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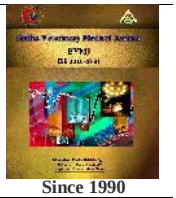

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Original Paper

Bacteriological evaluation to Foodborne Pathogens in Ready to Eat Meats

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ABSTRACT

This study was conducted on 90 random samples of cooked chicken and beef meats (45 of each) with 125 gm weight of each sample .The beef meat samples were represented by beef kabab , beef kofta and shawerma (15 samples each) while the chicken meat samples were represented by fried pane , fried drum sticks and chicken shawerma (15samples each) which were collected from Kafr Elzayat city, Gharbia governorate to evaluate the bacteriological quality of ready to eat chicken and beef meats. The results of bacteriological examination for beef and chicken recorded that *E.coli* were isolated from three beef kabab samples from four grilled beef kofta samples, and from seven beef shawerma. *E.coli* isolates from fried chicken pane were 4. While 6 isolates were retrieved from fried chicken drumstick samples and 9 isolates from chicken shawerma. The study revealed the edibility of samples based on *Staph. aureus* count. *Salmonella entretidis* (*S. entretidis*) was isolated from grilled beef kofta , beef shawerma, chicken pane and chicken shawerma with a percentage 6.67,13.33, 6.67 and 6.67% respectively Also, *S. typhimurium* was isolated from the examined samples of beef kabab, beef kofta, beef shawerma, chicken drumstick and chicken shawerma , chicken drumstick and chicken shawerma.

1. INTRODUCTION

Ready to eat meats area unit thought-about as a perfect substance for growth of many organisms, as its high content of moisture, minerals, element compound, some possible carbohydrates (glycogen) and its pH favorable for many microorganisms(Al-Mutairi, 2011).

Contamination of food with infective microorganisms, specially enteric bacteria and coliform may be due to poor microbial quality of raw materials, inadequate personal hygiene and an extended amount between production and consumption at temperature (Kiipliilii et al., 2003).

E.coli is common non-virulent organism however some strains have infective or toxigenic virulence factors that render them virulent to human. It is recognized as a heavy foodborne infectious agent and related to varied outbreaks of sickness ensuing from contaminated meat product (Gi et al., 2009).

Enteric bacteria are one among the microorganisms significantly related to foodborne outbreaks of malady. Meat product and poultry area unit the foremost widespread sources of malady by enteric bacteria (Siqueira et al., 2003). *Salmonella* species are the main reason for gastroenteritis, illness, hospitalization and death every year (CDC, 2006).

Staph. aureus is one among the microorganisms oftentimes related to foodborne outbreaks of malady infecting intestinal flu and barely any inheritable directly from meat however largely happens because of excessive handling or contamination throughout or once change of state of meat and meat product (Busani et al., 2005).Therefore, the aim of the current study was to evaluate the bacteriological quality of ready to eat chicken and meats samples

2. MATERIAL AND METHODS

2.1. Collection of samples:

Ninety random beef and chicken meats (45 each) were collected from different markets in Kafr Elzayat town, Gharbia governorate, Egypt. The beef samples were represented by kabab, kofta and meat shawerma (15 each), whereas the chicken meats were represented by fried pane, fried drumstick and chicken shawerma (15 each). Every sample was kept in separated sterile bag and preserved in an ice box then transferred to the laboratory to be examined bacteriologically.

2.2. Sampling according to (ISO 4833-1, 2013):

Sterile peptone water (225 ml, 0.1%) were added to 25 gram of the examined sample, thoroughly mixed using sterile blender for 1.5 minutes, from which tenfold serial dilutions were prepared. The prepared samples were subjected to bacteriological examinations.

3.2. Bacteriological examination:

3.2.1. Isolation and Identification of Staphylococcus aureus

Using Baired parker agar plate at 37°C for 48 hrs.

Determination of Staph. aureus count was done according to FDA, (2001).

Morphological examination (ISO, 1995).

Biochemical identification after McFadden (2000).

Detection and typing of enterotoxin following shingaki et al. (1981).

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3.2.2. Isolation and Identification of E.coli Using MacConkey broth tubes at 37°C for 24 hours
Identification of *Enteropathogenic E.coli* followed ISO (1995) and MacFaddin (2000)
Serological identification of *E. coli* (Kok et al. , 1996).
The isolates were serologically identified by using rapid diagnostic *E.coli* antisera sets (DENKA SEIKEN Co., Japan) for diagnosis of the Enteropathogenic types.

