#### MULTI-DRUG RESISTANT (MDR) ESCHERICHIA COLI ORIGINATED FROM CLINICAL AND ENVIRONMENTAL SOURCES IN ISMAILIA-EGYPT

Amira M. Zakaria Biotechnology Research Center Suez Canal University EGYPT Mohamed H. Abdel Aziz Faculty of Science Suez Canal University EGYPT Samy A. Selim. Faculty of Science Al Juof University SUADI ARABIA

Corresponding Author Email: <a href="mailto:amirazakaria2000@yahoo.com">amirazakaria2000@yahoo.com</a>

#### ABSTRACT

The exaggerated use of antibiotics has led to the selection of new strains of bacteria that resist to antibiotics, a situation which is found in the case of Escherichia coli strains. This study was conducted to evaluated the antibiotic resistance profiles of Escherichia coli (E. coli), isolated from different Clinical and Environmental sources in Egypt. A total of 384 samples from human, animal and environmental sources were collected from different locations in Ismailia, Egypt. E. coli isolates (n = 283) were identified by conventional microbiology culture and were phenotypically characterized using biochemical and motility tests. From the overall number of E. coli isolates, 31.4% (89/283) were isolated from stools of humans with diarrhea, 17.3% (49/283) from stools of sheep, cattle and chicken with diarrhea, 16.6% (47/283) from urine of humans with urinary tract infection, 17.3% (49/283) from surfaces water, 6.4% (18/283) from sea-food, 6% (17/283) from processed meat products, 3.9% (11/283) from dairy products and 1.1% (3/283) from poultry products (liver). The antibiotic resistance patterns showed that the isolates carried multi-drug resistance (MDR) phenotype to at least four commonly used antibiotics belonging to different classes: Erythromycin (E), Gentamicin (CN), Cefazolin (CZ), Thiampinicol (TP), Vancomycin (VA), Ciprofloxacin (CIP) and Ampicillin (AM).

Keywords: Escherichia Coli, Antibiotics, Egypt, MDR.

# INTRODUCTION

*Escherichia coli* (*E. coli*) bacteria normally live in the intestines of people and animals. Most *E. coli* are harmless and actually are an important part of a healthy human intestinal tract. However, some *E. coli* are pathogenic, meaning they can cause illness, either diarrhea or illness outside of the intestinal tract. The types of *E. coli* that can cause diarrhea can be transmitted through contaminated water or food, or through contact with animals or persons (CDC 2003).

Antibiotic resistance in *E. coli* has been globally identified in isolates from environmental, animal and human sources (Dromigny JA 2005). This is a consequence of the use of antimicrobials in medicine and their application in animal husbandry, which have brought about phenotypic changes, often due to chromosomal mutations (Erb A *et al* 2007). Studies have shown that many pathogenic organisms have developed some degree of resistance to antimicrobials and they confer resistance through different mechanisms, with a negative impact on veterinary and human medicine. (Yismaw G 2010) These mechanisms of resistance include the alteration of receptor-binding sites of drugs, a decreased intake of drugs

by altering the entry or active efflux of the drug, the destruction or inactivation of the drug, and development of resistant metabolic pathways (Yismaw G 2010).

They are usually considered to be opportunistic pathogens, which constitute a large portion of the normal intestinal flora of humans. The bacteria can, contaminate, colonize, and subsequently cause infection of extra intestinal sites, and are a major cause of septicemia, peritonitis, abscesses, meningitis, and urinary tract infections (UTI) in humans. *E. coli* is an incredibly diverse bacterial species with the ability to colonize and persist in numerous niches both in the environment and within animal hosts (Okonko IO *et al* 2009). The organisms are known to cause enteric infections and diarrhea (gastroenteritis) in humans and animals.

The surfacing of antibiotic resistance usually results from the misuse of antibiotics as growthpromoters in animal production, for therapy and prophylaxis (Chees bourgh M 1991) Because humans consume these animal products, there is a probability of the spread of resistant strains from animals to humans and thus healthy individuals can become carrier hosts for multiple antibiotic-resistant bacteria (Bauer A.W.1996 et al ).Several studies have revealed that *E. coli* is resistant to a number of antibiotics. (von Baum H, Reinhard M. 2000, Gangoué JP *et al* 2004, Orrett FA, Shurl SM. 2001, Iqbal MK, Patel IK 2002 and Al-Tawfiq JA. 2006).Adding to the consequences of microbial resistance to antibiotics on human health, contamination of surface water bodies with resistant bacterial strains from human activities and livestock operations has also been reported (Iqbal MK, Patel IK 2002).

