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Hygienic evaluation of meat handlers and equipment in meat preparation sector in some markets in Qalyubia Governorate.

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ABSTRACT

From two retail markets (A and B) in the Qalyubia Governorate of Egypt, 90 samples of raw beef burger, kofta and sausage were gathered and 30 swabs of employee hands, table surfaces and knives (10 of each) were equally collected to assess the hygienic practices used to handle these products, the collected samples were tested for the presence of *Salmonella* and *E. coli*. Additionally, it was done to check for food poisoning bacteria on knives, tabletop surfaces, and people who handle food. To be precise, in market (A), *S. aureus* was identified from 20% of worker hands and table surfaces and 30% of knife swabs. The results from the swabs taken from the worker hands, tabletops, and knives in market (B) were 30%, 30%, and 40%, respectively. However, *E. coli* was recovered from 20% of knives, 10% of worker hands, and table surfaces in market (A) and from 10% of worker hands, 20% of knives and table surfaces in market (B). Furthermore, the tested swabs from markets (A) and (B) were free from *Salmonellae*. *Salmonellae* were found in 6.67% of the beef burger and kofta samples from market (A), and in 6.67%, 6.67% and 20% of the beef burger, Kofta and sausage samples from market (B), respectively, Good hygienic processing practices for processing of meat products should be implemented, these practices include selection of good quality raw materials, cleaning and hygiene of work station, cleaning and sanitation of tools and equipment, good personal hygiene and hand sanitation and control of CCPs of the production process including temperature control from receiving of raw materials till displaying the end products.

1. INTRODUCTION

Food can be contaminated with surfaces and food employees during chopping, shredding, and serving. Pathogenic microorganisms are transmitted by direct contact with food or indirectly with airborne particles. this study aimed to determine the prevalence and the relationship between pathogenic microorganisms isolated from food, kitchen equipment and food handler's hands (Erdogani et al., 2020), Contamination Beef available at retail outlets has gone through a long chain process before it is ready at the retails. The contamination risks were increased during the slaughter and processing of the carcasses. Contaminations also can be compounded during transportation, storage, and handling of meat by retailers (Ahmad *et al*., 2013).

Salmonella is a bacterium that can cause food products to become contaminated during or after processing, according to (Gilbert *et al.* 2016). Ready-to-eat (RTE) food is not subjected to any testing to assure its safety prior to consumption, so the danger of contracting a disease transmitted by food must be taken into account if salmonella is present.

4% of the chicken meat tested positive for *E. coli.* According to Hashem 2015, 50% of isolated *E. coli* were *E. coli* O55 and 50% were *E. coli* O86A.

High amounts of cross-contamination are primarily caused by the water used to dress chickens. Samples from Lusaka's two primary poultry abattoirs were bacteriologically analysed and found that *E. coli* and *Salmonella* contamination were detected in 70% and 2.5% of the selected dressed chickens respectively. The number of total coliforms and *E. coli* were observed to be significantly higher in samples from washed carcasses than pre-washed carcasses (65 and 35%) (Mpundu *et al.*, 2019).

In this study, microorganisms crucial for food safety and public health will be found on food, surfaces, and staff hands. Additionally, the relationship between cross-contamination and sources of contamination between these isolates will be analyzed.

Tafida *et al* (2013) reported that Salmonella is among the most important food borne pathogens worldwide contaminating a wide range of animal products including meat products. Human illnesses due to this pathogen are attributed to poor biosecurity in production, improper processing and handling of meat and meat products. This is more likely where surveillance and regulatory control is weak.

Foods contaminated with *S. aureus* are a potential vehicle for the transmission of enterotoxigenic *S. aureus* to humans. This contamination can occur in the following ways: i) food contact surfaces, ii) food handlers, iii) food-producing animals, iv) tools used in processing, v) air, and vi) dust. Among these, the primary source of food contamination is via manual contact or respiratory secretions, which is caused by food handlers carrying *S. aureus* producing enterotoxin in their noses or on their hands (Chaalal *et al.,* 2018).