3.2.3. Isolation and Identification of Salmonellae according to ISO (1995) Using XLD media at 37 °C for 24 hrs.
Serological identification of Salmonellae (Kauffman, 1974): Serological identification of *Salmonellae* was carried out for the determination of Somatic (O) and flagellar (H) antigens using *Salmonella* antiserum (DENKA SEIKEN Co., Japan).

3.3. Statistical analysis
Data were analyzed using the descriptive statistic SPSS (Version 20). Min., Max., Mean and Standard Error were calculated.

3. RESULTS AND DISCUSSION

Foodborne diseases caused by *E. coli, Salmonella* species and *S. aureus* represent a significant public unhealthiness worldwide. These pathogens square measure transmitted in the main through consumption of contaminated food and therefore the presence of those organisms in meat has relevant public health implications (Normanno et al., 2005 and Sousa, 2008). Dangerous microorganisms are widely spread in soil, water, animals and food handlers. These microorganisms spread on hands, clothes, utensils and cutting boards; the slight contact will transfer them to meat and chicken meals and cause food-borne diseases. Raw food, particularly meat, poultry and their juices have dangerous microorganisms which can be transferred to different food throughout preparation and storage (*FAO/WHO, 2003*). USFDA (2012) reported that S*taphylococci* exist in air, dust, sewage and food or on food equipment, environmental surfaces, humans and animals. Humans and animals are the primary reservoirs.
Staphylococci are present in the nasal passages and throats and on the hair and skin of 50 % or more of healthy individuals. Although food handlers are usually the main source of food contamination in food poisoning outbreaks, equipment and environmental surfaces can also be sources of contamination with *S .aureus*.
The results of the present work illustrated in table (1) indicated the mean value of total staphylococcal count in the examined samples of ready to eat beef and chicken meat meals were $5.08\times10^2\pm 0.79\times10^3$/g for beef kabab, $1.34\times10^3\pm 0.21\times10^3\times10^3$/g for beef kofta, $2.81\times10^3\pm 0.46\times10^3$/g for beef shawerma, $1.96\times10^3\pm 0.28\times10^3$/g for chicken pane, $4.71\times10^3\pm 0.52\times10^3$/g for chicken drum stick and $6.29\times10^3\pm 1.04\times10^3$/g for chicken shawerma.
Table (2) revealed that SET- RPLA test proved that only one enterotoxin A&D strain was isolated from beef kofta with a percentage 6.67%, one enterotoxin A and one enterotoxin C&D strain from beef shawerma. while, chicken meals samples one enterotoxin A strain and one strain enterotoxin C of chicken drum stick, two enterotoxin A and one enterotoxin A&D strain were isolated from chicken shawerma. Nearly similar results were obtained by Nassar (1988); Yassien and El-Essawy (1990) and Moussa et al. (1992) who found that the mean values of *S.aureus* count (cfu/g) was 5.8×10^4 within the examined samples of ready to eat meat, Al-Kour (2001) found that the mean of total *staph. aureus* count was $4.13\times10^3 \pm 1.25\times10^3$/g within the examined samples of kofta. El-Taher (2009) found that the mean of the *staphylococci* count was 6.91×10^3 within the examined samples of cooked kofta. Whereas, Mohamed (2000*)* did not isolate *S.aureus* from any of the examined samples of heat treated meat product. Abd Allah and Hassan (2000) founded that the mean of *staphylococci* count was 1.2×10^2(cfu/g) within the examined samples of grilled shawerma. However, higher findings were obtained by Kirralla (2007) who found that the mean of *S.aureus* counts within the examined samples of cooked meat was 2.45×10^5/g . Al-Tawwab (2004) found that the mean of *staph aureus* count was $3.1\times10^6 \pm 4.9\times10^5$, $5\times10^4 \pm 6.7\times10^3$ within the examined samples of kofta and shawerma, respectively. Regarding the results of SET- RPLA nearly like that recorded by Rosec et al. (1997)*;* Abd alslam et al. (2014) and Ezzat et al.(2014);Abdalrahman et al. (2015); Abd El-Tawab et al. (2015) and Afifi-Dina (2016)who found enterotoxin A; C;D and A&C in meat meal samples.

Table 1 *S. aureus* count/g in the examined samples of ready to eat meats (n=15).

	Ready-to-eat meats	Min.	Max.	Mean ± S.E*
	Kabab	1×10^2	9×10^2	$5.08\times10^2 \pm 0.79\times10^3$
Beef	Kofta	1×10^2	4×10^3	$1.34\times10^3 \pm 0.21\times10^3$
	Shawerma	1×10^2	7×10^3	$2.81\times1^3\pm 0.46\times10^3$
	Pane	1×10^2	5×10^3	$1.96\times10^3\pm 0.28\times10^3$
Chicken meat:	Drumstick	1×10^2	9×10^3	$4.71\times10^3\pm 0.52\times10^3$
	Shawerma	1×10^2	2×10^4	$6.29\times10^3\pm 1.04\times10^3$

S.E* = standard error of mean

Table 2 Prevalence of enterotoxin secreted by *S. aureus* strains isolated from ready to eat meats.