## MATERIALS AND METHODS Sample Collection

A total of 384 samples were collected from different sources in Ismailia governorate. Sampling included various types of food, water, stool from diarrheal human and animal cases, and urine from patients with urinary tract infection (UTI). A total of 99 food samples were randomly collected in sterile plastic bags from different local markets. The collected foods included meat products, poultry products, sea-food products and dairy products. Twenty five grams of each sample was treated with 225 ml of modified tryptic soy broth (mTSB- DifcoLa Jolla, CA/USA) and incubated with agitation (120 rpm) for 24 h at 37°C as described by Cowan (1985) and Ethelberg (2009). After enrichment for 24 h at 37°C, aliquots of 100 µl were plated onto Eosin Methylene Blue (EMB-Difco) agar to presumptively identify isolates as Gram-negative enteric bacteria and presumptive E. coli (green-metallic colonies), and onto MacConkey agar (MAC-Difco) .After 18 to 24 h at 37°C, characteristic colonies from EMB agar and MAC agar were transferred onto Tryptic Soy agar (TSA, Difco) and used for biochemical identification tests. Thirty one water samples were collected from the Ismailia freshwater canal and examined according to standard methods (APHA, 1998) isolates yielding typical green metallic colonies were inoculated into MAC agar and incubated at 37°C for 24 h (APHA, 1998). Fecal specimens (n=179) were collected following standard methods.

# ANTIBIOTIC SUSCEPTIBILITY TESTS

The whole number of *E. coli* isolates (n = 283) were tested for their susceptibility to 16 antibiotics listed in Table 1. Testing was performed on Mueller-Hinton Agar plates using the Kirby-Bauer disk diffusion technique (Jorgensen *et al.*, 1997). The antibiotic resistance of each *E. coli* isolate was determined based on the breakpoints of the inhibition zone diameters for individual antibiotic agents and as recommended by the disk manufacturer. The results

were interpreted according to the guidelines of the Clinical Laboratory Standards Institute for antimicrobial susceptibility testing (CLSI, 2010). Resistance to more than four antibiotics was as multidrug resistance (MDR). MDR index (MDRI) of individual isolates was calculated by dividing the number of antibiotics to which the isolate was resistant by the total number of antibiotics to which the isolate was exposed (Chandran *et al* 2008). Isolates with MDRI values of more than 0.2 or 20% were considered highly resistant.

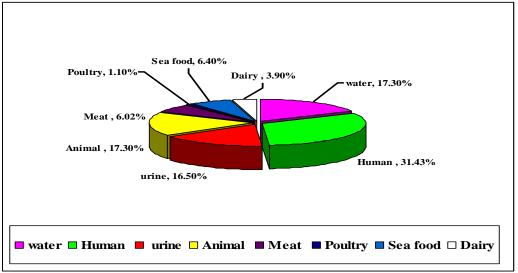
# Table (1): The evaluation of inhibition zone diameter (mm) of antibiotic susceptibility test according to National Committee for Clinical Laboratory Standards (NCCLS) (2003)

			<b>References measurements for</b> <i>Enterobactericeae</i> in mm						
Antibiotics list	Conc.µg	Symbol.	Resistance	Intermediate	Sensitive				
Amoxicillin/Calvulanic Acid	30	AMC 30	≤13	14-17	≥ 18				
Ampicillin	10	AM 10	≤13	14-16	≥ 17				
Cefazolin	30	CZ 30	≤14	15-17	≥ 18				
Ciprofloxacin	5	CIP	≤15	16-20	≥ 21				
Clindamycin	2	DA	≤14	15-20	≥ 21				
Erythromycin	15	E 15	≤13	14-22	≥ 23				
Gentamicin	120	CN 120	≤6	79	≥ 10				
Imipenem	10	<b>IPM 10</b>	≤13	14-15	≥ 16				
Nitrofurantion	300	F 300	***	***	***				
Nitrofloxacin	10	<b>NOR 10</b>	***	***	***				
Penicillin	10	P 10	****	****	****				
Rifampin	5	RA 5	≤16	17-18	≥ <b>20</b>				
Spectinomycin	100	SPT 100	≤14	1214	≥ 15				
Tetracycline	30	TE 30	****	****	****				
Thiampinicol	30	TP 30	****	****	****				
Vancomycin	30	VA 30	****	****	****				

(\*\*\* means that this antibiotic mainly inhibit Enterobactericeae)

# **RESULTS Prevalence of** *E. coli* recorded in all samples:

From figure (1): The number of *E. coli* which isolated from all studied samples were estimated as (283) isolates. The highest percentage of E. coli prevalence was recorded as (31.43%) in human stool followed by 17.33%, 17.3% and 16.52% in animal stool, raw water and human urine respectively. The lowest percentages of E. coli prevalence was recorded as 1.1% in poultry products and raised to 3.9%, 6.02%, 6.4% in diary products, meat products and seafood respectively.



