



Assessment of Microbiological Quality of Imported Broiler Chicken Carcasses Retailed for Sale in Al Beida City, Libya

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ABSTRACT

A total of 100 random samples of imported frozen broiler chicken carcasses including; breast and thigh (50 samples / each) were collected from supermarkets at Al Beida City, Libya to assess their quality through microbiological analysis. It was recorded that mean value of APC for breast samples was 7.6×10^5 CFU/g and 4.9×10^6 CFU/g for thigh samples. Also, it was recorded that mean value of Staphylococci count for breast samples was 6.3×10^2 CFU/g and 2.5×10^3 CFU/g for thigh samples. In addition, it was recorded that mean value of EC for breast samples was 1.6×10^5 CFU/g and 3.6×10^5 CFU/g for thigh samples while mean value of CC for breast samples was 6.4×10^4 CFU/g and 3.7×10^5 CFU/g for thigh samples. It was clear that mean value of EC and CC of thigh samples was higher than that of breast samples. Finally, mean value of Mold count for breast samples was 3.6×10^2 CFU/g and 6.2×10^2 CFU/g for thigh samples while mean value of Yeasts count for breast samples was 1.9×10^3 CFU/g and 8.9×10^3 CFU/g for thigh samples. Detection of potential pathogenic bacteria revealed that the incidence of *E. coli* was higher in the examined samples of breast (54%) compared to that of thigh (48%) while the incidence of *Salmonella* was in the examined samples of breast and thigh was 9% for each. The obtained results clarified that imported chicken carcasses had higher bacterial counts and significant potential pathogenic bacteria that may be attributed to cross contamination and unsanitary personal hygiene during handling, packaging, storage and distribution.

Keywords: Microbiological, Quality, Imported, Chicken, Carcasses

1. Introduction

Chicken meat is one of the most popular foods amongst advanced and developing countries due to its role in solving the problem of animal meat shortage especially in the last decade. Poultry meat is considered an excellent source of high quality, easily digested animal protein which is of a high biological value and contains most of essential amino acids beside many minerals and vitamins which are necessary for maintaining life and promoting growth. The importation of food of animal origin as frozen meat was increased in Egypt especially from India and Brazil (FAO, 2009). Inspection of frozen meat plays an important role in controlling of number of diseases of public health importance. Theoretically, when chicken is frozen correctly and maintained at the optimal freezing temperature, it should stay good forever. However, it is hard to guarantee that the chicken will remain safe and of good quality for eating but once frozen chicken is thawed the microbes come back to life and continue their life's work, which is to multiply and consume (Wallace, 2003).

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Unfortunately, Chicken has higher pathogenic and spoilage bacterial counts than most other foods. The microflora in raw chicken carcasses is very heterogeneous, and it may already be present at the time of slaughter, introduced by the workers' handling and the cutting tools, or by water and air during dressing, evisceration, cutting, and packing. Evaluation of microbial hazards and their indicators would help to provide hygienic chicken meat for consumers.

Aerobic plate count is an important factor for evaluation of microbial quality assessment in food products and is an indicator of the overall degree of microbial contamination of foods (ICMSF, 1996). Presence of coliform in ready to eat food indicates that there are degrees of ignorance of the handlers to the proper hygienic practices (Lues et al., 2006).

E. coli is a normal inhabitant of the intestinal tract of humans and worm-blooded animals and meat is a common source of *E. coli* contamination, which may be acquired during slaughter through fecal contact besides some pathogenic strains are responsible for enteric and diarrheal diseases, and they have been increasingly recognized as the most important causes of food borne diseases and outbreak all over the world (Bettelheim and Goldwater, 2014).

Salmonella is a member of the Enterobacteriaceae, Gram negative, motile, with peritrichous flagella and non-spore forming rods. Also, *Salmonella* is a facultative anaerobic (can grow with or without oxygen) catalase positive and oxidase negative bacteria. However, *Salmonella* is not included in the group of organisms referred to as coliforms (Lawley et al., 2008). More than 2,500 different types of *Salmonella* exist, some of which cause illness in both animals and people. Some types cause illness in animals but not in people. Some serotypes are only present in certain parts of the world (Brands, 2006).

