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Bacterial status of broiler chicken meat meals served at governmental hospital Edris A.M.¹, Islam Sabek¹, Ahmed Maroaf², Heba Rabea³

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ABSTRACT

Keywords	A grand total of 90 random samples with 250 gm weight of each sample of boiled, grilled and
Coliform.	fried broiler chicken meat meals (30 of each) were collected from a governmental hospital at various times in Kalyobia governorate, Egypt were collected for microbiological examination.
Enterobacteriaceae	The average values of APC, Enterobacteriaceae, coliform and staphylococcal counts were
Meat Meals	4.81x1040.65x104, 2.16x1040.41x104 cfu/g, 1.45x103036x103 and 3.10x103048x103 in
Staphylococci	boiled chicken meat meals,9.97x1032.18x103,5.73x1030.96x103, 9.74x1022.07 x102 and 1.26x1030.19x103 cfu/g in grilled chicken meat meals, 6.02x1031.33x103.,1.81x 1030.27x103,4.19x1020.53x102 and 7.58x1021.25x102 cfu/g in fried chicken meat meals.
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	Moreover, the incidence of serologically identified E. coli as Enteropathogenic E. coli (E. coli
Received 10/11/2020 Accepted 04/12/2020 Available On-Line 20/01/2021	O1:H7, E. coli O2: H6, E. coli O78, E. coli O124, E. coli O126: H21E. coli O128: H2and E. coli O146: H21), Enterotoxigenic E. coli (E. coli O2:H6, E. coli O78, E. coli O158, E. coli O128: H2 and E. coli O91: H21) Enterohaemorrhagic E. coli (E. coli O1:H7, E. coli O78 and E. coli O126:H21) and Enteroinvasive E. coli (E. coli O146 :H21). The public health importance of the isolated microorganisms and the recommended points were discussed.
	importance of the isolated microorganisms and the recommended points were discussed.

1. INTRODUCTION

Chicken meat is the most palatable easily prepared meat meals. and it is main source of protein, vitamins, fat, essential amino acids and minerals (Biesalski, 2005). More ever, it considered as a perfect media for growth of different organisms due to its relative humidity, the high level of nitrogenous compounds, minerals, glycogen and suitable pH for different microorganisms (Al-Mutairi, 2011). Food borne diseases are still occur and increased despite the improvement in food technology. The food which prepared in hospital play important role in increasing or decreasing the risk of disease as food free from contamination fast patient recovery (Custovicand Ibrahimagic, 2005). This risk of contamination of food differ according to nature of the microorganism, the level of contamination, the nature of food and especially the physiological state of consumer. Chicken meats need many processes to be ready to eat so the risk of contamination may increase during processing from hands, worker's clothes, knives or from the surrounding environment resulting in an inferior or unfit for human. The level of contamination can be measured using the aerobic plate count, Total Enterobacteriaceae, Total Coliforms and Escherichia coli biotype 1, which used as indicator for fecal contamination (Paulsen et al., 2006). E.coli, Salmonellae and coagulase positive S. aureus are the most pathogens detected in chicken meat. (Abdaslam et al., 2014; Ezzat et al., 2014). Some strains of E. coli have pathogenic or toxigenic virulence factors that make them virulent to human. So, it considered as a serious food borne pathogen which responsible for many outbreaks of disease (Gi et al., 2009). Food contaminated by S. aureus due to excessive

handling of food during or after cooking or due to ingestion of raw meat contaminated with this organism. Staphylococcal food poisoning characterized by nausea, vomiting, diarrhea lasting from 24 to 48 h and complete recovery occur within 1-3 days. The cooking method should be carefully applied to produce a temperature enough to kill all these microorganisms as most of them destroyed between 72°C to 83°C (Murphy et al., 2001). Therefore, the aim of present study was to evaluate the bacteriological status of chicken meat meals at governmental hospital.

2. MATERIAL AND METHODS

2.1. Collection of Samples:

Ninety random samples of boiled, grilled and fried broiler chicken meat meals (30 of each) with 250 gm weight of each sample were collected from a governmental hospital at various times in Kalyobia governorate, Egypt. Each sample was kept in a separated sterile plastic bag and preserved in an ice box then transferred to the laboratory under complete aseptic conditions without undue delay and then subjected to following examination.