Figure(1): Percentages of Prevalence of *E. coli* in different represented sources

# Antimicrobial susceptibility results (Antibiogram) of E. coli isolates:

Antibiotics disc diffusion technique indicated ideal results among the isolated strains. More detailed prevalence of resistance, intermediate and sensitive E. coli strains (Antibiogram for 283) were tabulated in table : (2,3,4). The resistant isolates of *E. coli* against Amoxicillin/Calvulanic Acid (AMC) were (141) isolates, the distribution and prevalence of E coli isolates were as following: Four from raw water, 52 from human stool, 36 from animal stool 27 from UTI, and 11 ,1 ,2,8 isolates from meat products poultry, diary and seafood respectively. The intermediate pattern to (AMC 30) was represented in (59) isolates and grouped as: 14 raw water, 13 human stool, 9 animal stool 13 UTI, 3 meat products , 1 poultry, 1 diary , and 5 seafood While the sensitive patterns of E. coli against to (AMC 30) were observed in (83) isolates and were grouped as following: 31 raw water, 24 human stool, 4 animal stool 3 UTI, 3 meat products , 1 poultry, 8 diary , and 5 seafood.

The resistant isolates of *E. coli* against Ampicillin (AM 10) were (211) isolates their distribution and prevalence were as following: 36 from raw water, 55 from human stool, 43 from animal stool 36 UTI, 15 from meat products, 3 from poultry, 8 from diary, and 15 from seafood. The intermediate pattern to (AM 10) was found in (30) isolates and were grouped as following: 9 raw water, 6 human stool, 2 animal stool 7 UTI, 1 meat products, 0 poultry, 1diary, and 4 seafood. While the sensitive patterns to (AM 10) were recorded in (42) isolates and were grouped as: Tow from raw water, 28 from human stool, 1 from animal stool 4 from UTI, 2 from meat products, 1, 2, 2 in poultry, diary, and seafood respectively.

## European Journal of Advanced Research in Biological and Life Sciences

#### Vol. 3 No. 1, 2015 ISSN 2056-5984

## Table (2): Resistance pattern of *E. coli* isolates originated from different sources.

		Raw	Human	Animal					Sea		
		water	stool	stool	UTI	Meat	Poultry	Diary	food		
								11			
		49	89	49	47	17	3	(3.9)	18		resistant
Antibiotic agent	Symbol/Conc.	(17.3%)	(31%)	(17.3%)	(16.5%)	(6.02%)	(1.1%)	%	(6.4%)	isc	olates
Amoxicillin/Calvulanic											
Acid	AMC 30	4	52	36	27	11	1	2	8	141	50%
Ampicillin	AM 10	36	55	43	36	15	3	8	15	211	75%
Cefazolin	CZ 30	21	49	33	32	9	0	0	12	156	55.10%
Ciprofloxacin	CIP	13	53	45	30	13	1	0	1	156	55.1
Clindamycin	DA	25	51	45	24	9	1	0	6	161	57%
Erythromycin	E 15	27	43	44	30	13	0	0	12	169	60%
Gentamicin	CN 120	24	34	29	20	9	0	0	3	119	42%
Imipenem	IPM 10	14	23	31	7	6	0	0	1	82	29%
Nitrofurantion	F 300	27	23	16	23	4	2	3	3	101	36
Nitrofloxacin	NOR 10	23	45	33	9	4	2	0	2	118	42%
Penicillin	P 10	23	51	49	33	13	0	1	10	180	64%
Rifampin	RA 5	22	51	46	27	10	1	0	10	167	59%
Spectinomycin	SPT 100	31	25	31	16	6	0	0	4	113	40%
Tetracycline	TE 30	32	35	42	22	4	0	0	6	141	50%
Thiampinicol	TP 30	31	40	38	28	9	0	0	7	153	54%
Vancomycin	VA 30	35	46	49	35	10	1	1	18	195	69%