Therefore, great emphasis is being placed on the microbiological aspects of poultry carcasses and on searching for alternative mechanisms to reduce both natural and cross contamination, thus avoiding major public health problems so it is important to adopt hazard analysis and critical control point principles in production, processing and handling of poultry carcasses to achieve pathogen free products. So, the present work was carried out to evaluate the microbial fitness of imported chicken carcasses retailed for sale in Al Beida City, Libya beside isolation and identification of some potential pathogenic bacteria of public health significance.

2. Material and methods

2.1. Collection of samples:

A total of 100 random samples of imported frozen broiler chicken carcasses including; breast and thigh (50 samples / each) were collected from supermarkets at Al Beida City, Libya. Each sample (represented by one carcass) was kept in a separate plastic bag and transferred directly with a minimum of delay to the laboratory of Preventive Medicine and Public Health Department, Faculty of Veterinary Medicine, Omar Al- Mukhtar University in an insulating refrigerated container under complete aseptic condition to avoid any changes in the quality of the sample.

2.2. Microbiological evaluation of imported chicken carcasses:

1. Preparation of samples was performed according to APHA, (2001).
2. Determination of aerobic plate count (APC) was carried out according to ISO 4833: (2003).
3. Determination of Staphylococci count was carried out according to ICMSF, (2002).

4. Determination of Enterobacteriaceae count (EC) was carried out according to ISO 4833: (2003).
 5. Determination of Coliforms counts (CC) was carried out according to ISO 4833: (2003).
 6. Determination of molds and yeasts counts was carried out according to ISO 21527-1: (2008).
 7. Isolation and Identification of E. coli ICMSF, (1996).
 8. Isolation and Identification of Salmonellae according to modified ISO. 6579: (2002).
- 2.3. Statistical analysis was carried out according to SAS, (2014).

3. Results

Table (1): Statistical analytical results of Aerobic Plate and Staphylococci counts (CFU/g) of imported chicken carcasses

Chicken carcasses (n=50/each)	APC				Staphylococci Count			
	Min	Max	Mean	±SEM	Min	Max	Mean	±SEM
Breast	2.9×10 ⁴	1.7×10 ⁶	7.6×10 ^{5a}	7.2×10 ⁴	1.7×10 ²	4.6×10 ³	6.3×10 ^{2a}	2.6×10 ²
Thigh	3.2×10 ⁴	4.1×10 ⁷	4.9×10 ^{6b}	9.2×10 ⁵	2.6×10 ²	4.9×10 ³	2.5×10 ^{0b}	7.4×10 ²

Means with similar letters are not significantly different at P ≤0.05. Aerobic plate count must not exceed 105 CFU/g. Chicken carcasses must be free from Staphylococci.

Table (2): Statistical analytical results of Enterobacteriaceae and Coliforms Counts (CFU/g) of imported chicken carcasses

Chicken carcasses (n=50/each)	Enterobacteriaceae count				Coliforms count			
	Min	Max	Mean	±SEM	Min	Max	Mean	±SEM
Breast	2.6×10 ⁴	4.2×10 ⁵	1.6×10 ^{5a}	6.2×10 ⁴	1.3×10 ³	2.9×10 ⁵	6.4×10 ^{0a}	2.2×10 ⁴
Thigh	3.4×10 ⁴	5.3×10 ⁵	3.6×10 ^{5b}	8.2×10 ⁴	3.7×10 ³	5.3×10 ⁵	3.7×10 ^{0b}	4.6×10 ⁴

Means with similar letters are not significantly different at P ≤0.05. There is no permissible limit for Enterobacteriaceae. Coliforms Count for chicken carcasses must not exceed 102 CFU/g

Table (3) Statistical analytical results of Mold and Yeast counts (CFU/g) of imported chicken carcasses

Chicken carcasses (n=50/each)	Mould count				Yeast count			
	Min	Max	Mean	±SEM	Min	Max	Mean	±SEM
Breast	2×10	1.4×10 ³	3.6×10 ^{2a}	8×10	5×10	5.3×10 ³	1.9×10 ^{3a}	1.3×10 ²
Thigh	7×10	5.1×10 ³	6.2×10 ^{2b}	2.1×10 ²	1.6×10 ²	3.1×10 ⁴	8.9×10 ^{0b}	1.9×10 ²

Chicken carcasses must be free from mold and yeasts. Means with similar letters are not significantly different at P ≤0.05.

