Prevalence and Antibiotic Susceptibility of *Escherichia Coli* Isolated from Meat-Based Products in Port-Said Markets

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Abstract
Eight hundred of meat products were collected randomly from Port-Said markets for *E. coli* isolation that was (43%). *E. coli* was recorded in (19.77%) minced meat, (17.44%) in raw meat, (17.15%) in sausage, (16.57%) in burger, (15.70%) in pastirma, (6.10%) in luncheon, (4.07%) in salami, and (3.20%) in frankfurter. Serologically, *E. coli* isolates were categorized into the following serotypes: (O25, O26, O55, O63, O78, O86, O111, O112, O114, O119, O124, O125, O126, O136, O142, O157, O158), and un-typed serotypes. Antibiotic sensitivity test revealed that all isolates were sensitive to Imipenem, Chloramphenicol, Nalidixic acid, and Ofloxacin while they were resistant to Erythromycin, Penicillin, Bacteracin, Cephradin, Rifampin, Tetracycline, Ciprofloxacin, and Vancomycin.

Introduction

Escherichia coli is Gram-negative rod-shaped bacterium ***Gansheroff and O'Brien (2000)*** that its presence in beef carcasses during processing considered as an indicator of fecal contamination ***Rantsiou et al. (2012).*** There are over 170 known serogroups of *E. coli*, classified according to somatic (O), flagellar (H), and/or capsular (K) antigens ***Wong et al. (2000).***

E. coli antibiotic resistance is of a particular concern because it is the most common Gram-negative pathogen in humans, the most common cause of urinary tract infections, a common cause of both community and hospital-acquired bacteremia ***Salvadori et al. (2004)*** and a cause of diarrhea ***Kaper et al. (2004).*** Because of resistance to most first-line of antibiotics, treatment of *E. coli* infection became complicated ***Sabaté et al. (2008).***
Therefore, this study aimed to study prevalence, serotyping, and antibiotic sensitivity of *E. coli* isolated from meat and meat-based products.

Material and Methods

Sample collection: Eight hundred samples consisted of 100 raw meat samples and 700 raw meat-based products samples included (minced meat, sausage, burger, pastirma, luncheon, salami, frankfurter) were collected from Port Said governorate markets as 100 from each during period from September 2016 to September 2018.

Sample preparation: Twenty-five grams of each product represented the product sample added aseptically to 225 ml buffered peptone water then enriched by incubation at 37ºC for 24 hours ***ICMSF (1978)***.

Bacterial isolation: Enriched samples were streaked on Eosin Methylene Blue agar (EMB) and MacConkey's agar and incubated at 37°C for 24 hours while on Tryptone Bile Glucuronic Agar (TBX) were incubated first at 37°C for 4 hours then at 44°C for 20 hours ***Koneman et al. (1997)***.

Serological examination: Isolates were submitted to serological typing by slide agglutination test using O somatic antigens ***Edwards and Ewing (1972)***.

Antibiotic sensitivity test: Antibiotic sensitivity test was done by disc diffusion test ***Bauer et al., (1966)*** and isolates were classified as sensitive, intermediate, or resistant according to ***NCCLS/CLSI (2007)***.

Results and Discussion

Meat and meat products are highly susceptible to microbial contamination as they are rich in essential nutrients ***Dave and Ghaly (2011).*** *E. coli* colonies exhibited unique metallic green sheen on EMB agar plate **Figure (1)**, blue green on TBX agar **Figure (2)**, and lactose-fermenting pink colonies on MacConkey's agar **Figure (3)** that go parallel with ***Omoruyi et al. (2018), Verhaegen, et al. (2015),*** and ***Abd El Tawab et al. (2015)*** respectively. The typical biochemical responses of *E. coli* listed in **Table (1)** were in line with ***Tafida et al. (2014).***

The prevalence of *E. coli* in examined raw meat and meat based products samples was 344/800 (43%) which is nearly similar to ***El-Sharkaway et al. (2016)*** who reported *E. coli* as (41%). High rate of *E. coli* can be attributed to high initial contamination in slaughterhouses, fecal contamination of carcass during dressing, contamination during products preparation by using raw materials with poor quality, contamination via workers, contaminated working surfaces, contaminated instruments, inappropriate transportation and refrigeration conditions. Results listed in **Table (2)** illustrated *E. coli* prevalence in each meat product. The present results is lower than ***El-Sharkaway et al. (2016)*** as they recorded *E. coli* highest ratio in burger as (29.26%) followed by minced meat as (26.82%), sausage as (24.39%), and pastirma as (19.51%).