#### Vol. 3 No. 1, 2015 ISSN 2056-5984

#### Table (3): Intermediate pattern of *E. coli* isolates originated from different sources.

Antibiotic agent		Raw water 49	Human stool 89	Animal stool 49	UTI 47	Meat 17	Poultry 3	Diary 11 (3.9)	Sea food 18	No of intermediate isolates 59	
A mariaillin (Calendaria	Symbol/Conc.	(17.3%)	(31%)	(17.3%)	(16.5%)	(6.02%)	(1.1%)	%	(6.4%)		
Amoxicillin/Calvulanic Acid	AMC 30	14	13	9	13	3	1	1	5	59	21%
Ampicillin	AM 10	9	6	2	7	1	0	1	4	30	11%
Cefazolin	CZ 30	20	20	12	9	2	0	1	4	68	24.00%
Ciprofloxacin	CIP	16	33	1	12	3	0	2	2	69	25.00%
Clindamycin	DA	19	12	1	11	2	0	2	4	51	18%
Erythromycin	E 15	12	9	0	6	2	2	3	6	40	14%
Gentamicin	CN 120	19	10	15	6	2	0	3	6	61	22%
Imipenem	IPM 10	19	13	9	6	1	1	3	3	55	19%
Nitrofurantion	F 300	9	13	17	4	2	1	2	3	51	18%
Nitrofloxacin	NOR 10	20	12	12	6	3	1	3	6	63	22%
Penicillin	P 10	15	8	0	10	2	2	2	2	41	14%
Rifampin	RA 5	10	29	1	20	4	1	3	7	75	27%
Spectinomycin	SPT 100	7	11	6	5	5	2	2	5	43	15%
Tetracycline	TE 30	4	10	3	9	1	1	2	2	32	11%
Thiampinicol	TP 30	18	19	9	18	3	1	2	3	73	25%
Vancomycin	VA 30	12	16	0	9	2	1	2	0	42	15%

#### Vol. 3 No. 1, 2015 *ISSN 2056-5984*

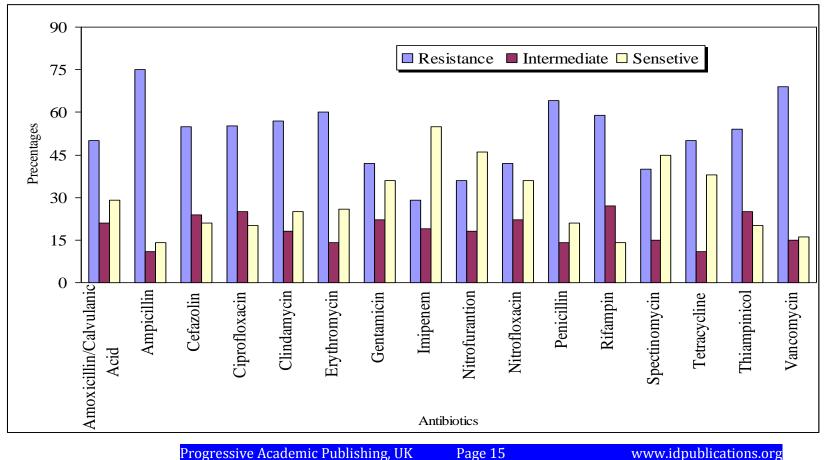
## Table (4):Sensitive pattern of *E. coli* isolates originated from different sources

Antibiotic agent	Symbol/Conc.	Raw water 49 (17.3%)	Human stool 89 (31%)	Animal stool 49 (17.3%)	UTI 47 (16.5%)	Meat 17 (6.02%)	Poultry 3 (1.1%)	Diary 11 (3.9) %	Sea food 18 (6.4%)		sensitive solutes
Amoxicillin/Calvulanic Acid	AMC 30	31	24	4	7	3	1	8	5	83	29%
Ampicillin	AM 10	2	28	1	4	2	1	2	2	42	14%
Cefazolin	CZ 30	8	20	4	6	6	3	10	2	59	21.00%
Ciprofloxacin	CIP	20	3	4	5	1	2	9	14	58	20.00%
Clindamycin	DA	5	26	3	12	6	2	9	8	71	25%
Erythromycin	E 15	10	37	3	11	2	3	8	0	74	26%
Gentamicin	CN 120	6	45	5	21	6	3	8	9	103	36%
Imipenem	IPM 10	16	53	9	34	10	2	8	14	146	55%
Nitrofurantion	F 300	13	53	16	20	11	0	6	12	131	46%
Nitrofloxacin	NOR 10	6	32	4	32	10	0	8	10	102	36%
Penicillin	P 10	11	30	0	4	2	1	8	6	62	21%
Rifampin	RA 5	17	9	2	0	3	1	8	1	41	14%
Spectinomycin	SPT 100	11	53	12	26	6	1	9	9	127	45%
Tetracycline	TE 30	13	44	2	16	12	2	10	11	110	38%
Thiampinicol	TP 30	0	30	2	1	5	2	9	8	57	20%
Vancomycin	VA 30	2	27	0	3	5	1	8	0	46	16%

#### European Journal of Advanced Research in Biological and Life Sciences

Figure (2): Antibiotic Resistance, Intermediate and Sensitive percentages patterns of *E coli* isolates against 16 type of antibiotics

Vol. 3 No. 1, 2015 ISSN 2056-5984



The resistant isolates of *E. coli* against Cefazolin (CZ 30) were recorded in (156) isolates, their prevalence and distributed were as following: 21 from raw water, 49 human stool, 33 animal stool 32 UTI, 9 meat products, 0 poultry, 0 diary, and 12 seafood. Intermediate pattern was recorded in (68) isolates and grouped as : 20 from raw water, 20 from human stool, 12, 9, 2, 0, 1, 4 from animal stool UTI, meat products, poultry, diary, and seafood respectively. Sensitive patterns to (CZ 30) were observed in (59) isolates and were grouped as 8 from raw water, 20 from human stool, 4 from animal stool 6 UTI, 6 meat products, 3 poultry, 10 diary, and 2 seafood.