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2. MATERIAL AND METHODS

2.1. Collection of samples:
In the butchery departments of two different hypermarkets (A and B) in Benha city, Qalyubia governorate, Egypt, 45 samples of meat products of chilled beef burger, kofta and sausage (15 of each) and 30 swabs of employee hands, table surfaces and knives (10 of each) were collected.
This research was approved by Institutional Animals Care and Use Committee of faculty of veterinary medicine, Benha University (approved number BUFVTM 07-04-23).

2.2. Bacteriological examination:
Preparation of samples (ICMSF, 1996):
For the preparation of tenfold serial dilutions, 225 ml of sterile peptone water 0.1% was added to 25 g. of sample and carefully blended using a sterile blender for 1.5 minutes. The following tests were performed on the prepared samples.
Preparation of swabs:
Plastic tubes with sterile cotton screw caps that are ready for use that were used to simulate swabs.
Preparation of templates:
To define the sampling region, a metal template with an exposed inner area of 10 cm2 (2 x 5 cm) was employed. The template was sterilized in a hot air oven at 180oC for 20 minutes while being wrapped in aluminum foil.
Preparation of rinsing fluid:
As a rinse and diluting fluid, 1% buffered peptone water was employed. Small heat-resistant screw-capped tubes containing 10 ml of washing fluid each were filled with the solution before being sterilized in the autoclave for 20 minutes at 121° C.
Swabbing of selected surfaces:
After using a sterile cotton swab and template, swabs were obtained from worker hands, table surfaces, and blades. To restrict the region being studied, the sterilised template was firmly pressed against the surface. Using a sterile cotton swab that was removed from plastic tubes with screw-on caps and soaked with 1% buffered peptone water for rinsing. then moved over the constrained space.
Screening for Salmonellae:
Pre-enrichment broth:
One ml of the original dilution was used to inoculate sterile peptone water, and the mixture was then cultured for 18 hours at 37°C
Enrichment broth:
A 9 ml Rappaport Vassilidis broth tube was inoculated with 1 ml of the original dilution, and the tube was then incubated at 43 °C for 24 hours (Harvey and Price, 1981).
Selective Plating:
The agar Xylose Lysine Deoxycholate (XLD) was employed. After being individually streaked onto XLD agar, for 24 hours at 37°C. Suspected colonies were red with or without black centers. The suspected colonies were sub-cultured onto nutrient agar plate and incubated at 37□C for 24 hours. Thus, the separate colonies were selected and streaked onto slope nutrient agar for further identification.
Serological identification of Salmonellae, (Kauffman, 1974).
Statistical Analysis: Feldman et al(2003).
Morphological examination (ISO, 1995).
Biochemical identification (MacFaddin, 2000).
Screening for Enteropathogenic Escherichia coli:
Pre-enrichment (ICMSF, 1996):
One milliliter of the initial dilution was added to MacConkey broth tubes along with inverted Durham's tubes as an additional source of inoculum. The inoculated tubes underwent a 24-hour incubation period at 37°C.
Enrichment broth:
One ml of a positive MacConkey tube was used to inoculate the MacConkey broth tubes, which were then incubated for 24 hours at 37°C. Suspected colonies were metallic green in color. Suspected colonies were purified and inoculated into slope nutrient agar tubes for further identification.
Plating media:
Eosin Methylene Blue agar medium (EMB) was streaked on MacConkey broth tubes and incubated at 37°C for 24 hours.
Staining (Cruickshank et al., 1975):
Biochemical identification (MacFaddin, 2000):
Serodiagnosis of E. coli:
According to Kok et al. (1996), the isolates were identified using a serological test.
Determination of S. aureus count (FDA, 2001):
Using a sterile bent glass spreader, successive dilutions from each of the previously prepared one ml preparations were distributed over a Baired Parker agar plate. The plates were kept upright for about 10 minutes while the inoculums were absorbed by the agar, or they were left upright in the incubator for about an hour. The inoculated and control plates were turned over and left to incubate for 48 hours at 37°C. The developed black colonies surrounded by clear halo zones were enumerated and *S. aureus* count /g was calculated. Also, the colonies were picked up and purified on nutrient agar slopes for further identification.

