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Bacteriological profile of frozen chicken meat cuts at Qalubiya governorate markets

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ABSTRACT

The study was conducted on 100 random samples of frozen chicken meat cuts represented by chicken wings, drumstick, thigh and breast (25 of each) purchased from different markets at Qalubiya Governorate, Egypt. The collected samples were bacteriological examination to investigate the bacteriological quality. The obtained results cleared that the mean values of APC, psychrotrophs; coliforms and *S. aureus* counts (CFU/g) of the wing samples were the most contaminated with such bacterial groups followed by drumstick, thigh, and breast samples. Further, bacteriological isolation of some food poisoning bacteria revealed detection of *E. coli* and coagulase positive *S. aureus* (CoPSA) in 11, and 21% of the examined samples, respectively. On the other hand, Salmonella species could not be detected in any of the examined samples. Concerning detection and typing of some enterotoxigenic CoPSA, 5 isolates were randomly examined using SET- RPLA test and the results indicated detection of Staphylococcal enterotoxin A in 3 isolates (60%), while Staphylococcal enterotoxins C and D were detected in one isolate (20% of each). Therefore, the sources and public health significance as well as trials for control of such serious food poisoning bacteria were discussed.

1. INTRODUCTION

Chicken meat is a major component of the human healthy diet worldwide that is low in fat and cholesterol as compared to other meats as well as it is an excellent source of high-quality animal proteins, vitamins, and minerals (Liu *et al*., 2012). Unfortunately, chicken carcasses are excellent media for enhancing the proliferation of variable foodborne microorganisms, especially, Salmonella; *E. coli*; Campylobacter and *S. aureus* that considered as important causes of foodborne outbreaks in people (Bhaisare *et al*., 2014). Consumers are expecting the chicken meat to be fresh, properly chilled, and tender, with the typical texture of fresh meat, without drip or leakage from the muscle and without pathogenic microorganisms (Boerrigter-Eenling *et al*., 2017). In contrast, the commercial interests of chicken meat producers and meat markets are for longer shelf-life and prolonged storage without signs of spoilage or quality losses. For these reasons, manufacturers and retailers prefer chicken carcasses and chicken meat cuts that are frozen than such in chilled storage (Atanassova *et al*. 2018).

Although freezing is considered an excellent method for keeping quality of chicken meat for long period (9-12 months) at temperature below -18ºC, psychrotrophic bacteria can grow leading to many undesirable changes in the sensory characters of the food products (Atanassova *et al*. 2018).

The APC is considered as an index of food quality, which gives an idea about the hygienic measures during processing and help in assessing the keeping quality of such food item (Aberle *et al*., 2001). In addition, the coliform bacteria are reliable indicators of fecal pollution, improper handling and storage of meat and meat products (Paulsen *et al*., 2006). Meat-borne *E. coli*, Salmonellae and coagulase positive *S. aureus* have been recorded to be the most important bacterial food poisoning outbreaks worldwide (Bhaisare *et al*., 2014 and Noori and Alwan, 2016).

Avian strains of *E. coli* show many resemblance with human extra intestinal *E. coli* strains, in that most of the virulence genes they possess and they can infect consumers through eating the contaminated foods causing variety of diseases, including hematological, urinary, respiratory, neural, and circulatory affections (Johnson *et al.*, 2007). Moreover, their presence in poultry meat and its products indicates lack of proper sanitation and possible fecal contamination (Synge, 2000).

Salmonella is considered the most frequent foodborne pathogen worldwide (Capita *et al*., 2007). Most Salmonella serovars of poultry meat origin lost their host-specificity revealing it able to cause human food poisoning (Muth, 2009).