The resistant isolates of *E. coli* against Ciprofloxacine (CIP5) was observed in (156) isolates their prevalence and distribution were as following : 13 from raw water, 53 human stool, 45 animal stool 30 UTI, 13 meat products , 1 poultry, 0 diary , and 1 seafood. Intermediate pattern was recorded in (69) isolates and were grouped as : 16 from raw water, 33 from human stool, 1 animal stool 12 UTI, 3 meat products , 0 poultry, 2 diary , and 2 seafood. While (58) isolates were sensitive to (CZ 30) and grouped as : 20 from raw water, 3 from human stool, 4 animal stool 5 UTI, 1 meat products , 2 poultry, 9 diary , and 14 seafood.

The resistant isolates of *E. coli* against Clindamycin (DA) was (161) isolates and their distributions were as following: 25 raw water, 51 human stool, 45 animal stool 24 UTI, 9 meat products, 1 poultry, 0 diary, and 6 seafood. intermediate pattern was represented in (51) isolates and was grouped as: 19 from raw water, 12 from human stool, 1 animal stool 11 UTI, 2 meat products, 0 poultry, 2 diary, and 4 seafood. While (71) isolates were sensitive to (DA) and were grouped as: 5 from raw water, 26 from human stool, 3 from animal stool 12, 6, 2, 9, 8 isolated from UTI, meat products, poultry, diary, and seafood respectively.

The resistant pattern of *E. coli* against Erythromycin (E 15) were observed in (169) isolates and were distributed as : 27 from raw water, 43 human stool, 44 animal stool 30 UTI, 13 meat products, 0 poultry, 0 diary, and 12 seafood. Intermediate pattern was recorded in (40) isolates and were grouped as: 12 from raw water, 19 human stool, 0 animal stool 6 UTI, 2 meat products, 2 poultry, 3 diary, and 6 seafood. While (74) isolates were sensitive to (E 15) and were grouped as : 10 raw water, 37 human stool, 3 animal stool 11 UTI, 2, 3, 8,0 from meat products , poultry, diary , and seafood respectively.

The resistant pattern of *E. coli* against Gentamicin (CN 120) was observed in (119) isolates and their prevalence and distribution were as following: 24 isolated from raw water, 34 from human stool, 29 animal stool 20 UTI, 9 meat products, 0 poultry, 0 diary, and 3 seafood. The intermediate patterns were recorded in (61) isolates and were grouped as 19 from raw water, 10 from human stool, 15 from animal stool 6 UTI, 2 meat products, 0 poultry, 3 diary, and 6 seafood. While (103) isolates were sensitive to (CN 120) and were recorded as 6 from raw water, 45 human stool, 5 animal stool 21, 6, 3, 8, 9 isolated from UTI, meat products, poultry, diary, and seafood respectively.

The resistant isolates of *E. coli* against Imipenem (IPM 10) were (82) isolates and their distribution and prevalence were as following: 14 from raw water, 23 from human stool, 31 animal stool 7 UTI, 6 meat products, 0 poultry, 0 diary, and 1 seafood. While (55) isolates shown intermediate pattern and grouped as : 19 from raw water, 13 from human stool, 9 animal stool 6 UTI, 1 meat products, 1 poultry, 3 diary, and 3 seafood. Sensitive profiles to

(IPM 10) were observed in (146) isolates and grouped as : 16 from raw water, 53 human stool, 9 animal stool 34 UTI, 10 meat products, 2 poultry, 8 diary, and 14 seafood.

The resistant isolates of E. coli against Nitrofurantion (F 300) were (101) isolates their distribution and prevalence were as following: 27 from raw water, 23 from human stool, 16 from animal stool 23 UTI, 4 meat products, 2 poultry, 3 diary, and 3 seafood. While (51) isolates shown intermediate pattern and were grouped as : 9 isolated from raw water, 13 from human stool, 17 animal stool 4 UTI, 2 meat products, 1 poultry, 2 diary, and 3 seafood. Sensitive patterns to (F 300) were observed in (131) isolates and were represented as: 13 from raw water, 53 from human stool, 16 from animal stool 20 UTI, 11 meat products, 0 poultry, 6 diary, and 12 seafood.