	Ready-to-eat meats	A		C		A & D		C & D	
		No.	%	No.	%	No.	%	No.	%
	Kabab	-	-	-	-	-	-		-
Beef	Kofta	-	-	-	-	1	6.67		
	Shawerma	1	6.67	-	-			1	6.67
	Pane	-	-	-	-	-	-	-	-
Chicken meat:	Drumstick	1	6.67	1	6.67	-	-	-	-
	Shawerma	2	13.33	-	-	1	6.67	-	-

As presented in table (3&4) *E.coli was isolated as* 3 (20%) from beef kabab samples, 4 (26.67%) from grilled beef kofta samples, 7 (46.67%) isolates from grilled beef shawerma. The serological identify were O_{26}:H_{11}(6.67%),

O_{127}:H_6(6.67%) and O_{146}:H_{21}(6.67%) from beef kabab, in add to O_{91}:H_{21}(6.67%), O_{111}:H_2(6.67%) and O_{128}:H_2 (13.33%) from beef kofta , from beef shawerma were O_8:H_{21}(6.67%), O_{26}:H_{11}(20%), O_{44}:H_{18}(6.67%), O_{124} (6.67%) and O_{128}:H_2(6.67%).

Chicken meat samples were detected that *E.coli* recorded as 4 (20%) from chicken pane , 6 (26.67%) isolates from fried chicken drumstick samples, 9 (46.67%) isolates from grilled chicken shawerma. The serological identification was O_2:H_6 (6.67%), O_{78} (6.67%), O_{91}: H_{21}(6.67%) and O_{121}:H_7(6.67%) from chicken pane. O_1:H_7(6.67%) ,O_{78}(13.33%) , O_{91}:H_{21}(6.67%), O_{153}:H_2(6.67%) and O_{159}(6.67%) was isolated from chicken drumstick. wherever chicken shawerma isolates were O_1:H_7(6.67%),O_{78}(20%),O_{26}:H_{11}(6.67%),O_{113}:H_2(6.67%), O_{117}:H_4(6.67%)and O_{125}:H_{21}(13.33%). Such *Enteropathogenic E.coli* were antecedently isolated from completely different ready-to-eat meat product by Yassien (1992), Soliman and El-Tabiy (2006) and El-Rayes(2008) achieved that the incidence of serologically known *E-coli* isolated from the examined samples of kofta were five isolates one O26:K60 (B6) EHEC (4%) whereas different 2 serotype were O_{86}:K_{61} (B_7) EPEC (8%) on the opposite hand 2 serotype were O_{124}:K_{72} (B_{17}) EIEC (8%). Alfaro (2017) found that *E.coli* bacteria can be transmitted through consumption of raw or rare ground beef. It can be also passed from person to person through improper hygiene. Salmonella is an enteric bacterium found in domestic and wild animal populations. *Salmonella* colonized animals and expose humans to this pathogen through meat, raw milk, water, and environment and direct contact (Guo *et al.* 2011).

Table 3 Prevalence of *E. coli* isolated from the examined samples of ready to eat meat (n=15).

Beef Meals / *E.coli* strains	Kabab		Kofta		Shawerma		Strain characteristics
	No.	%	No.	%	No.	%	
O8 : H21	-	-	-	-	1	6.67	EPEC
O26 : H11	1	6.67	-	-	3	20	EHEC
O44 : H18	-	-	-	-	1	6.67	EPEC
O91 : H21	-	-	1	6.67	-	-	EHEC
O111 : H2	-	-	1	6.67		-	EHEC
O124	-	-	-	-	1	6.67	EIEC
O127 : H6	1	6.67	-	-	-	-	ETEC
O128 : H2	-	-	2	13.33	1	6.67	ETEC
O146 : H21	1	6.67	-	-	-	-	EPEC
Total	3	20	4	26.67	7	46.67	

Table (4) Prevalence of *E. coli* isolated from the examined samples of ready to eat chicken meat (n=15).