Staphylococcus aureus had been ranked in the third place as one of the most important foodborne diseases worldwide (Normanno *et al*., 2007). It is used as heat treatment sufficiency indicator, hygienic conditions during food processing, production and preparation (Malheiros *et al*., 2010). It secrets much types of staphylococcal enterotoxins (SEs) which demonstrated emetic activity (María *et al*., 2010). Staphylococcal enterotoxins are associated with Staphylococcal food poisoning characters such as fast sudden emergence of GIT disturbances lasting from 24 to 48h (Llewelyn and Cohen, 2002).Moreover, staphylococcal

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enterotoxin type A is the most common enterotoxin recovered from food poisoning outbreaks (María *et al*., 2010). As the hygienic level of frozen chicken meat cuts with variable foodborne microorganisms represent serious health impact to the consumers, the current study was performed to throw out light over the bacteriological profile and sanitary status of some frozen chicken meat cuts sold in Benha city, Qalubiya Governorate, Egypt.

2. MATERIAL AND METHODS

2.1. Collection of samples

One hundred random samples of frozen chicken meat cuts represented by wings, drumstick, thigh and breast (25 of each), weighed about 15g for wing samples and 100-250 g for the other samples, were purchased from different supermarkets at Qalubiya Governorate. The collected samples were subjected to the following bacteriological examinations after their thawing in the refrigerator overnight:

2.2. Bacteriological examination:

2.2.1. Preparation of samples was performed following APHA (2001)

2.2.2. Determination of APC (CFU/g), using pour plate technique following ISO (2013).

2.2.3. Enumeration of psychrotrophic bacteria using pour standard plate count agar following APHA (2001).

2.2.4. Enumeration of coliforms count using pour plate of tempered melted Violet Red Bile agar following ISO (2006).

2.2.5. Detection of pathogenic E. coli was conducted following ISO (2001)

Typical *E. coli* colonies (greenish-bluish colonies with bluish halo zone) on TBX agar after incubation at 37º for 24h were purified and isolated for morphological identification by Gram stain; biochemically according to Edward and Ewing (1972), and serologically according to Markey *et al*. (2013) as tabulated in table (1).

Table 1 Antisera used in serological identification of *E. coli*

Polyvalent Sera	Monovalent sera						
Polyvalent 1	O1	O26	O86a	O111	O119	O127a	O128
Polyvalent 2	O44	O55	O125	O126	O146	O166	
Polyvalent 3	O18	O114	O142	O151	O157	O158	
Polyvalent 4	O6	O27	O78	O148	O159	O168	
Polyvalent 5	O20	O25	O63	O153	O167		
Polyvalent 6	O8	O15	O115	O169			
Polyvalent 7	O28ac	O112ac	O124	O136	O144		
Polyvalent 8	O29	O143	O152	O164			

B.H-sera H2, H4, H6, H7, H11, H18 and H21.

2.2.6. Isolation and enumeration of S. aureus on Baird Parker (BP) agar according to FDA (2001) appeared as black, shiny colonies with halo zone around them were picked up for morphological examination and biochemical identification according to Markey *etal*. (2013).

2.2.7. Detection of Enterotoxins producing S. aureus isolates by Reversed Passive Latex agglutination kit (SET-RPLA) test according to Igarashi et al. (1986).

2.2.8. Detection of Salmonella spp. following the instructions of ISO (2017): red colonies with or without black centers on XLD agar were speculated as salmonella isolate and identified morphologically and biochemically according to Markey *etal*. (2013).

2.2.9. Statistical analysis

Data were analyzed using the descriptive statistic SPSS (Version 20). Differences in mean of analyzed data were considered significant at P≤0.05.

3. RESULTS

The recovered results in table (2) showed that the wing samples recorded the highest APC followed by drumstick, thigh and breast samples in which statistical results showed a significant (P≤0.05) differences of wing and drumstick samples when compared with thigh and breast samples, and a significant (P≤0.05) increase of thigh samples results when compared with breast meat samples. Meanwhile, there was no difference of APC readings between wing and drumstick meat samples.

Table 2 Statistical analysis of Aerobic plate counts (CFU/g) in the examined samples of frozen chicken meat cuts (n=25 of each).