The resistant pattern of E. coli against Nitrofloxacin (NOR 10) (118) isolates and were distributed as following: 23 from raw water, 45 from human stool, 33 from animal stool 9 from UTI, 4 meat products, 2 poultry, 0 diary, and 2 seafood. Intermediate patterns to (NOR 10) were recorded in (63) isolates and were grouped as : 20 from raw water, 12 from human stool, 12 from animal stool 6 UTI, 3 meat products, 1 poultry, 3 diary, and 6 seafood. While (102) isolates were sensitive to (NOR 10) and were grouped as: 6 from raw water, 32 from human stool, 4 animal stool 32 from UTI, 10 from meat products, 0 poultry, 8 diary, and 10 seafood.

The resistant pattern of E. coli against Penicillin (P10) were observed in (180) E coli isolates and were distributed as following: 23 isolated from raw water, 51 from human stool, 49 animal stool 33 UTI, 13 meat products, 0 poultry, 1 diary, and 10 seafood. Intermediate pattern to (P10) were observed in (41) isolates and were grouped as : 15 from raw water, 8 from human stool, 0 from animal stool 10 UTI, 2meat products, 2 poultry, 2 diary, and 2 seafood. While (62) isolates were sensitive to (P10) and were grouped as : 11 from raw water, 30 from human stool, 0 animal stool 4 UTI, and 2, 1, 8, 6 meat products, poultry, diary, and seafood respectively.

The resistant pattern of E. coli against Rifampin (RA5) were recorded in (167) isolates and their prevalence were as following: 22 from raw water, 51 from human stool, 46 animal stool 27 UTI, 10 meat products, 1 poultry, 0 diary, and 10 seafood. While (75) isolates shown intermediate pattern and were grouped as :10 from raw water, 29 from human stool, 1 animal stool 20 UTI, 4 meat products, 1 poultry, 3 diary, and 7 seafood. The sensitive profiles to (RA 5) were observed in (41) isolates and were recorded as: 17 from raw water, 9 from human stool, 2 animal stool 0 UTI, 3 meat products, 1 poultry, 8 diary, and 1 seafood.

The resistant pattern of E. coli against Spectinomycin (SPT 100) were estimated in (113) E coli isolates and were distributed as : 31 from raw water, 25 from human stool, 31 from animal stool 16 UTI, 6 meat products, 0 poultry, 0 diary, and 4 seafood. While (43) isolates shown intermediate pattern and were grouped as : 7 from raw water, 11 from human stool, 6 animal stool 5 UTI, 5 from meat products, 2 poultry, 2diary, and 5 seafood. The sensitive patterns to (SPT 100) were represented in (127) isolates and were recorded as : 11 from raw water, 53 from human stool, 12 from animal stool and 26, 6, 1, 9, 9 UTI, meat products, poultry, diary, and seafood respectively.

The resistant patterns of *E. coli* against Tetracycline (TE 30) were observed in (141) isolates and their prevalence and distribution was as following: 32 from raw water, 35 from human stool, 42 animal stool 22 UTI, 4 meat products, 0 poultry, 0 diary, and 6 seafood. While (32) isolates shown intermediate patterns and were grouped as: 4 from raw water, 10 from human stool, 3 from animal stool 9 UTI, 1 meat products, 1 poultry, 2diary, and 2 seafood. Sensitive profiles to (TE 30) were recorded in (110) isolates and were grouped as: 13 from raw water, 44 from human stool, 2 animal stool 16 UTI, 12 meat products, 2 poultry, 10 diary, and 11 seafood.

The resistant patterns of *E. coli* against Thiampinicol (TP 30) were rescored in (153) isolates and were grouped as following: 31 from raw water, 40 from human stool, 38 animal stool 28 UTI, 9 meat products, 0 poultry, 0 diary, and 7 seafood. While (73) isolates shown intermediate pattern and were distributed as: 18 raw water, 19 human stool, 9 animal stool 18 UTI, 3 meat products, 1 poultry, 2diary, and 3 seafood. The sensitive patterns to (TP 30) were recorded in (57) isolates and were grouped as: No from raw water, 30 human stool, 2 animal stool 1 UTI, 5 meat products, 2 poultry, 9 diary, and 8 seafood.

The resistant patterns of E. coli against Vancomycin (VA 30) were found in (195) isolates and were distributed as following: 35 from raw water, 46 from human stool, 49 animal stool 35 UTI, 10 meat products, 1 poultry, 1diary, and 18 seafood. While (42) isolates shown intermediate pattern and were grouped as : 12 from raw water, 16 from human stool, 0 animal stool 9 UTI, 2 meat products, 1 poultry, 2 diary, and 0 seafood. The sensitive profiles to (VA 30) were recorded in (46) and were grouped as: 2 from raw water, 27 from human stool, 0, 3, 5, 1, 8, 0 animal stool UTI, meat products, poultry, diary, and seafood respectively.