2.2. Methods:

2.2.1. Preparation of Samples:

Under complete aseptic conditions, 25 grams of the sample were weighed and transferred into a sterile homogenizer flask containing 225 ml of sterile peptone water (0.1%). The content of the flask was homogenized for 3 minutes at 14000 rpm then allowed to stand for 5 minutes at room temperature. One ml of the homogenate was transferred into a separate tube containing 9 ml of sterile peptone water (0.1%) from

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which tenfold serial dilution were prepared. The prepared samples were subjected to the following examination, was prepared. The prepared samples were subjected to the following examination:

2.2.1. Determination of aerobic plate count (ISO, 2002).

2.2.2.Determination of Enterobacteriaceae count (ISO, 2004)

2.2.3. Determination of coliform count (FDA, 2002).

2.2.4. Determination of total Staphylococci count (FDA, 2001).

2.2.5. Serological Screening for Enteropathogenic Escherichia coli (Kok et al., 1996)

2.3. Statistical Analysis:

The obtained results were statistically evaluated by application of ANOVA test according to Feldman et al. (2003).

3. RESULTS

It is clear from the results recorded in table (1) that APC cfu/g in the examined samples differ from 6.3×10^3 to 2.0×10^5 with an average $4.81 \times 10^4 \pm 0.65 \times 10^4$ /cfu/g in boiled chicken meat meals, 5.8×10^3 to 4.1×10^4 with an average $9.97 \times 10^3 \pm 2.18 \times 10^3$ /cfu/g in grilled chicken meat meals and 1.2×10^3 to 1.5×10^4 with an average $6.02 \times 103 \pm 1.33 \times 10^3$ /cfu/g in fried chicken meat meals.

Table (2) showed high significant differences in APC in the examined samples (p< 0.01). Moreover, the results recorded that the higher average of Aerobic Plate Counts was recorded in the examined boiled chicken meat meals samples $(4.81 \times 10^4 \pm 0.65 \times 10^4 \text{cfu/g})$ and the lower one was in the examined fried chicken meat samples which was $(6.02 \times 10^3 \pm 1.33 \times 10^3 \text{cfu/g})$.

Table 1 Statistical analysis of Aerobic Plate Counts "APC" (cfu/g) in the examined chicken meat meal samples (n=30).

Chicken meals	ken meals Min		$Mean \pm S.E^{\ast}$
Boiled	6.3×103	2.0×10 ⁵	$4.81{\times}10^4{\pm}0.65{\times}10^4$
Grilled	5.8×103	4.1×10^{4}	$9.97{\times}10^3 \pm 2.18{\times}10^3$
Fried	1.2×103	1.5×10^{4}	$6.02{\times}10^3{\pm}1.33{\times}10^3$

Table 2 Analysis of variance (ANOVA) of APC in the examined samples of chicken meat meals.

9 2	251654.15		
	73161.48	36580.72	17.83++
7 1	178492.67	2051.64	
	2	2 73161.48 2 7 178492.67	2 73161.48 36580.72 7 178492.67 2051.64

It is appeared from the result recorded in table (3) that Enterobacteriaceae counts in the examined samples differ from $4.3x 10^3$ to $8.5x10^4$ with an average $2.16x10^4 \pm 0.41x10^4$ cfu/g in boiled chicken meat meals, $1.9x10^3$ to $2.3x10^4$ with an average $5.73x10^3 \pm 0.96x10^3$ cfu/g in grilled chicken meat meals and $7.0x10^2$ to $6.1x10^3$ with an average $1.81x10^3 \pm$ $0.27x10^3$ /cfu/g in fried chicken meat meals. Table (4) showed high significant differences in Enterobacteriaceae count in the examined samples (p < 0.01). The higher average of Enterobacteriaceae counts/g was recorded in the examined boiled chicken meat meal samples ($2.16x10^4 \pm 0.41x10^4$ cfu/g) and the lower one was in the examined fried chicken meat meal samples which was ($1.81 \times 10^3 \pm$ 0.27×10^3 cfu/g). The results given in table (5), it is obvious that the total coliform counts/(cfu/g) in the examined samples of boiled, grilled and fried chicken meat meal at hospital restaurant ranged from 1.0×10^2 to 6.2×10^3 cfu/g with an average of $1.45 \times 10^3 \pm 0.36 \times 10^3$ cfu/g for boiled chicken meat meal , 1.0×10^2 to 3.5×10^3 cfu/g with an average of $9.74 \times 10^2 \pm 2.07 \times 10^2$ cfu/g for grilled chicken meat meal and 1.0×10^2 to 9.0×10^2 cfu/g with an average of $4.19 \times 10^2 \pm 0.53 \times 10^3$ cfu/g for fried chicken meat. Table (6) showed high significant differences in total coliform counts in the examined samples (p < 0.01).