The most predominant serotype was O125 as (25%) followed by O158 as (20.93%), un-typed serotypes as (12.5%), O111 as (10.47%), O55 as (8.43%), O157 as (5.81%), O26 as (4.07%), O119 as (2.33%), O142 as (2.03%), O114 as (1.74%), both O124 and O136 as (1.45%), O78 as (1.16%), O112 as (0.87%), both O63 and O126 as (0.58%), and both O25 and O86 as (0.29%). ***Ibrahim et al. (2015)*** isolated similar *E. coli* serotypes from meat products as O26, O55, O111, O114, O119, O124, and O125.

Antibiogram of *E. coli* listed in **Table (3)** and **Figure (4)** revealed that (95.93%) of isolates were highly sensitive to Imipenem which is in agreement with ***Gundogan and Avci (2013).*** About (94%) of isolates were highly sensitive to Chloramphenicol which goes parallel with ***Oja et al. (2010).*** Also, (74.13%) of isolates were sensitive to Nalidixic acid which is nearly similar to ***Nobili et al. (2017)*** who confirmed that all tested *E. coli* isolates were sensitive to Nalidixic acid. Moreover, none of *E. coli* isolates were resistant to Ofloxacin as (77.03%) were highly sensitive while (22.97%) were moderately sensitive to it.

In addition, (85.76%) of isolates were moderately sensitive to Neomycin while (2.91%) were sensitive to it. ***Rahman et al. (2017)*** recorded higher sensitivity rate, as all *E. coli* isolates were sensitive to Neomycin. ***Arya et al. (2008)*** reported that only (20%) of *E. coli* isolates were sensitive to Amikacin which is lower than the current result as (40.12%) were sensitive to Amikacin.

Furthermore, ***Srinivasa et al. (2011)*** illustrated that (10.7%) of *E. coli* isolates were resistant to Polymyxin B which is nearly similar to the present study as (8.72%) of *E. coli* isolates were resistant to Polymyxin B. ***Rahman et al. (2017)*** detected that (28.57%) of *E. coli* isolates were resistant to Doxycyclin which is higher than the present study as (17.15%) of isolates were resistant to Doxycyclin.

In addition, (51.45%) of isolates were intermediately sensitive to Amoxicillin/Clavulanic acid and (31.40%) of isolates were resistant to it. Higher resistance rate recorded by ***Ammar et al. (2016)*** as (93.75%) of isolates were resistant. About (40%) *E. coli* isolates were resistant to Trimethoprim/Sulfamethoxazole but lower resistance rate as (28.57%) recorded by ***Rahman et al. (2017)***.

Present result demonstrated resistance of isolated *E. coli* serotypes to Erythromycin that agrees with ***Al-Sultan et al. (2012).*** This may be resulted from long-term and widespread abuse of Erythromycin in livestock fields. All *E. coli* isolates were resistant to Bacitracin, Vancomycin ***Nicoline et al. (2015)*** and Tetracycline ***Anu and Gayathri (2015)*** which agree with present result. Ciprofloxacin resistance was (100%) which is higher than **Yang *et al*. (2004)** who recorded Ciprofloxacin resistance as (84%). All *E. coli* isolates (100%) were resistant to Penicillin G. Last researches described Penicillin as non-effective antibiotic against *E. coli* ***Kundu et al. (2019)***

Table (1): *Biochemical reactions of E. coli isolates*

Biochemical Tests	*E. coli*
Gram stain	Gram Negative bacilli
Oxidase test	-
Catalase test	+
Indole test	+
Methyl Red test	+
Voges-Proskauer test	-
H2S production on TSI	-
On TSI "slant"	Yellow color with acid production
On TSI "butt"	Yellow color, acid, and gas production
Citrate test	-
Urease test	-
Lactose, Mannitol, Glucose fermentation	+
Sucrose fermentation	Variable

Table (2): *Prevalence of E. coli in all raw meat and meat based products samples*

Meat and meat products	No. of *E. coli* isolates	%*
Pastirma	54	15.70
Minced meat	68	19.77
Frankfurter	11	3.20
Burger	57	16.57
Sausage	59	17.15
Luncheon	21	6.10
Raw meat	60	17.44
Salami	14	4.07
Total	344	100