# DISCUSSION

Antimicrobial resistance in E. coli has increased worldwide (Von Baum H, Reinhard M. 2000). To help characterize evolution of drug resistance in E. coli since antimicrobial drugs were widely used, 283 E. coli isolates were tested for their susceptibility to a common panel of 16 antimicrobial agents. We tested E. coli isolates from both clinical and environmental sources for susceptibility trends. In this study, the overall resistance of E. coli to antimicrobials was high. The result is consistent with the findings of previous studies (Orrett FA, Shurl SM. 2001).

The Antibiotiogram profile for the 283 E. coli isolates indicated that Antibiotic resistance pattern is greatly increasing among most of E. coli isolates Figure (2): 75% of isolates were resistant to Ampicillin followed by Vancomycin 69%, Penicillin 64%, Erthromycin 60%, while 55% demonstrated resistance to Ciprofloxacin and 50% resistance to tetracycline. In addition, we identified 50% resistance of the tested isolates to Amoxicillin-Clavulanic acid (AMC) which were mainly noticed among isolates from from human stool (n=52), animal stool (n=36),UTI (n=27), from meat products (n=11), seafood (n=8), (n=4) water (n=4)(n=2) from dairy product (n=2), and from poultry (n=1). We found that near to 93% of the Isolated E.coli were muliti drug resistant (MDR) as they were resistant to 4-13 types of the investigated antibiotics, this pattern were more pronounced among clinical samples and meat product as (100%) of these isolates were resistant to Amoxicillin/Calvulanic Acid, Ampicillin, Erythromycin, Penicillin, Rifampin, Tetracycline and Vancomycin.

The findings alarm to a serious impact in limiting the selection of treatment drug. This finding corroborated with the study reported by Mubita et al., 2008, who reported that both clinical and environmental strains displayed MDR phenotype to most of the previously mentioned antibiotics. Many authors documented that the use of antibiotics is strongly associated with the prevalence of antimicrobial resistance in E. coli isolates in foodproducing animals (kang et al., 2005).

Similar finding has been reported in other *E. coli* pathovar in many other studies from Egypt (Shaheen et al., 2004; Putnam et al., 2004) and different parts of the world (Okeke et al., 2000; Hoge et al., 1998; Shapiro et al., 2001; Turner et al., 1998). There is an increasing isolation rate of MDR strains belonged to Enteropathogenic E. coli in Nigeria (Okeke et al., 2000), Thailand (Hoge et al., 1998), Kenya (Shapiro et al., 2001) and Israel (Turner et al., 1998). Despite the MDR pattern reported in other studies involving traveler's diarrhea, yet susceptibility to ciprofloxacin was noted among the E.coli strains which makes it remain as the drug of choice for the treatment of (Ericsson et al., 2003; Dupont, 2006).

## REFERENCES

- Al-Tawfiq JA. Increasing antibiotic resistance among isolates of *Escherichia coli* recovered from inpatients and outpatients in a Saudi Arabian Hospital. Infect Control Hosp Epidemiol 2006; 27: 748-753.
- APHA. (1998). Standard methods for the examination of water and waste, water 19th ed. American Public Health Association Washington D.C.
- Bauer, A.W., Kirby, W.M.M., Sherris, J.C. and Turck, M. Antibiotic susceptibility testing by standard single disc method. Am J. Clin Pathol 1996; 45: 493-496.
- Centers for Disease Control and Prevention. (2003). Outbreak of Escherichia coli O157:H7 infections associated with drinking un pasteurized commercial apple juice. Morbid. Mortal. Weekly Rep. 45:44.
- Chandran A, Hatha AAM, Varghese S, Sheeja KM (2008). Prevalence of multidrug resistant E. coli serotypes in a Tropical Estuary, India. Microbes EnvIRON. 23(2): 153-158.
- Cheesbourgh M. Medical laboratory manual for tropical countries. 2nd edition: England: Butter worth-Heineman LTD, 1991; 114-6.
- Clinical and Laboratory Standards Institute CLSI, (2003). Performance standards for antimicrobial susceptibility testing document M100-S14., Wayne, PA.
- Dupont, H.L., (2006). Travellers' diarrhoea: Contemporary approaches to therapy and prevention. Drugs, 66: 303-314.
- Dromigny JA, Nabeth P, Juergens-Behr A, Perrier-Gros-Claude JD. Risk factors for antibiotic resistant Escherichia coli isolated from community-acquired urinary tract infections inDakar, Senegal. J Antimicrobial Chemother. 2005; 56: 236-239.
- Erb A, Stürmer T, Marre, R Brenner H. Prevalence of antibiotic resistance in *Escherichia coli*: overview of geographical, temporal, and methodological variations. Eur J Clin Microbial Infect Dis. 2007; 26: 83-90.
- Ericsson, C.D., (2003). Travellers' diarrhoea. Int. J. Antimicrob. Agents, 21:116.-124. Gangoué JP, Koulla-Shirob S, Ngassama P, Adiogo D, Njine T, Ndumbe P. Antimicrobial resistance of Gram-negative bacilli isolates from inpatients and outpatients at Yaounde CentralHospital, Cameroon. Inter J Infect Dis. 2004; 8: 147-154.
- Hoge, C.W., J.M. Gambel, A. Srijan, C. Pitarangsi and P. Echeverria, (1998). Trends in antibiotic resistance among diarrheal pathogens isolated in Thailand over 15 years. Clin. Infect. Dis., 26: 341-345.
- Iqbal MK, Patel IK. Susceptibility patterns of Escherichia coli: Prevalence of multidrugresistant isolates and extended spectrum beta-Lactamase phenotype. J Pak Med Asso. 2002; 52: 407-417.