Results in table (7) indicated that the mean values of total staphylococcal count(cfu/g) in the examined samples of boiled, grilled and fried chicken meat meals at hospital restaurant were $3.10 \times 10^3 \pm 0.48 \times 10^3$ /cfu/g in boiled chicken meat, $1.26 \times 10^3 \pm 0.19 \times 10^3$ /cfu/g in grilled chicken meat, $7.58 \times 10^2 \pm 1.25 \times 10^2$ /cfu/g in fried chicken meats.

Table 3 Statistical analysis of Enterobacteriaceae counts (cfu/g) in the examined chicken meat meal samples(n=30).

Chicken meals	Min	$Mean \pm S.E^{\ast}$	
Boiled	4.3×103	8.5×10^{4}	$2.16{\times}10^4{\pm}0.41{\times}10^4$
Grilled	1.9×103	2.3×10^{4}	$5.73{\times}10^3{\pm}0.96{\times}10^3$
Fried	7.0×10^{2}	6.1×10^{3}	$1.81{\times}10^3{\pm}0.27{\times}10^3$

Table 4 Analysis of variance (ANOVA) of Enterobacteriaceae counts in the examined samples of chicken meat meals.

Source of variance	D.F	S.S	M.S	F. value
Total	89	147127.90		
Between Meals (M)	2	30889.81	15444.84	11.56++
Error	87	116238.09	1336.07	

++ = High significant differences (P<0.01)

Table 5 Statistical analysis of coliform counts (cfu/g) in the examined chicken meat meals samples (n=30).

Chicken meals	Min	Max	Mean \pm S.E [*]
Boiled	1.0×10^{2}	6.2×10 ³	$1.45{\times}10^3{\pm}0.36{\times}10^3$
Grilled	1.0×10^{2}	3.5×103	$9.74{\times}10^2{\pm}2.07{\times}10^2$
Fried	1.0×10^{2}	9.0×10^{2}	$4.19{\times}10^2{\pm}0.53{\times}10^2$

Table 6 Analysis of variance (ANOVA) of coliform counts in the examined samples of chicken meat meals.

Source of variance	D.F	S.S	M.S	F. value
Total	89	82796.73		
Between Meals (M)	2	10921.69	5460.86	6.61++
Error	87	71875.04	826.15	

++ = High significant differences (P<0.01)

Table 7 Statistical analysis of Staphylococcus counts (cfu/g) in the examined chicken meat meal samples (n=30).

Chicken meals	Min	Max	Mean \pm S.E [*]
Boiled	>10 ²	8.0×10 ³	$3.10{\times}10^3{\pm}0.48{\times}10^3$
Grilled	>10 ²	5.0×10 ³	$1.26{\times}10^3{\pm}0.19{\times}10^3$
Fried	>10 ²	1.3×103	$7.58{\times}10^2{\pm}1.25{\times}10^2$

Furthermore, table (8) declared that the incidence and serotyping of Enteropathogenic *E.coli* isolated from the examined samples of boiled, grilled and fried chicken meat at hospital were $O_1 : H_7 EPEC (6.67\%), O_{78} EPEC (10\%), O_2$: $H_6 EPEC (3.33\%), O_{128} : H_2 ETEC (6.67\%), O_{146} : H_{21}EPEC (3.33\%) and <math>O_{124} EIEC (3.33\%)$ were isolated in grilled chicken meat $O_2 : H_6 EPEC (3.33\%), O_{78} EPEC (6.67\%), O_{91} : H_{21} EHEC (3.33\%), O_{128} : H_2 ETEC (3.33\%) and <math>O_{158} EPEC (3.33\%)$ were isolated in fried chicken meat $O_1 : H_7 EPEC (3.33\%), O_{78} EPEC (6.67\%), O_{91} : H_{21} ETEC (3.33\%), O_{78} EPEC (3.33\%) and <math>O_{158} EPEC (3.33\%), O_{78} EPEC (6.67\%), O_{126} : H_{21} ETEC (3.33\%), and O_{146} : H_{21} EPEC (3.33\%).$

Table 8 Prevalence of different *E. coli* serotypes isolated from the examined samples of chicken giblets (n=30).

