan Afr Med J. 26:41. doi: 10.11604/pamj.2017.26.41.10975.

Baron, E. J; Peterson, L. R; Finegold, S. M; Bailey and Scott’s. (1994). Diagnostic Microbiology. 9th ed. St Louis: Mosby; 1994:249–257.

Bauer, A. W; Kirby, W. M. M; Sherris, J. C; Turk, M. (1966). Antibiotic susceptibility testing by a standardized single disc method. Am J Clin Pathol. 45: 493–496.

Bitew, A; Molalign, T; Chanie, M. (2017). Species distribution and antibiotic susceptibility profile of bacterial uropathogens among patients complaining urinary tract infections. BMC Infect Dis. 17(1):654. doi:10.1186/s12879-017-2743-8

Butler, C. C; Hawking ,M. K; Quigley, A; McNulty, C. A. (2015). Incidence, severity, helps seeking, and management of uncomplicated urinary tract infection: a population-based survey. Br J Gen Pract. 65(639):e702-7. doi: 10.3399/bjgp15X686965.

Carlos, R; Kiffer,, Caio, M; Carmen, P; Oplustil, J. L; Sampaio. (2007). Antibiotic Resistance and Trend of Urinary Pathogens in General Outpatients from a Major Urban City. International Braz J Urol Vol. 33 (1): 42-49.

Cheesebrough, M. (2006). District Laboratory Practice in Tropical Countries Part II. 2nd ed. London: Cambridge University Press; 105–114.

Clinical Laboratory Standards Institute (CLSI). (2014). Performance Standards for Antimicrobial Susceptibility Testing; Twenty-Fourth Informational Supplement. CLSI Document M100-S24. Vol. 34. Wayne, PA: Clinical and Laboratory Standards Institute; 50–57.

Das, R. N; Chandrashekhar, T. S; Joshi, H. S; Gurung, M; Shrestha, N; Shivananda, P. G. (2006). Frequency and susceptibility profile of pathogens causing urinary tract infections at a tertiary care hospital in western Nepal. Singapore Med J 47(4) : 281.

Denamur, S; Tyteca, D; Marchand, B. J; Van, B. F; Tulkens, P. M; Courtoy, P. J *et al*. (2011). Role of oxidative stress in lysosomal membrane permeabilization and apoptosis induced by gentamicin, an aminoglycoside antibiotic. Free Radical Biol Med. 51(9):1656-65.

Donne, C; Hershed, H; Mary, W; Carrofl, B. (2013). Microbiology Laboratory Manual Bio/2421L.

Eltahawy, A. T; Khalaf, R. M. F. (1988). Urinary tract infection at a University Hospital in Saudi Arabia: incidence, microbiology, and antimicrobial susceptibility. Ann Saudi Med. 8(4):261–266. doi:10.5144/ 0256-4947.1988.261.

Falagas, M. E; Kastoris, A. C; Kapaskelis, A. M; Karageorgopoulos, D. E. (2010). Fosfomycin for the treatment of multidrug-resistant, including extendedspectrum beta-lactamase producing, Enterobacteriaceae infections, a systematic review. Lancet Infect. Dis. 10:43–50.

file:///D:/F+NA%20U%202021/cip,cn,f,na/Gentamicin.

Foxman, B. (2014). Urinary tract infection syndromes: occurrence, recurrence, bacteriology, risk factors, and disease burden. Infect Dis Clin North Am. 28(1):1-13. doi: 10.1016/j.idc.2013.09.003

Gilbert, J. A; Blaser, M. J; Caporaso, J. G; Jansson, J. K; Lynch, S. V; Knight, R. (2018). Current understanding of the human microbiome. Nat Med. 24(4):392- 400. doi: 10.1038/nm.4517.

Guermazi, T. S; Boujlel, S; Assoudi, M; Issaoui, R; Tlili, S; Hlaiem, M. E. (2018). Susceptibility profiles of bacteria causing urinary tract infections in Southern Tunisia. J Glob Antimicrob Resist. 12:48-52. doi: 10.1016/j.jgar.2017.09.004.