- Jorgensen, J.H., J.D. Turnide, and J.A. Washington. (1999). Antibacterial Susceptibility Tests: Dilution and DiskDiffusion Methods. In: Manual of Clinical Micro-biology, 7th ed. Murry, P.R., M.A. Pfaller, F.C. Tenover, E.J. Baron, and R.H. Yolken (eds.), ASM Press, Wa-shington, D.C. pp. 1526-1543.
- Kang, H.Y.; Jeong Y.S.; Oh, JY.; Tae, S.H.; Choi, C.H.; Moon D.C.; Lee, W.K.; Y.C.; Soel, S.Y. and Cho, D.T. (2005).Characterization of antimicrobial resistance and class I integrons found in E.coli isolates from human and animals in Korea J. Antimicro. Chemother.55:639-644.
- Mubita, C., M. Syakalima, C. Chisenga, M. Munyeme and M. Bwalya et al., (2008). Antibiograms of faecal Escherichia coli and Enterococci species isolated from pastoralist cattle in the interface areas of the Kafue basin in Zambia-short communication. Veterinarski Arhiv, 78: 179-185.
- Okonko IO. Soleye FA, Amusan TA, Ogun AA, Ogunnusi TA Ejembi J. Incidence of multi-drug resistance (MDR) organisms in Abeokuta, Southwestern Nigeria. *Global J Pharm.* 2009; 3(2): 69-80.
- Putnam, S.D., M.S. Riddle, T.F. Wierzba, B.T. Pittner and R.A. Elyazeed et al., (2004). Antimicrobial susceptibility trends among Escherichia coli and Shigella spp. isolated from rural Egyptian paediatric populations with diarrhoea between 1995 and 2000. Clin. Microbiol. Infect, 10: 804-810.
- Okeke, I.N., A. Lamikanra, H. Steinrck and J.B. Kaper, (2000). Characterization of Escherichia coli strains from cases of childhood diarrhea in provincial Southwest Nigeria. J. Clin. Microbiol., 38: 7-12.
- Orrett FA, Shurl SM. Prevalence of resistance to antimicrobial of *E. coli* isolates from clinical sources at a private hospital in Trinidad. *Jpn J Infect Dis.* 2001; 54: 64-68.
- Shaheen, H.I., S.B. Khalil, M.R. Rao, R. Abu Elyazeed and T.F. Wierzba et al., (2004). Phenotypic profiles of enterotoxigenic Escherichia coli associated with early childhood diarrhea in rural Egypt. J. Clin. Microbiol., 42: 5588-5595.
- Shapiro, R.L., L. Kumar, P. Phillips-Howard, J.G. Wells and P. Adcock et al., (2001). Antimicrobial- resistant bacterial diarrhea in rural Western Kenya. J. Infect. Dis., 183: 1701-1704.
- Turner, D., N. Porat, D. Cohen, M. Yavzori, D. Fraser, N. Peled, O. Ohama and R. Dagan, (1988). Antibiotic resistance pattern of enterotoxigenic E. coli isolated from infants and young adults in Israel. Eur. J. Clin. Microbiol. Infect. Dis., 17: 666-669.
- Von Baum H, Reinhard M. Antimicrobial resistance of *Escherichia coli* and therapeutic implications. *Inter J Med Microbiol*. 2000; 295: 503–511.
- Yismaw G, Abay S, Asrat D, Yifru S, Kassu A Bacteriological profile and resistant patterns of clinical isolates from pediatric patients, Gondar University Teaching Hospital, Gondar Northwest Ethiopia. *Ethiop. Med. J.* 2010; 48(4): 293-300.