3. RESULTS

Tables 1 and 2 show the prevalence of the food poisoning bacteria (S. *aureus, E. coli*, and Salmonellae) in swabs taken from worker hands, table surfaces, and knives in Markets (A) and (B) respectively. The results showed that the incidence of S. aureus was 20%, 20%, and 30% in worker hands, table surfaces and knives while, E. coli was 10% in both samples of worker hands, knives in market (A). In market (B), the incidence of 30%, 30%, and 40% was in *S. aureus*, 10%, 10%, and 20% was in E coli in worker hands, table surfaces, and knives samples , respectively . All swab samples were free from Salmonellae.

Table 1 Incidence of food poisoning bacteria in the swabs taken from meat handlers and equipment's at retail markets (A) (n=10).

Swabs	Worker hands		Table surfaces		Knives	
Pathogens	No.	%	No.	%	No.	%
Salmonella	0	0	0	0	0	0
E. coli	1	10	0	0	1	10
S. aureus	2	20	2	20	3	30

Table 2 Incidence of food poisoning bacteria in the swabs taken from meat handlers and equipment's at retail markets (B) (n=10).

Swabs / Pathogens	Worker hands		Table surfaces		Knives	
	No.	%	No.	%	No.	%
Salmonella	0	0	0	0	0	0
E. coli	1	10	1	10	2	20
S. aureus	3	30	3	30	4	40

Table 3 shows that *Salmonellae* were found in 6.67%, 13.33%, and 6.67% of the analyzed kofta and sausage in market (A) and in 6.67%, 6.67%, and 20% of the samples in market (B), respectively.

Table 3 Incidence of Salmonellae contaminating meat products at retail markets (n=15).

Butchery section	A		B	
Meat products	No.	%	No.	%
Beef burger	-	-	1	6.67
Kofta	1	6.67	1	6.67
Sausage	2	13.33	3	20
Total (45)	3	6.67	5	11.11

Tables 4 and 5 recorded the identification of *Salmonellae* species, which isolated from examined meat samples in the market (A&B). Results showed that *S. Enteritidis* and *S. Montevideo* were isolated from 6.67% of sausage samples while *S. Typhimurium* was isolated from 6.67% of kofta in market (A). On the other hand in market (B) *S. Enteritidis* was isolated from 6.67% of kofta, *S.Haifa*, *S. infantis* and *S.Typhimurium* were isolated from 6.67% of sausage while *S.Typhimurium* was isolated form 6.67% from beef burger samples.

Table 4 identification of Salmonellae detected in meat products at retail market (A) (n=15).

Meat products / Salmonella Strains	Beef burger		Kofta		Sausage		Group	Antigenic structure	
	No.	%	No.	%	No.	%		O	H
S. Enteritidis	-	-	-	-	1	6.67	D1	1,9,12	g,m : 1,7
S. Montevideo	-	-	-	-	1	6.67	C1	6,7	g,m,s : 1,2,7
S. Typhimurium	-	-	1	6.67	-	-	B	1,4,5,12	i : 1,2
Total	-	-	1	6.67	2	13.33			

Table 5 Identification of Salmonellae detected in meat products at retail market (B) (n=15).

Products / Strains	Beef burger		Kofta		Sausage			Antigenic structure group	
	No.	%	No.	%	No.	%		O	H
S. Enteritidis	-	-	1	6.67	-	-	D1	1,9,12	g,m : 1,7
S. Haifa	-	-	-	-	1	6.67	B	1,4,5,12	Z10: 1,2
S. Infantis	-	-	-	-	1	6.67	C1	6,7	r : 1,5
S. Typhimurium	1	6.67	-	-	1	6.67	B	1,4,5,12	i : 1,2
Total	1	6.67	1	6.67	3	20			

According to Table (6), the *Salmonellae*-based unacceptability of the analyzed samples was 6.67% and 13.33% in kofta and sausage in market (A) and 6.67%, 6.67%, and 20% in beef burger, kofta, and sausage samples in market (B).