Samples	Min.	Max.	Mean ±SE
Wings	3.9×10^5	1.15×10^6	$8.16\times10^5\pm0.37\times10^{5a}$
Drumstick	3.8×10^5	1.05×10^6	$7.85\times10^5\pm0.35\times10^{5a}$
Thigh	3.0×10^5	9.8×10^5	$6.76\times10^5\pm0.37\times10^{5b}$
Breast	1.5×10^5	9.3×10^5	$5.58\times10^5\pm0.43\times10^{5c}$

Different superscript litters (a, b, c) means significant difference of frozen chicken meat cut samples (P≤0.05).

Table (3) indicated that the mean values of psychrotrophic count in the examined wing and breast samples were 4.91×10^5 and 3.88×10^5 CFU/g; which proved that wings were the most contaminated samples, while breast samples were the lowest. Moreover, the statistical results showed that, wing and drumstick meat samples showed a significant (P≤0.05) increase of counts when compared with breast meat samples. However, there was no difference of psychrotrophic counts between breast meat samples and others.

Table 3 Statistical analysis of Psychrotrophic counts (CFU/g) in the examined samples of frozen chicken meat cuts (n=25 of each).

Samples	Min.	Max.	Mean ±SE
Wings	2.3×10^5	7.2×10^5	$4.91\times10^5\pm0.25\times10^{5a}$
Drumstick	2.1×10^5	6.9×10^5	$4.74\times10^5\pm0.25\times10^{5a}$
Thigh	2.0×10^5	6.7×10^5	$4.41\times10^5\pm0.24\times10^{5ab}$
Breast	1.1×10^5	6.6×10^5	$3.88\times10^5\pm0.30\times10^{5b}$

Different superscript litters (a, b, c) means significant difference of frozen chicken meat cut samples (P≤0.05).

However, all the examined samples were contaminated with APC and psychotrophic microorganisms, the counts were considered within the safe permissible limits stipulated by EOS (1090/2005) (as all examined samples did not exceeded 10^5 CFU/g so all samples were accepted).

Regarding to the coliforms count. Table (4) showed that the coliform counts were detected in 72% of the examined wing and drumstick samples, while it was detected in 60% of both examined breast and thigh samples.

Table 4 Statistical analysis of Coliform counts (CFU/g) in the examined samples of frozen chicken meat cuts (n=25 of each)

Samples	Positive		Min.	Max.	Mean ±SE	MPL	No. of accepted samples
	No.	%*					
Wings	18	72	$< 1\times10^2$	4.3×10^3	$2.66\times10^3\pm0.29\times10^{3a}$	$\leq 10^2$	7 (28%)
Drumstick	18	72	$< 1\times10^2$	4.0×10^3	$2.12\times10^3\pm0.23\times10^{3b}$	$\leq 10^2$	7 (28%)
Thigh	15	60	$< 1\times10^2$	3.3×10^3	$2.01\times10^3\pm0.18\times10^{3b}$	$\leq 10^2$	10 (40%)
Breast	15	60	$< 1\times10^2$	2.9×10^3	$1.84\times10^3\pm0.16\times10^{3b}$	$\leq 10^2$	10 (40%)

* Percentage in relation to total number of sample in each row (25). Different superscript litters (a, b) means significant difference of frozen chicken meat cut samples (P≤0.05). MPL: Maximum permissible limit according to Egyptian Organization for standardization (EOS, 1090/ 2005).

According to the listed mean counts, the wing samples had the highest contamination followed by drumstick, thigh and breast samples, respectively. In addition, the statistical findings showed that wing meat samples revealed significant (P≤0.05) increase of total coliform counts when compared with breast meat samples. Meanwhile, there were no difference of coliform counts between drumstick and thigh when compared with breast meat samples. Moreover, 34 examined frozen chicken meat cuts samples were contaminated and the counts were higher than the safe permissible limits stipulated by EOS (1090/2005) for coliform count (not exceed 10^2 CFU/g) so, they were unaccepted.

Referring to the incidence of *E. coli* and serotyping, Table (5 and 6) showed that out of 100 examined samples, 11 *E. coli* strains were isolated where wing samples were the highest contamination. Serotyping of the isolates revealed detection of O_{55}:H_7, O_{111}:H_2, O_{125}:H_{21} and O_{146}:H_{21} with different prevalence in the examined samples. Referring to EOS (1090-2005) legislations, these 11 contaminated samples were unaccepted for human consumption.