E. coli			Chicken meals				Strain
strains	Bo	oiled	Gr	Grilled Fried		ried	Characteristics
	No.	%	No.	%	No.	%	-
O1 : H7	2	6.67	0	0	1	3.33	EPEC
O2 : H6	1	3.33	1	3.33	0	0	EPEC
O78	3	10	2	6.67	2	6.67	EPEC
O91 : H21	0	0	1	3.33	0	0	EHEC
0124	1	3.33	0	0	0	0	EIEC
O126 : H21	0	0	0	0	1	3.33	ETEC
O128 : H2	2	6.67	1	3.33	0	0	ETEC
O146 : H21	1	3.33	0	0	1	3.33	EPEC
0158	0	0	1	3.33	0	0	EPEC
Total	9	30	6	20	4	13.33	

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

4. DISCUSSION

The result recorded in table (1) recorded that the total APC in examined samples high than that obtained by Oumokhtar (2000) who mentioned that the mean value of aerobic plate count in chicken meat was 2.9×10^4 cfu/g. Meanwhile, they lower than that recorded by Kirralla (2007) ($2.20 \times 10^6 \pm 2.12 \times 10^5$); El-Taher (2009) ($9.05 \times 10^3 \pm 2.51 \times 10^3$); Arab (2010) ($6.3 \times 10^4 \pm 0.35 \times 10^4$); Ali (2011) ($4.78 \times 10^5 \pm 0.96 \times 10^5$); Ibrahim et al. (2014) ($7.35 \times 10^4 \pm 1.17 \times 10^4$); Abd El-Aal (2015) $9.64 \times 10^4 \pm 2.25 \times 10^4$ in boiled chicken meat and $7.18 \times 10^4 \pm 1.44 \times 10^4$ in fried ones at hospital restaurant.

Accordingly, the boiled chicken meat was the most contaminated examined food followed by grilled chicken meat. This could be attributed to the fact that chicken meat may receive more handling during processing as well as scalding which may be source of contamination with larger number of microorganisms. Also, frying done at very high temperature which my reach to 220°C while boiling reach maximally to 100°C.

The obtained results may be explained as cooking cannot destroy all microorganisms, therefore, the holding of cooked foods at ambient temperature for several hours is the primary contributory factor for the growth and multiplication of such organisms (Bryan et al., 1997).

The result recorded in table (3) recorded that Enterobacteriaceae count in examined samples agree with those of Ibraheem (2005) in Assuit and El-Azhar universities. Meanwhile, the counts in boiled samples were higher than those recorded by Abd El-Daiem (2004) (4.2×10^3); Meanwhile, the counts in samples were lower than those recorded by Abbass (2011) ($2.84 \times 10^4 \pm 0.35 \times 10^4$); Saad et al., (2011) ($9.81 \times 10^3 \pm 2.66 \times 10^3$) and Abd El-Aal (2015) ($2.21 \times 10^4 \pm 0.38 \times 10^4$).

Enterobacteriaceae have a great resistance to the environment than coliforms also they can be colonized in an inadequate sanitation so they can be used be as indicators of sanitation (GMPs) (Kornacki and Johnson, 2001).

The high Enterobacteriaceae counts in food indicates improper processing/or recontamination due to cross contamination which occur through raw materials, contaminated equipment or unclean handling (Ikeme, 1990).

5. CONCULOSIONS

The obtained results in this study allow to know that boiled, grilled and fried chicken meat meals at hospital restaurant Presence of Enterobacteriaceae in the food is an indication of improper hygienic measures (Gill and Landers, 2004).