Table 6 Acceptability of Salmonellae in the examined samples of meat products at retail markets according to EOS (2005).

Butchery	*Salmonellae* /25g*	A				B			
		Accepted		Unaccepted		Accepted		Unaccepted	
Meat Products		No.	%	No.	%	No.	%	No.	%
Beef burger	Free	15	100	0	0	14	93.33	1	6.67
Kofta	Free	14	93.33	1	6.67	14	93.33	1	6.67
Sausage	Free	13	86.67	2	13.33	12	80	3	20

Escherichia coli was isolated from 6.67%, 13.33 and 26.67% of the samples in market (A) and 20%, 33.33% and 33.33% of the samples in market (B) in beef burger, kofta and sausage samples, respectively (Table 7). In addition to serotyping of the isolated *E.coli* from the meat samples in Market (A), were O26:H11 (EHEC) (6.67%) in beef burger, O111:H2 (EHEC) (6.67%) and O127:H6 (ETEC) 6.67% in kofta, O86 (EPEC) (6.67%), O111:H2 (EHEC) (13.33%) and O121:H7 (EHEC) (6.67%) in sausage samples (Table 8).

Table 7 Incidence of E. coli contaminating meat products at retail markets (n=15).

Butchery	A		B	
Meat products	No.	%	No.	%
Beef burger	1	6.67	3	20
Kofta	2	13.33	5	33.33
Sausage	4	26.67	5	33.33

Table 8 Serological identification of E. coli detected in meat products at retail market (A) (n=15).

Products / *E.coli* strains	Beef burger		Kofta		Sausage		Strain characteristics
	No.	%	No.	%	No.	%	
O26 : H11	1	6.67	-	-	-	-	EHEC
O86	-	-	-	-	1	6.67	EPEC
O111 : H2	-	-	1	6.67	2	13.33	EHEC
O121 : H7	-	-	-	-	1	6.67	EHEC
O127 : H6	-	-	1	6.67	-	-	ETEC
Total	1	6.67	2	13.33	4	26.67	

Results in Table (9) recorded serotyping of *E. coli* from examined meat samples in the market (B) were *O20* (EPEC) (6.67%) from sausage, *O26: H11* (EHEC) (6.67%) from each beef burger and kofta, O44: H18 (EHEC) (6.67%) from sausage, while *O111: H2* (EHEC) (6.67%, 13.33 and 6.67%) from burger, kofta and sausage, while *O114:H4* (EPEC) (6.67%) and *O124* (EIEC) (6.67%) from kofta samples only.

Regarding the acceptability of *E. coli* in the examined samples according to EOS (2005) 26.67, 33.33% and 46.33 % in the market (A), 40%, 53.33% and 60%in beef burger, kofta and sausage samples in the market (B) were unacceptable (Table10).

Table 9 Serological identification of E. coli detected in meat products at retail market (B) (n=15).

Products / *E.coli* strains	Beef burger		Kofta		Sausage		Strain characteristics
	No.	%	No.	%	No.	%	
O20	-	-	-	-	1	6.67	EPEC
O26 : H11	1	6.67	1	6.67	-	-	EHEC
O44 : H18	-	-	-	-	1	6.67	EHEC
O111 : H2	1	6.67	2	13.33	1	6.67	EHEC
O114 : H4	-	-	1	6.67	-	-	EPEC
O124	-	-	1	6.67	-	-	EIEC
O128 : H2	1	6.67	-	-	2	13.33	ETEC
Total	3	20	5	33.33	5	33.33	

Table 10 Acceptability of the examined samples of meat products at retail markets based on their E. coli (n=15).

Butchery / Meat Products	*E. coli* /25g*	A				B			
		Accepted		Unaccepted		Accepted		Unaccepted	
		No.	%	No.	%	No.	%	No.	%
Beef burger	Free	11	73.33	4	26.67	9	60	6	40
Kofta	Free	10	66.67	5	33.33	7	46.67	8	53.33
Sausage	Free	8	53.33	7	46.67	6	40	9	60

4. DISCUSSION

In order to eradicate or reduce microorganisms of concern to an acceptable level, food must be cooked or processed before it is ready for human consumption (CFS, 2014).