Table 5 Incidence of *E. coli* detected in the examined samples of frozen chicken meat cuts (n=25)

Sample	Positive		No. of accepted samples**	No. of non-accepted samples**
	No.	%*		
Wings	4	16.0	21	4 (16%)
Drumstick	3	12.0	22	3 (12%)
Thigh	2	8.0	23	2 (8%)
Breast	2	8.0	23	2 (8%)
Total (100)	11	11.0	89	11 (11%)

* Percentage in relation to total number of sample in each row.

Table 6 Incidence and serotyping of *E. coli* isolated from positive samples of frozen chicken meat cuts (n=25 of each)

E. coli serotype	Wings		Drumstick		Thigh		Breast		Strain characteristic
	n	%*	n	%*	n	%*	n	%*	
O_{55}:H_7	2	8	1	4	0	0	1	4	EPEC
O_{111}:H_2	0	0	1	4	1	4	0	0	EHEC
O_{125}:H_{21}	1	4	0	0	1	4	1	4	ETEC
O_{146}:H_{21}	1	4	1	4	0	0	0	0	EPEC
Total	4	16	3	12	2	8	2	8	-

* % was calculated in relation to total number of each sample (25). EPEC: Enteropathogenic *E. coli.* ETEC: Enterotoxigenic *E. coli.* EHEC: Enterohaemorrhagic *E. coli*

The recorded results in Table (7 and 8) revealed detection of *S. aureus* in 28, 24, 16 and 16% of the examined wing, drumstick, thigh and breast, respectively. The mean counts (CFU/g) proved that wing samples were the most contaminated samples. Referring to EOS (1090/2005) legislations, out of 100 examined samples, 21 samples were rejected due to contamination with coagulase positive *S. aureus*.

The results of SET-RPLA test in Table (9) cleared that out of eight randomly examined *S. aureus* isolates, five *S. aureus* (62.5%) strains were enterotoxigenic and classified according to type of toxin into (3A; 1C and 1D).

It is of great importance to mention that Salmonella species could not be detected in any of the examined samples.

Table 7 Incidence and counts of *Staphylococcus aureus* in examined frozen chicken meat cuts samples (n=25 of each)

Samples	Positive		Min.	Max.	Mean ±SE
	No.	%*			
Wings	7	28	$< 1\times10^2$	1.9×10^3	$1.20\times10^3\pm0.18\times10^{3a}$
Drumstick	6	24	$< 1\times10^2$	1.3×10^3	$1.00\times10^3\pm 0.10\times10^{3a}$
Thigh	4	16	$< 1\times10^2$	1.2×10^3	$0.90\times10^3\pm0.13\times10^{3a}$
Breast	4	16	$< 1\times10^2$	1.0×10^3	$0.78\times10^3\pm0.16\times10^{3a}$

* Percentage in relation to total number of sample in each row (25). (a) statistical analysis of means (ANOVA) revealed no significant differences between the examined samples.

Table 8 Incidence of Coagulase Positive *Staphylococcus aureus* in examined samples of frozen chicken meat cuts (n=25 for each sample)

Samples	Positive		No. of accepted samples**	No. of non-accepted samples**
	No.	%*		
Wings	7	28	18	7
Drumstick	6	24	19	6
Thigh	4	16	21	4
Breast	4	16	21	4
Total (100)	21	21.0	79	21

* Percentage in relation to total number of sample in each row. **Accepted and non-accepted samples according to the isolation of Coagulase Positive *S. aureus* (EOS, 1090 / 2010) (free).