Chicken meats / *E.coli* strains	Pane		Drumstick		Shawerma		Strain characteristics
	No.	%	No.	%	No.	%	
O1 : H7	-	-	1	6.67	1	6.67	EPEC
O2 : H6	1	6.67	-	-	-	-	EPEC
O26 : H11	-	-	-	-	1	6.67	EHEC
O78	1	6.67	2	-	3	20	EPEC
O91 : H21	1	6.67	1	6.67	-	-	EHEC
O113 : H2	-	-	-	-	1	6.67	EPEC
O117 : H4	-	-	-	-	1	6.67	EHEC
O121 : H7	1	6.67	-	-	-	-	EHEC
O125 : H21	-	-	-	-	2	13.33	ETEC
O153 : H2	-	-	1	6.67	-	-	EPEC
O159	-	-	1	6.67	-	-	EIEC
Total	4	20	6	26.67	9	46.67	

EPEC = Enteropathogenic *E.coli.* ETEC = Enterotoxigenic *E.coli.* EIEC = Enteroinvasive *E.coli.* EHEC = Enterohaemorrhagic *E.coli*

Table 5 Prevalence of *Salmonella* organisms isolated from the examined samples of ready to eat meats (n=15).

Beef Meals	Kabab		Kofta		Shawerma		Group	Antigenic structure	
Salmonella Strains	No.	%	No.	%	No.	%		O	H
S. Enteritidis	-	-	1	6.67	2	13.33	D1	1,9,12	g,m : -
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. Muenster	-	-	1	6.67	-	-	E1	3,10,15,34	e,h : 1,5
S. Typhimurium	1	6.67	2	13.33	1	6.67	B	1,4,5,12	i : 1,2
S. Virchow	-	-	-	-	1	6.67	C1	6,7,14	r : 1,2
Total	2	13.33	4	26.67	5	33.33			

Table 6 Prevalence of *Salmonella* organisms isolated from the examined samples of ready to eat chicken meats (n=15).

Chicken meats	Pane		Drumstick		Shawerma		Group	Antigenic structure	
Salmonella Strains	No.	%	No.	%	No.	%		O	H
S. Apeyeme	-	-	1	6.67	-	-	C3	8,20	Z38 : -
S. Enteritidis	1	6.67	-	-	1	6.67	D1	1,9,12	g, m : -
S. Kentucky	1	6.67	1	6.67	3	20	C3	8,20	i : Z6
S. Labadi	-	-	-	-	1	6.67	C3	8,20	d : Z6
S. Molade	-	-	-	-	1	6.67	C2	8,20	Z10 : Z6
S. Papuana	-	-	-	-	-	-	C1	6,7	r : e,n,Z15
S. Tsevie	-	-	1	6.67	-	-	B	4,5	i : e,n,z15
S. Typhimurium	-	-	1	6.67	1	6.67	B	1,4,5,12	i : 1,2
Untypable	1	6.67	-	-	-	-	------	------	------
Total	3	20	4	26.67	7	46.67			

As tabulated in table (5&6) Concerning isolates of *salmonella* organisms in the examined samples; *S.* entretidis was isolated from grilled beef kofta (6.67%), beef shawerma (13.33%), chicken pane (6.67%) and chicken shawerma (6.67%). Also, *S.* typhimurium was isolated from 6.67%, 13.33 %, 6.67%, 6.67% and 6.67% of the examined samples of beef kabab, beef kofta, beef shawerma, chicken drumstick and chicken shawerma. Also S. *Kentucky* was isolated from chicken pane (6.67%) , chicken drumstick (6.67%) and chicken shawerma (20%). *S. muenster* was isolated only from beef kofta (6.67%), *S. Virchow* and *S. infantis* were isolated only from beef shawerma(6.67%), *S. haifa* were isolated only from beef kabab (6.67%), *S. Apeyeme* and *S. Tsevie* were isolated only from chicken drum stick (6.67%), *S. labadi* and *S. molade* were isolated only from chicken shawerma (6.67%) and un-typable salmonellae were isolated from chicken pane (6.67%). *Salmonella* microorganisms were antecedently isolated from ready-to-eat meat product by Soliman (1988), Ghoshal (1992), Elsherif and Mossalami (1998), Al-Kour (2001), Soliman et al. (2002) & Richardson and Stevens (2003). But no record for *Salmonella* isolates from ready-to-eat meat product by El-Hosseiny (1987), Rafaie & Mostafa (1990), Khalafalla (1996) & Kirralla (2007).

4. CONCULOSIONS

The achieved results in the current study allow concluding that ready to eat meat and chicken meats were contaminated with different types of microorganisms. This may be attributed to many causes mainly bad hygiene and post cooking contamination. Furthermore, the examined samples of chicken shawerma, chicken drumstick and beef shawerma were more contaminated with the highest level of microorganisms because such products may receive more handling during preparation as well as addition of spices which act as a source of contamination.

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