S. aureus, E. coli and *Salmonellae* were detected in swabs taken from worker hands, table surfaces, and knives in both Markets (A) and (B) as Table (1). Incidence of *S. aureus was* less than what recorded by Mohtaram *et al.* (2017) who isolated it from 46% of the samples and 10% in both swab samples of worker hands and knives contained *E. coli* but table surfaces samples were free. Also, this is less than that (Mohtaram *et al* 2017) results, who was not isolated from all the samples from Market (A) but from 29% of the samples in addition to *Salmonallae.* Additionally, the prevalence of *S.aureus* poisoning found in samples from worker hands, table surfaces, and knives in Market (B) was 30%, 30%, and 40%, respectively. *Salmonella* was not isolated from all samples, while *E. coli* was isolated from 10%, 10%, and 20% of the analyzed exchanges, respectively.

In Table (3) results outcome is better than that was observed by (Osama *et al.,* 2021) who isolated *Salmonellae* from 5% of the Kofta samples.

Salmonellae species that were identified from meat samples in the market (A) are included in Table (4).

The obtained results at tables (4&5) revealed that burger at market A were similar to those recorded by Ibrahim (2001) and Zaki –Eman (2003) as they failed to isolate *Salmonellae spp.* in burger samples.

The results of burger at market B were nearly similar to that recorded by *Saad et al. (2011)* (5%), but lower than that obtained by Mousa et al (2014) (20%), also higher than that reported by Usama (2009) (2.5%).

Furthermore, the achieved results of kofta were nearly similar to that recorded by Zaki (2003) (5%) and Shaltout *et al*. (2013) (8%), but lower than that recorded by Ghanem (2009) (13.33%), Also higher than that obtained by Usama (2009) (2.5%).

The obtained results of sausage were lower than those obtained by Mousa *et al* (2014) (40%) and Sobieh (2014) (26.67%), but higher than that recorded by Zaki (2003) (5%) and El Maghraby (2014) (12%).

According to Table (6), Such *Salmonella spp*. were previously isolated from RTE meat products by Ghanem (2009) isolated *S. Enteritidis* (4.4%) and *S. Typhimurium* (6.67%), Shaltot *et al*. (2013) isolated *S. Enteritidis* (4%) and *S. Typhimurium* (4%) and Sobieh (2014) *S. Enteritidis* (4.4%) and *S. Typhimurium* (6.6%).

This outcome is better than that (Osama et al., 2021) isolated *E. coli* from 8% of tested Kofta samples. Food reputation refers to opinions on how food affects its consumers. It is seen to be essential for hotel guests' health and safety.

Regarding to burger, higher incidences were recorded by Ibrahim (1991) (36%) and Zaki (2003) (35%).

The current results of kofta were agree to some extent to that obtained by Sobieh (2014) (13.33%), but higher than *Tavakoli and Riazipour (2008)* (12.6%) and *Saad et al. (2011)* (10%), and lower than Zaki (2003) (50%) and Abdel Fattah (2014) (40%).

On the other hand, the present study for sausage was nearly similar to that obtained by Ibrahim (2008) (25%) and Sobieh (2014) (26.67%), but higher than Al-Mutairi (2011) (12%) and lower than Zaki (2003) (40%) and Alrais (2008) (36%).

5. CONCLUSION

All food serving facilities should adhere to Good Hygienic Practices (GHP), which are crucial for ensuring the safety of the food being served. Contamination of meat products with such serious pathogens remains as a public health problem, thus all precautions of proper sanitation during manufacture, handling and storage of such meat products should be adopted to control these serious pathogens and to obtain a maximum limit of safety to consumers.

Good hygienic practices for processing of meat products should be implemented, these practices include selection of good quality raw materials, cleaning and hygiene of workstation, cleaning and sanitation of tools and equipment, good personal hygiene and hand sanitation and control of CCPs of the production process including temperature control from receiving of raw materials till displaying the end products.

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