***(%) calculated according to total no. of *E. coli* isolates= 344**

Table (3): *Antibiogram of E. coli isolates.*

Antimicrobial agent	***E. coli* (No. = 344)**					
	Sensitive		**Intermediate**		**Resistant**	
	No.	**%***	**No.**	**%***	**No.**	**%***
Doxycyclin (30µg)	0	0.00	285	82.85	٥٩	17.15
Nalidixic acid (30µg)	255	74.13	59	١٧,١٥	30	8.72
Neomycin (30µg)	10	2.91	295	85.76	39	11.34
Rifampin (5µg)	0	0.00	0	0.00	344	100
Chloramphenicol (30µg)	324	94.19	10	2.91	10	٢,٩١
Tobramycin (10µg)	0	0.00	344	100	0	0.00
Ofloxacin (5µg)	265	77.03	79	22.97	0	0.00
Erythromycin(15µg)	0	0.00	0	0.00	344	100
Penicillin(10µg)	0	0.00	0	0.00	344	100
Bacteracin (10 µg)	0	0.00	0	0.00	344	100
Amikacin (30µg)	138	40.12	197	57.27	٩	2.62
Trimethoprim/Sulfamethoxazole(25µg)	٩٨	28.49	108	31.40	138	40.12
Cephradin (30µg)	0	0.00	0	0.00	344	100
Polymyxin B (300µg)	0	0.00	٣١٤	91.28	30	8.72
Amoxicillin/Clavulanic acid (30µg)	59	17.15	١٧٧	51.45	108	31.40
Oxalinic acid (2µg)	167	48.55	128	37.21	49	14.24
Vancomycin (30µg)	0	0.00	0	0.00	344	100
Ciprofloxacin (5µg)	0	0.00	0	0.00	344	100
Imipenem (10µg)	٣٣٠	95.93	١٤	4.07	0	0.00
Tetracycline (30µg)	0	0.00	0	0.00	344	100

***(%) calculated according to total no. of *E. coli* isolates= 344.**

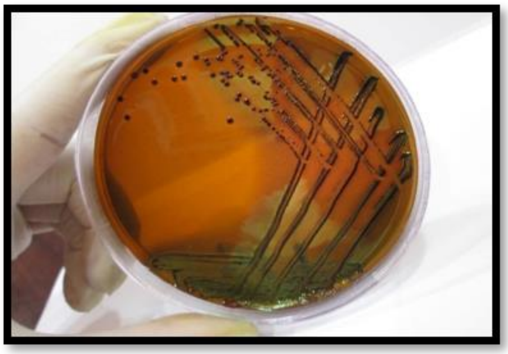

Figure (1): *E. coli* colonies on EMB.

Figure (2): *E. coli* colonies on TBX.

Figure (3) *E. coli* colonies o MacConkey's.

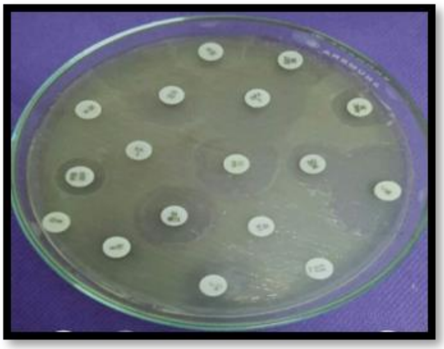

Figure (4): *E. coli* antibiogram.

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مدى تواجد الاشريشيا كولاي المعزولة من منتجات اللحوم في أسواق بورسعيد وحساسيتها للمضادات الحيوية

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الملخص العربي

جُمعت ٨٠٠ منتجات اللحوم عشوائياً من أسواق بورسعيد لعزل الاشريشيا كولاي و تحديد حساسيتها للمضادات الحيوية. تواجد الاشريشيا كولاي كان (٤٣٪) حيث كانت (19.٧٧%) في اللحم المفروم, (١٧.٤٤٪) في اللحم النيئ, (17.15%) في السجق, (16.57٪) في البرجر, (١٥.٧٪) في البسطرمة, (6.10%) في اللانشون, (4.07%) في السلامي و (3.2%) في الفرانكفورتر. الفحص السيرولوجي أوضح تواجد (O25, O26, O55, O63, O78, O86, O111, O112, O114, O119, O124, O125, O126, O136, O142, O157, O158 و عينات لم تصنف سيرولوجيًا). المعزولات حساسة للاميبينيم و الكلورامفينيكول و النالدكسيك اسيد و الاوفلوكساسين في حين كانت مقاومة للاريثروميسين و البنيسيلين و البكتيرسين و السيفراجين و الريفامبين و السيبروفلوكساسين و التيتراسيكلين و الفانكوميسين.