Regarding the epidemiological importance, as some of members Enterobacteriaceae are pathogenic and cause serious infections and food poisoning outbreaks to human. Furthermore, the Enterobacteriaceae count can be taken as indicator of possible enteric contamination in the absence of coliform organisms (Mosupye and Van Holy, 2000).

The result recorded in table (5) recorded that *coliform* count In chicken meat samples were nearly similar to those of El-Taher (2009)which was $(7.95 \times 10^2 \pm 1.59 \times 10^2)$ and Raphael et al.(2014) (8.85×10^2)but much lower than that of Saad et al.(2011) ($4.85 \times 10^3 \pm 0.77 \times 10^3$); Ibrahim et al.(2014) was($1.18 \times 10^3 \pm 0.26 \times 10^3$)and Abd El-Aal (2015)($1.06 \times 10^4 \pm 0.17 \times 10^4$) for boiled chicken meat and ($6.40 \times 10^3 \pm 1.23 \times 10^3$)for fried ones.

High *coliform* count indicated inferior quality of meat. The contamination with coliforms may occur during slaughtering, or dressing of carcasses, , shopping blocks, soiled hands or knives used for handling and cutting or contaminated water (Yadav et al., 2006).

The result recorded in table (7) recorded that staphylococci count in examined samples were lower than those obtained by El-Taher (2009) $(3.59 \times 10^3 \pm 0.76 \times 10^3)$ in chicken meat; Arab (2010) $(2.60 \times 10^4 \pm 1.05 \times 10^4)$ in chicken meat; Ibrahim et al. (2014) $(3.01 \times 10^3 \pm 0.26 \times 10^3)$ in cooked chicken meat and Abd El-Aal (2015) $(4.42 \times 10^3 \pm 0.75 \times 10^4)$ for boiled chicken meat and 2.10×10³± 0.32×10³ for fried chicken meat. Comparing to the safe permissible limits stipulated by Center for Food Safety (2014) for the total staphylococcal count (not exceed 10^4 cfu/g),

Higher count obtained by Farag (2009) found Staphylococcus count were5.9% more than permissible limit.

Presence of E. coli in meat indicates a general lack of cleanness during slaughtering, evisceration, dressing, transportation and handling of meat (ICMSF, 1996c). Moreover, the incidence of serologically identified E. coli as Enteropathogenic E. coli (E. coliO1:H7, E. coliO78 and E. coliO₁₁₄: H₂₁), Enterotoxigenic E. coli (E. coli O₁₂₅:H₁₈) Enterohaemorrhagic E. coli (E. coli O26:H11 and E. coli O₁₁₁:H₄) and Enteroinvasive E. coli (E. coli O₁₂₄). Nearly similar results were obtained by Ali (2011). Higher results obtained by EL-Abbasy (2010). These results came in accordance with those obtained by El-Taher (2009) 13.3%; Arab (2010) 6.67%; Marzano and Balzaretti (2011)and El Masry et al. (2015). The same serotypes of E.coli were previously isolated from chicken meat by Maarouf and Nassif (2008); Lamada-Hananet al. (2012)Windham et al. (2013) and Abd El-Salam(2014). These results coincided with the fact of Wilson et al., (1997) and Woody et al., (1998) who recorded that the same serogroups were Enteropathogenic E.coli and causing infantile enteritis; hemorrhagic colitis; hemorrhagic gastroenteritis and diarrheal illness in different settings.

The presence of *E. coli* used as an indicator for fecal pollution. Which occur due to improper slaughtering techniques, contaminated surfaces and/or handling of the meat by hands of infected person (Nelet al., 2004). Also, the contamination by E. coli can occur during the meat processing at slaughterhouse or due to the poor handling of the retailers of meat (Kagambèga et al., 2011).

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 and during processing (scalding) which consider big source of contamination. Also fried chicken meat meals less contaminated than boiled chick meat meals mainly due to that fried meat firstly boiled then fried so expose to high temperatures for long time which kill most food poisoning microorganism on the other hand my contaminated by some types of bacteria do not present in boiled one through cross contamination and bad personnel hygiene. This can be controlled by applying Hygienic measures during slaughtering, struggling should be considered. HACCP (Hazard Analysis and Critical Control Points) is a system of preventive control designed to improve the safety of the poultry product.

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