Table (4): Incidence of Enteropathogenic E. coli isolated from imported chicken carcasses

Chicken carcasses E. coli serotypes	Breast (n=50)		Thigh (n=50)		Total (n=100)	
	No.	%	No.	%	No.	%
O ₁₁₁ :H ₂ (EHEC)	6	12.0	5	10.0	11	11.0
O ₉₁ :H ₂₁ (EPEC)	4	8.0	4	8.0	8	8.0
O ₁₂₇ :H ₆ (ETEC)	5	10.0	6	12.0	11	11.0
O ₁₁₃ :H ₄ (EPEC)	8	16.0	3	6.0	11	11.0
O ₁₂₄ (EIEC)	4	8.0	6	12.0	10	10.0
Total	27	54.0	24	48.0	51	51.0

Table (5): Incidence of Salmonella serotypes isolated from imported chicken carcasses

Chicken carcasses Salmonella serotypes	Breast (n=50)		Thigh (n=50)		Total (n=100)	
	No.	%	No.	%	No.	%
S. Enteritidis	3	6.0	4	8.0	7	7.0
S. Typhimurium	4	8.0	3	6.0	7	7.0
S. Kentucky	2	4.0	2	4.0	4	4.0
Total	9	18.0	9	18.0	18	18.0

4. Discussion

Poultry meat are subjected to the risk of contamination of various pathogens from different sources, primary during preprocessing and processing steps and secondary after processing through packaging, marketing and storage. Such contamination may render these food articles unfit for human consumption or even harmful to consumers. The Microbiological examination of chicken meat is used to determine conformance to the chicken meat specification (Microbiological criteria) is often used for testing conformance to provide only limited production to consumer against food poisoning and/or foodborne disease.

4.1. Aerobic Plate Count (APC)

Higher mean values of APC in frozen chicken carcasses may be attributed to contamination accompanying handling, transportation and retailing in the markets in insufficient chilling temperature. Also, it may be traced back to the habit of most supermarkets in retailing frozen chicken in semi chilled state and exposed them to repeated freezing and thawing throughout the day.

The recorded data in Table (1) clarified that APC for breast samples ranged from 2.9×10⁴ to 1.7×10⁶ CFU/g with mean value of 7.6×10⁵ CFU/g while APC for thigh samples ranged from 3.2×10⁴ to 4.1×10⁷ CFU/g with mean value of 4.9×10⁶ CFU/g. It was clear that mean value of APC of thigh samples was higher than that of breast samples. This finding was similar to that recorded by Habib, (2017) who found that APC for frozen carcasses ranged from 2.3×10⁴ to 1.3×10⁷ CFU/g with mean value of 3.6×10⁶ CFU/g.

Also, as shown in Table (1), it was observed that staphylococci count for breast samples ranged from 1.7×10² to 4.6×10³ CFU/g with mean value of 6.3×10² CFU/g while for thigh samples ranged from 2.6×10² to 4.9×10³ CFU/g with mean value of 2.5×10³ CFU/g. It was clear that mean value of Staphylococci count of thigh samples was higher than that of breast samples. In addition, it was recorded that all of the examined samples of chicken carcasses had Staphylococci count exceeding the permissible limit. Nearly similar results were recorded by Habib, (2017) who found that Staphylococci Count for frozen carcasses ranged from 1.2×10² to 4.1×10³ CFU/g with mean value of 8.9×10² CFU/g. Staphylococci are commonly found on the skin and in upper respiratory tract of man and animals and can easily contaminate the carcass, the presence of staphylococci in chicken carcass may be due to contaminated equipment and worker's hands with abrasion and wounds.

The recorded data in Table (2) clarified that EC for breast samples ranged from 2.6×10⁴ to 4.2×10⁵ CFU/g with mean value of 1.6×10⁵ CFU/g while EC for thigh samples ranged from 3.4×10⁴ to 5.3×10⁵ CFU/g with mean value of 3.6×10⁵ CFU/g. It was clear that mean value of EC of thigh samples was higher than that of breast samples. This finding was similar to that recorded by Habib, (2017) who found that EC for frozen carcasses ranged from 2.9×10⁴ to 4.1×10⁵ CFU/g with mean value of 2.2×10⁵ CFU/g.

The presences of Enterobacteriaceae in chicken carcasses indicate a microbiological proliferation which can allow multiplication of wide range of pathogenic and toxigenic microorganisms constituting public health hazard. Consequently, the Enterobacteriaceae count could be applied to monitor the hygienic level during handling of chicken carcasses.