Table 9 Incidence of enterotoxins production from isolated *Staphylococcus aureus*

No. of *S. aureus*	Enterotoxigenic strains		Type of enterotoxin		
	NO.	%	A	C	D
8	5	62.5	3	1	1

4. DISCUSSION

The mean values of APC in Table (1) were nearly similar to those recorded by Javadi and Safarmashaei (2011) ($3.53x10^5$ for thigh and $6.71x10^5$ CFU/g for breast samples, respectively). Meanwhile, they were higher than those reported by Daoud et al. (2014) ($2.1x10^3$ and $2.7x10^3$ CFU/g for breast and thigh, respectively) and Hassanen *et al.* (2017a) (7.47x104, 6.51x104 and 6.13x104 CFU/g in drumstick, thigh and breast samples, respectively), but lower than recorded by Mohamed (2016) (4.38×10^6 for thigh, 3.78×10^6 for breast and 4.0×10^6 CFU/g for wings).

Although there were significant differences among the examinedsamples, all the examined samples were accepted referring to the permissible limits of EOS (1090 /2005) ($<10^5$ CFU/g) which may be attributed to good poultry manufacturing processing and efficient freezing process that inhibits bacterial growth.

The total psychrotrophic counts provides useful information about the keeping quality of chicken meat cuts, as their counts indicated the sanitation level adopted during all stages of manufacturing, transportation, storage and retailing (Jay *et al*., 2005). It is evident from the result recorded in Table (2) that, the total psychrotrophic counts infrozen chicken meat cut samples came in harmony with those

obtained by Al-Hamadany (2009) (1.06×10^6 CFU/g in the examined frozen chicken meat cuts samples), but disagree with those reported by Mohamed (2016) who recorded higher counts (4.59×10^6 for thigh and 5.71×10^6 CFU/g for breastfrozen samples). The recovered Psychrotrophic counts were little lower than total aerobic bacterial count and all examined samples were accepted, this could be due to good manufacturing practices which has a role in decreasing the count (Mead, 2000) or the samples were bought fresh and the products did not stay for long periods in shops and markets and so the psychrotrophic bacteria did not have enough time to increase(Jay *et al*., 2005).

The obtained coliform counts in Table (3) came in parallel with those reported by Mohamed (2016) (2.61×10^3 and 2.07×10^3 CFU/g for thigh and breast samples, respectively). Meanwhile, they disagreed with those reported by Daoud et al. (2012) who recorded lower counts ($6.9x10^2$ and 6.4×10 CFU/g for thigh and breast samples, respectively); and with those of Marwan-Heba (2016) who recorded higher counts ($5.56\times10^5$and 1.35×10^4 CFU/g, respectively). Detection of pathogenic *E. coli* in meat samples hypothesis contamination with gastric content and implies processing faults evisceration.

The results of *E. coli* isolation (Table, 4) agreed with those of Abd El-Alim (2017) who detected *E. coli* in 7.3% of his examined frozen chicken meat samples, but lower than those of Marwan-Heba (2016) (26.9%). Moreover, the serogroups obtained in Table (5) were detected in frozen meat samples by Marwan-Heba (2016); Mohamed (2016) and Abd El-Alim (2017). These complied with the reported results of Son et al. (2014) who recorded that the same serovars were enteropathogenic *E. coli* and causing puerile enteritis; hemorrhagic colitis, HUS, hemorrhagic gastroenteritis and profuse diarrheal disorders. The results of *S. aureus* counts (Table, 6) came in accordance with the reported results of Mahmoud and Hamouda-Seham (2006) who reported that, the mean value of *S. aureus* counts in chicken meat samples were 8.9×10^3 CFU/g. Meanwhile, they were disagreed with those recorded by Mohamed (2016) who recorded higher results (1.14×10^4 and 1.12×10^4 CFU/g in thigh and breast samples, respectively); and with those of Al-Dughaym and Al-Tabari (2009) who recorded lower counts ($<10^2$ CFU/g). Moreover, isolation rates of coagulase positive *S. aureus* (Table, 7) were nearly similar to the results of Marwan-Heba (2016) and Abd El-Alim (2017) that isolated *S. aureus* from frozen chicken meat with incidences of 26.7% and 20.7%, respectively. Meanwhile, they were disagreed with those of Abdalrahman *et al.* (2015) and Afifi-Dina (2016) who reported higher incidences for *S. aureus* isolation (53.8% and34.3%, respectively); and with those of Al-Hamadany (2009) who failed to detect *S. aureus* in frozen chicken meat cuts samples. The results of SET-RPLA test (Table, 8) were incompliance with those recorded by Abdalrahman *et al*. (2015); Afifi-Dina (2016) and Hassanen *et al*. (2017b). The occurrence of *S. aureus* in frozen chicken meat cuts could be due to poor personal hygiene of workers and the technique used in eviscerating the chicken carcasses during processing. Detection of *S. aureus* in poultry meat and its products throw light on poor personal hygiene as well as scarcity sterilization of the used equipment. *Staphylococcus aureus* can grow with keeping both acceptable odor and taste of food products while secreting heat resistant enterotoxins leading to food intoxication with rapid onset of symptoms within 3-8h post-ingestion as nausea, vomiting, abdominal cramps severe diarrhea and gastroenteritis among consumers (Zogg *et al*., 2016).