Gupta, K; Hooton. T. M; Stamm. W. E. (2001). Increasing antimicrobial resistance and the management of uncomplicated community-acquired urinary tract infections. Ann Intern Med. 135:41–50.

hakur, S; Pokhrel, N; Sharma, M. (2013). Prevalence of multidrug resistant Enterobacteriaceae and extended spectrum b lactamase producing Escherichia coli in urinary tract infection. Res J Pharm Biol Chem Sci; 4(2): 1615-1624.

Hryniewicz, K; Szczypa, K; Sulikowska, A *et al*. (2001). Antibioticsusceptibility of bacterial strains isolated from urinary tract infections in Poland. J Antimicrob Chemother. 47: 773–80

Hummers, P. E; Koch, M; Ohse, A. M; Heizmann, W. R; Kochen, M. M. (2005). Antibiotic resistance of urinary pathogens in female general practice patients. Scand J Infect Dis. 37: 256-61.

Hummers, P. E; Ohse, A. M; Koch, M. Heizmann, W. R; Kochen, M. M. (2005). Management of urinary tract infections in female general practice patients. Fam Pract. 22(1):71–77. doi:10.1093/ fampra/cmh720.

Jeena, P; Coovadia, H, *et al*. (1995). A prospective study of bacteriuria and pyuria in catheter specimens from hospitalized children, Durban, South Africa. Annals of Tropical Paediatrics 15: 153-158.

Jeena, P; Coovadia, H, *et al*. (1996). Bacteriuria in children attending a primary health care clinic: a prospective study of catheter stream urine samples. Annals of Tropical Paediatrics 16: 293-298.

Kala, U and Jacobs, W. (1992). Evaluation of urinary tract infection in malnourished black children. Annals of Tropical Paediatrics 12: 75-81.

Khalifa, S. G; Amal, R; Khaled, T; Abdulaziz, Z and Ezzedin, F. (1993). Antimicrobial resistance in Libya: 19702011. ISSN: 2820 (Print) 1819-6357.

Latika, J. S; Geeta, M. V; Hetvi, M. (2015). URINARY TRACT INFECTION: BACTERIOLOGICAL PROFILE AND ITS ANTIBIOTIC SUSCEPTIBILITY IN WESTERN INDIA. NJMR│Volume 5│Issue 1│

Laupland, K; Ross. T; Pitout .J; Church. D; Gregson. D. (2007). Community-onset urinary tract infections: a population-based assessment. Infection. 35:150–3.

Mcquiston, H. J; Rosborg, D. M; Sternhagen, N. A. B; Llor, C; Bjerrum, L. (2013). Different recommendations for empiric first-choice antibiotic treatment of uncomplicated urinary tract infections in Europe. Scand J Prim Health. 31:235–40

Mihankhah, A; Khoshbakht, R; Raeisi, M; Raeisi, V. (2017). Prevalence and antibiotic resistance pattern of bacteria isolated from urinary tract infections in Northern Iran. J Res Med Sci. 22:108. doi: 10.4103/jrms.JRMS_889_16.

Mihankhah, A; Khoshbakht, R; Raeisi, M; Raeisi, V.(2017). Prevalence and antibiotic resistance pattern of bacteria isolated from urinary tract infections in Northern Iran. J Res Med Sci. 22:108. doi: 10.4103/jrms.JRMS_889_16.

Mohammed, M. A; Alnour, T. M; Shakurfo, O. M; Aburass, M. M. (2016). Prevalence and antimicrobial resistance pattern of bacterial strains isolated from patients with urinary tract infection in Messalata Central Hospital, Libya. Asian Pac J Trop Med. 9(8):771-6. doi: 10.1016/j.apjtm.2016.06.011.

Mostafa, M. M; Albakosh, A. M; Alrtail, A; Rzeg, M. M and Aboukay,A. M. (2016). Etiology of uropathogenic bacteria in patients with urinary tract infection in Zliten, Libya. J Human Applied Sci.;29:16-32.

Nili, H & Asasi, K. (2002). Natural cases and an experimental study of H9N2 avian influenza in commercial broiler chickens of Iran. Avian Pathology, 31(3), 247-252.

Okonko, I. O, Ijandipe, L. A; Ilusanya ,O. A; Donbraye, E. O. B; Ejembi, J; Udeze, A. O, *et al*. (2009). Incidence of urinary tract infection (UTI) among pregnant women in Ibadan, South-Western Nigeria. Afr J Biotechnol. 8(23):6649- 57.