The recorded data in Table (2) also clarified that CC for breast samples ranged from 1.3×10³ to 2.9×10⁵ CFU/g with mean value of 6.4×10⁴ CFU/g while CC for thigh samples ranged from 3.7×10³ to 5.3×10⁵ CFU/g with mean value of 3.7×10⁵ CFU/g. Moreover, it was clear that mean value of CC of thigh samples was higher than that of breast samples. In addition, it was found that all of the examined samples of

chicken carcasses had CC exceeding the permissible limit. This finding was similar to that recorded by Hassan, (2015) who noticed that the mean value of CC in frozen chicken carcasses was 1.4×10^2 CFU/g and Habib, (2017) who found that CC for frozen carcasses ranged from 1.8×10^3 to 3.6×10^5 CFU/g with mean value of 1.9×10^5 CFU/g.

Presence of coliforms in the broiler chicken carcasses may be attributed to the unsanitary conditions during different stages of processing (from slaughtering till final product) as they are indicator of fecal pollution either from workers and/or poultry. Efforts should be directed towards thoroughly cleaning and sanitizing of all equipment come in contact with poultry and workers and hygienic measures should be adopted during different steps of dressing (Neculita et al., 2007).

The recorded data in Table (3) clarified that Mould count for breast samples ranged from 2×10 to 1.4×10^3 CFU/g with mean value of 3.6×10^2 CFU/g while for thigh samples ranged from 7×10 to 5.1×10^3 CFU/g with mean value of 6.2×10^2 CFU/g. It was clear that mean value of Mould count of thigh samples was higher than that of breast samples. In addition, it was recorded that all of the examined samples of chicken carcasses had Mould count exceeding the permissible limit. Nearly similar results were recorded by Habib, (2017) who found that mold count for frozen carcasses ranged from 7×10 to 4.7×10^3 CFU/g with mean value of 6.6×10^2 CFU/g.

Mold count was used as an index of proper sanitation and high quality products. Molds were an assist in the putrefactive processes and in other cases. They may import a moldy odor and taste to food stuffs. Molds can grow over an extremely wide range of temperature. Therefore, one can find Mould on particularly all foods at almost any temperature under which food are held. Besides Mould can assist in the putrefactive process and produce toxic substances namely mycotoxins which are harmful to man and animal (Frazier and Westhoff, 1998).

Yeast normally played a small role in spoilage because they constituted only a small portion of the initial population, because they grew slowly in comparison with most bacteria and because their growth may be limited by metabolic substance which can produced by bacteria. Spoilage yeast those found their way into foods being widely distributed in nature resulting in undesirable change in physical appearance of food (Walker, 1976).

The recorded data in Table (3) clarified that Yeast counts for breast samples ranged from 5×10 to 5.3×10^3 CFU/g with mean value of 1.9×10^3 CFU/g while for thigh samples ranged from 1.6×10^2 to 3.1×10^4 CFU/g with mean value of 8.9×10^3 CFU/g. It was clear that mean value of Yeast count of thigh samples was higher than that of breast samples. In addition, it was recorded that all of the examined samples of chicken carcasses had yeast count exceeding the permissible limit. Nearly similar results were recorded by Habib, (2017) who found that yeasts count for frozen carcasses ranged from 1.4×10^2 to 3.1×10^4 CFU/g with mean value of 8.9×10^3 CFU/g.

E. coli was a related coliform bacteria predominant among aerobic commensal flora in gut of man, animal and poultry, so there presence in poultry carcasses is an indicator of fecal contamination. The pathogenic bacteria like *E. coli* induce undesirable changes and constitutes a public health hazard in form of gastroenteritis or intoxication in consumers especially for children up to 2 years also cause urinary tract infection (Miskimin et al., 1976).

The recorded results in Table (4) revealed that the overall incidence of *E. coli* was 51% and it was higher in the examined samples of thigh (54%) compared to that of breast (48%). The obtained results of the rate of isolation of *E. coli* from chicken carcasses was higher than that recorded by Zhao et al., (2001) (38.7%), Samaha et al., (2003) (22.86%) and Hossam, (2012) (10%). At the same time, this finding was lower than that recorded by Hassan, (2015) (72%) and Habib, (2017) (70 %).