Results of Salmonella detection came in harmony with those reported by Al-Hamadany (2009), Mohamed (2016), Marwan-Heba (2016) and Abd El-Alim (2017) who failed to detect salmonella serovars from frozen chicken meat cuts samples; meanwhiledisagreed with the results recorded by Noori and Alwan (2016) who isolated salmonella serovars from frozen chicken meat samples. Moreover, the obtained results indicated good evisceration process at the slaughterhouse and good handling to the chicken carcasses.

5. CONCULOSION

Results of the hygienic profile of the examined samples showed that frozen wing samples were the most contaminated samples followed by drumstick, thigh and breast samples. in addition, it proved that the frozen chicken meat cuts are considered of public health hazard and the presence of aerobic bacteria, psychrotrophic bacteria, coliforms mainly pathogenic *E. coli* and enterotoxigenic coagulase positive *S. aureus* with relatively high rates might be due to insufficient sanitation and disinfection procedures of equipment and surfaces; or poor ill-knowledge personal hygiene that leading to frequent thawing and freezing of chicken meat resulting in an inferior or even unfit quality for human consumption. Therefore, it was concluded that these pathogens are meat borne pathogens of public health important.

6. REFERENCES

1. Abd El-Alim, H.A. 2017.Bacteriological studies on some bacteria isolated from flesh of imported and local frozen poultry. Thesis, Master of Veterinary Medicine (Bacteriology, Immunology and Mycology), Benha Univ., Egypt.
2. Abdalrahman, L.S., Stanley, A., Wells, H., Fakhr, M.K. 2015. Isolation, virulence, and antimicrobial resistance of methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin sensitive *Staphylococcus aureus* (MSSA) strains from Oklahoma retail poultry meats.Int. J. Environ. Res. Public Health, 12, 6148-6161.
3. Aberle, E.D., Forrest, J., Gerrard, D.E., Mills, E.W. 2001.Principles of meat science.(4th edition).Hunt Puplishing Co., Kendall, USA.
4. Afifi-Dina, H.M. 2016. Characterization of methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from food products of poultry origin in Egypt. Thesis, Master of Veterinary Medicine (Bacteriology, Immunology and Mycology), Benha Univ., Egypt.
5. Al-Dughaym, A.M. and Al-Tabari, G.F. 2009.Safety and quality of some chicken meat products in Al-Ahsa markets-Saudi Arabia. Saudi J. Biological Sciences 17, 37–42.
6. Al-Hamadany, M.S. 2009.Studying certain quality properties of imported frozen chicken thighs.Thesis, Master of Science, College of Education for Women, University of Baghdad.
7. APHA "American Public Health Association" 2001.Compendium of methods for the microbiological examination of Foods.4th Ed. F.P. Downes and K. Ito (editors), APHA. Washington D.C., USA.
8. Atanassova, S., Stoyanchev, T., Yorgov, D., Nachev, V. 2018.Differentiation of fresh and frozen-thawed poultry breast meat by near infrared spectroscopy.Bulgarian J. Agricultural Science 24, 162-168.
9. Bhaisare, D.B., Richard, T.D., Punniamurthy, C.N. 2014. Bacterial pathogens in chicken meat: Review. Int. J Life Sci Res 2(3), 1-7.
10. Boerrigter-Eenling, R., Alewijn, M., Weesepoel, Y., Van Ruth, S. 2017. New approaches towards discrimination of fresh/chilled and frozen/thawed chicken breasts by HADH