Oladeinde, B. H; Omoregie, R; Olley, M; Anunibe, J. A. (2011). Urinary tract infection in a rural community of Nigeria. N Am J Med Sci. 2011;3 (2):75–77. doi:10.4297/najms.2011.375.

Oladeinde, B. H; Omoregie, R; Olley, M; Anunibe, J. A.(2011). Urinary tract infection in a rural community of Nigeria. N Am J Med Sci.;3 (2):75–77. doi:10.4297/najms.2011.375

Patterson, T. F; Andriole, V. T. (1987). Bacteriuria in pregnancy. Infect Dis Clin N Am 1(4): 807-822.

Pfaller, M. A; Jones, R. N; Doern, G. V; Kugler, K. (1998). The Sentry Participants Group. Bacterial pathogens isolated from patients with blood stream infection: frequencies of occurrence and antirmcrobial susceptibility patterns from SENTRY Antimicrobial Surveillance Program (United States and Canada, 1997). Antimicrob Agents Chemother 42: 1762-70

Prakash, D; Saxena, R. S. (2013). Distribution and antimicrobial susceptibility pattern of bacterial pathogens causing urinary tract infection in urban community of meerut city, India. ISRN Microbiol. 749629. doi: 10.1155/2013/749629.

Rabasa, A and Shattima, D. (2002). Urinary tract infection in severely malnourished children at the University of Maidugiri Teaching Hospital. Journal of Tropical Pediatrics 48: 359-361.

Rampure, R; Gangane, R; Oli, A. K. (2013). Prevalence of MDR-ESBL producing Klebsiella pneumoniae isolated from clinical samples. J Microbiol Biotechnol Res 3(1): 32-39.

Razak, S. K; Gurushantappa, V. (2012). Bacteriology of urinary tract infection and antibiotic susceptibility pattern in a tertiary care hospital in South India. Int J Med Sci Public Health. 1:109-112

Sader *et al*. (1999). Antimicrobial suseptibility of bacteria causing urinary tract infections. Clinical Microbiology and infection, V o l u m e 5 Number 8.

Sader, H. S, Jones, R. N. Gales, A. C, *et al*. (1998). Antimicrobial susceptibility patterns for pathogens isolated from patients in Latin American medical centers with a diagnosis of pneumonia: analysis of results from the SENTRY antimicrobial surveillance program (1997). Diagn Microbiol Infect Dis 31: 289-301.

Sahm, D. F; Thornsberry, C; Mayfield, D. C; Jones, M. E; Karlowsky, J. A. (2001). Multidrug resistant urinary tract isolates of Escherichia coli: prevalence and patient demographics in the United States in antimicrobial agents and chemotherapy. Antimicrob Agents & Chemother. 45(5): 1402-1406.

Salim, F. A; Murad, S. K; Elbareg, A. M. (2017). Isolation of bacterial pathogens causing urinary tract infections and their antimicrobial susceptibility pattern among patients at Misurata teaching hsospital, Libya. Microbiol. Infect Dis. 1(2):1-5.

Schaeffer, A. J. (2002). The expanding role of fluoroquinolones. Am J Med. 113:45–54.

Sobel, J. D; Kaye, D. (2014). Urinary tract infections. In: Mandell GL, Bennett JE, eds. Principles and Practice of Infectious Diseases, 8th ed. Philadelphia: Elsevier Saunders, 886-913.

Sweileh, W. M. (2009). A prospective comparative study of gentamicin-and amikacin-induced nephrotoxicity in patients with normal baseline renal function. Fundamental Clin Pharmacol. 23(4):515-20.

World Health Organisation. (2014). Antimicrobial resistance: global report on surveillance. http://www.who.int/drugresistance/documents/surveillancereport/en/. Accessed 25 Oct 2014.

Wurtz R *et al*,. 1997 Antimicrobial Dosing in Obese Patients. Clinical Infectious Diseases 25: 112-8**.**

Scientific Journal for the Faculty of Science-Sirte University - SJFSSU

sjsfsu@su.edu.ly
journal.su.edu.ly/index.php/JSFSU

تصميم : عبدالله نصرالدين