Moreover, serotyping of the obtained isolates of Enteropathogenic *E. coli* from chicken carcasses was recorded also in Table (4). It was found that serotype O111:H2 (EHEC) (11 isolates) was identified in breast and thigh samples with an incidence of 12 and 10%, respectively, serotype O91:H21 (EPEC) (8 isolates) identified in breast and thigh samples with an incidence of 8% for each, O127:H6 (ETEC) (11 isolates) identified in breast and thigh samples with an incidence of 12 and 10%, respectively, O113:H4 (EPEC) (11 isolates) identified in breast and thigh samples with an incidence of 16 and 6%, respectively and lastly O124 (EIEC) (10

isolates) identified in breast and thigh samples with an incidence of 8 and 12%, respectively.

The predominant serotypes were EPEC and EHEC, Shiga-toxin producing *E. coli* (STEC) was identified as a worldwide cause of serious human gastrointestinal disease and the life threatening Haemorrhagic Uremic Syndrome (HUS). The most common serotype causing (HUS) was *E. coli* O157:H7 (Karch et al., 2005) characterized by acute renal failure, haemolytic anaemia and thrombocytopenia, usually occurs in young children (under 5 years of age). Moreover, it was a major cause of acute renal failure in children in UK and several other countries and up to 10% of patients infected with VTEC O157 developed HUS (Stewart et al., 1997).

Generally, the presence of *E. coli* in examined chicken considered as an indicator for improper handling or unhygienic conditions which agreed with Frazier and Westhoff, (1998) and Hashim, (2003). *E. coli* related coliform bacteria predominant among aerobic commensal flora in gut of man, animal and poultry, so there presence in poultry carcasses is an indicator of fecal contamination.

Salmonellae were one of the most frequent causes of food borne illness worldwide and transmission involves foods of animal origin (Khaitas et al., 2007).

The recorded results in Table (5) revealed that the total incidence of Salmonellae was 18%; 9 isolates were recovered from breast samples and 9 samples were recovered from thigh samples. This finding was nearly similar to that recorded by Habib, (2017) who found that the rate of isolation of Salmonellae from frozen chicken carcasses was 16.67%.

The obtained results of the rate of isolation of Salmonellae from chicken carcasses was lower than that recorded by Zhao et al. (2006) (39%) and Donado-Godoy et al. (2012) (27%) and while it was higher than that recorded by Duarte et al. (2009) (9.6%).

The presence of Salmonellae in cooked foods is often attributed to inadequate sanitation, poor personal hygiene during food handling, processing and storage, presence of waste close to food preparation and food premises, and inadequate refrigeration.

Moreover, serotyping of the obtained isolates of Salmonellae from imported chicken carcasses was recorded also in Table (5) and revealed the detection of *S. Enteritidis* at the rate of 6 and 8 % from the examined samples of breast and thigh, respectively, *S. Typhimurium* at the rate of 8 and 6 % from the examined samples of breast and thigh, respectively and *S. Kentucky* from the examined samples of breast and thigh at the rate of 4 % for each. It is noticed that *S. typhimurium* and *S. Enteritidis* were the most prevalent Salmonella serotypes as well as many authors like Ulloa et al. (2010) who found that all six Salmonella isolates corresponded to *S. Enteritidis*, Chagas et al. (2013) who found that 94% of the chicken carcass samples were contaminated by 46 Salmonella isolates among these isolates 32% were genotyped as Salmonella Enteritidis and the most prevalent serotype isolated by Abdellah et al. (2009) was *S. Typhimurium* (40.35%).

Such results were expected due to handling contamination during processing or during storage and retailing in supermarkets, also throughout rodents as rats and mice excreta which contain *S. Typhimurium* that contaminated food directly or indirectly and cause cross contamination.

5. Conclusion

The obtained results in the current work clarified that chicken carcasses had a significant species of potential pathogenic bacteria that may be attributed to unsanitary conditions, cross contamination and personal hygiene conditions during handling, packaging, storage, distribution and selling. Therefore, to keep chicken carcasses with high quality to safeguard consumer's health, strict hygienic precautions must be followed without delay in the loading of imported frozen chicken and vehicles used for transport should be fitted with temperature records to monitor the environment, and the transfer should be as rapid as possible.

Conflict of interest statement

No conflicts of interest.

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