activity determination: Customized slope fitting and chemometrics. Meat Science 126, 43-49.
11. Capita, R., Alonso-Calleja, C., Prieto, M. 2007. Prevalence of *Salmonella enterica* serovars and genovars from chicken carcasses in slaughterhouses in Spain. J Appl Food Microbiol 103, 1366–1375.
12. Daoud, J.R., Farghaly, R.M., Maky, M. 2014. Microbial quality of frozen chicken meat at grocery stores in Qena city.International Conference and Exhibition on Food Processing & Technology, November 22-24, S S Hundal, J Clin Toxicol, 4:4:74
13. Edward, R.P. and Ewing, W.H. 1972. Edwards and Ewing's identification of Enterobacteriaceae, 3rd Ed. Burgess, Minneapolis.
14. EOS "Egyptian Organization for Standardization" No.1090, 2005. Egyptian Organization for Standardization and quality control. Egyptian Standards for frozen poultry and rabbit.
15. FDA "Food and Drug Administration" 2001.Evaluation and definition of potentially hazardous foods. Analysis of microbial hazards related to time/ temperature control of food for safety. Department of Health and Human Services. Food and Drug Administration Chapter 4:1-19.
16. Hassanen, F.S., Shaltout, F.A., Amani, M.S., Maarouf, A.A., Rasha, N.A. 2017[a]. Studies on bacteriological profile of chicken meat cuts in Kaliobia Governorate. Benha Vet Med J 33(2), 402-409.
17. Hassanen, F.S., Shaltout, F.A., Amani, M.S., Maarouf, A.A.A., Rasha, N.A. 2017[b]. Detection of virulence genes of enterotoxigenic *Staphylococcus aureus* isolated from chicken meat cuts by using PCR. Benha Vet Med J 33(2), 410-417.
18. Igarashi, H., Fujikawa, H., Shingaki, M., Bergdoll, M.S. 1986. Latex agglutination test for Staphylococcus toxic shock syndrome toxin1" J. Clinic. Microbial 23, 516-521.
19. ISO "International Organization of Standardization" 2001.International Organization for Standardization.No. 16649. Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of -glucuronidase-positive *Escherichia coli*.
20. ISO "International Organization of Standardization" 2006. No.4832-E. Microbiology of food and animal feeding stuffs – Horizontal methods for detection and enumeration of coliforms part, 3: colony count technique.
21. ISO "International Organization of Standardization" 2013.International Organization for Standardization.No. 4833-1. Microbiology of the food chain-Horizontal method for enumeration of microorganisms-Part 1: Colony count at 30^{O}C by the pour plate technique.
22. ISO "International Organization of Standardization" 2017.International Organization for Standardization.No. 6579-1.Microbiology of the food chain-Horizontal method for detection of Salmonella -Part 1.
23. Javadi, A. and Safarmashaei, S. 2011. Microbial profile of marketed broiler meat. Middle-East J Sci Res 9 (5), 652-656.
24. Jay, J.M., Loessner, M.J., Golden, D.A. 2005. Modern Food Microbiology. 7th Ed., Springer Science, Business Media Inc.
25. Johnson, T.J., Kariyawasam, S., Wannemuehler, Y., Mangiamele, P., Johnson, S.J., Doetkott, C. 2007. The genome sequence of avian pathogenic *Escherichia coli* strain O_1:K_1:H_7 shares strong similarities with human ExPEC genomes. J Bacteriol 189, 3228-3628.
26. Liu, X.D., Jayasena, D.D., Jung, Y., Jung, S., Kang, B.S., Heo, K.N., Lee, J.H., Jo, C. 2012. Differential proteome analysis of breast and thigh muscles between Korean native chickens and commercial broilers. Asian Australas J Anim Sci 25,895–902.
27. Llewelyn, M. and Cohen, J. 2002. Superantigens: Microbial agents that corrupt immunity. Lancet Infect Dis 2, 156-162.
28. Mahmoud, Y.E.L. and Hamouda-Seham, N. 2006. Quality evaluation of poultry meat carcasses in El-Gharbia Governorate markets. Assiut Vet J 52(110), 31-38.
29. Malheiros, P.S., Passos, C.T., Casarin, L.S., Serraglio, L., Tondo, E.C. 2010. Evaluation of growth and transfer of *Staphylococcus aureus* from poultry meat to surfaces of stainless steel and polyethylene and their disinfection. Food Control, 21, 298-301.
30. María, Á.A., María, C.M., María, R.R. 2010. Food poisoning and *Staphylococcus aureus* enterotoxins. Toxins (Basel) 2(7), 1751–1773.
31. Markey, B.K., Leonard, F.C., Archambault, M., Cullinane, A., Maguire, D. 2013.Clinical Veterinary Microbiology. 2nd Ed., MOSBY. Elsevier Ltd., Edinburgh, London, New York, Oxford, Philadelphia, St. Louis, Sydney, Toronto.
32. Marwan-Heba, A.E.I. 2016.Sanitary status of meat meals at hospital level in Kaliobia Governorate.Thesis, Master of Veterinary Medicine (Meat Hygiene), Benha University, Egypt.
33. Mead, G.C. 2000. Fresh and further processed poultry in: Microbiological safety and quality of food. Lund, B.M. (Ed.), Aspen Pub., 445-471.
34. Mohamed, S.E.R. 2016. Frozen chicken meat quality in governmental hospital. Thesis, Master of Veterinary Medicine (Meat Hygiene), Benha Univ., Egypt.
35. Muth, M.K. 2009. Analysis of salmonella control performance in U.S young chicken slaughter and pork slaughter establishments, J. Food Protect., 72 (1), 6-13.
36. Noori, T.E. and Alwan, M.J. 2016.Isolation and identification of zoonotic bacteria from poultry meat. Int J Adv Res Biol Sci 3(8), 57-66.
37. Normanno, G., La Salandra, G., Dambrosio, A., Quaglia, N.C., Corrente, M., Parisi, A., Santagada, G.,Firin, U.A., Crisetti, E., Celano, G.V. 2007. Occurrence, characterization and antimicrobial resistance of enterotoxigenic *Staphylococcus aureus* isolated from meat and dairy products. Int J Food Microbiol 115, 290-296.
38. Paulsen, P., Schopf, E., Smulders, F.J.M. 2006. Enumeration of total aerobic bacteria and *E. coli* in minced meat and on carcass surface samples with an automated most-probable-number method compared with colony count protocols. J Food Protect 69(10), 2500-2503.
39. Son, I., Binet, R., Maounounen-Laasri, A., Lin, A., Hammack, T.S., Julie A. and Kase, J.A. 2014.Detection of five Shiga toxin-producing *Escherichia coli* genes with multiplex PCR". J Food Microbiol 40, 31-40.
40. SPSS for windows, Version: 11 (19 September, 2001). Copyright SPSS Inc. 1989 - 2001. All rights reserved.
41. Synge, B.A. 2000. Verotoxin producing *E. coli*: veterinary view. J applied Microbiol Symposium Suppl 88, 315-375.
42. Zogg, L., Zurfluh, K., Inderbinen, M.N., Stephan, R. 2016. Institute for food safety and hygiene, Vetsuisse Faculty, University of Zurich, Switzerland, Characteristics of ESBL-producing Enterobacteriaceae and Methicillin resistant *Staphylococcus aureus* (MRSA) isolated from Swiss and imported raw poultry meat collected at retail level. J. SAT ASMV, 158(6), 451-456.