



## Microbiological quality of imported frozen broiler meat in Jordan

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Received 27 June 2013; Received in revised form 30 September 2013; Accepted 30 September 2013

**Aims:** The microbiological quality of broiler meat is of concern to health authorities and industries alike. The presence of pathogenic organisms is a major cause of food borne diseases and high numbers of organisms usually result in meat spoilage. This communication reports on the microbiological quality of imported frozen broiler meat to Jordan prior to distribution in to the market place.

**Methodology and results:** A total of 100 consecutive imported lots of frozen broilers, received at the laboratories of Jordan Food and Drug Administration were included in this investigation. All tests were performed in accordance with United States FDA or ISO recommended procedures. Results indicated that Mechanically Deboned Meat (MDM) was by far the most contaminated, with total plate count ranging between  $< 10^3$  and  $> 10^6$  CFU/g. Total coliform count varied between  $< 10$  and  $> 10^5$  CFU/g. For all other categories of meat (whole carcasses, off bones and cut chicken portions), total plate count was always  $< 10^3$  while total coliform was below 10 CFU/g. Other parameters tested included counts for *Escherichia coli*, *Staphylococcus aureus* in addition to *Clostridium perfringens* and in all samples; these organisms were recovered in counts below 10 CFU/g. *Salmonella* species and *E. coli* O157:H7 were not detected in any sample. Based on the results of this investigation, one lot of MDM failed to comply with specification of total plate count and another failed to pass the criterion of total coliforms.

**Conclusion, significance and impact study:** Results of this investigation suggested that the microbiological quality of imported broiler meat in Jordan was with acceptable microbiological quality in regard to the microorganisms investigated. This is probably due to the strict regulation followed in the kingdom for the registration of exporting companies.

**Keywords:** Microbiological quality, imported broiler meat, total plate count, coliforms, Jordan

### INTRODUCTION

Human consumption of broiler meat has consistently increased over the years and the same trend was observed in Jordan. In 2005 the amount of chicken consumed was 22.11 kg / person / year and this figure is expected to rise to 26.2 kg / person / year in 2015 (Freiji, 2008; Al-Masad, 2010). The ability of the domestic poultry farms to meet the demand of the local market is limited as they can produce up to 120 thousand tons annually (Sayegh, 2007); whereas, the current need stands at about 175 thousand tons per annum. Therefore, the deficit in market demand for this commodity is compensated by importation. Broiler meats are usually imported in to the kingdom mostly in the frozen state and include whole chicken carcasses, Mechanically Deboned Meat (MDM), off bones as well as cut chicken portions.

Broiler meat is highly susceptible to microbial contamination and continuous efforts are taken worldwide to minimize the level of contaminants. This is done in order to safe guard consumers from food borne diseases and to protect meat from microbial spoilage. Hazard Analysis Critical Control Points (HACCP) was developed to provide substantial improvements in the production of

safe foodstuffs including broiler meat (Unnevehr and Jensen, 1996). The goal of HACCP was to focus on the hazard in a particular food commodity that is likely to affect public health if left uncontrolled. However, product design, processing, hygienic production conditions, handling, transportation and commercialization were critically addressed in the HACCP regulations (Bonne *et al.*, 2005); the ultimate protection to consumers is the responsibility of governments. The quality of broiler meat should be checked before being distributed in the market. Testing results should be in compliance with pre set specifications which encompass; microbiological limits, odor, antibiotics concentration and physical appearance.

Compliance of the finished product with specifications reflects in most cases the extent of manufacturer adherence to HACCP based procedures and other hygiene control measures. In some countries, regular inspection visits to poultry industrial facilities by authorized personnel's affiliated to concerned institutions responsible for food safety are made. In Jordan, Food and Drug Administration (JFDA) is responsible for inspection and authorization of product release into the market. Inspection is possible if the product is sold in the country of origin but the situation is different when the

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product is imported. In the latter case exporters must provide evidence that processed foodstuffs were produced by HACCP certified companies and that imported commodities meet specifications.

The microflora of raw chicken meat is heterogeneous and originates either endogenously or from slaughtering premises, operators' hands, equipment, packaging materials water and air in the production premises. The Complete absence of microbial contamination is not possible but the risk of food borne diseases and spoilage can be reduced substantially by minimizing their numbers and the exclusion of pathogenic microorganisms. Epidemiological data about food-borne illness suggest that broiler meat consumption even in developed countries is an important cause of food-borne infections in humans (Fitzgerald *et al.*, 2001). The objective of this investigation was to establish the microbiological quality of imported frozen broiler meat into Jordan before being distributed in to the market.

## MATERIALS AND METHODS

A total of 100 consecutive samples of imported frozen broilers received and tested at the laboratories of JFDA were included in this study. The complete process of thawing, dilution and culturing was performed in a laminar flow to avoid contamination. Thawing of chicken meat was performed at room temperature before 50 g of sample was taken aseptically by scalpel excision and stomached in a sterile stomacher bag containing 450 mL of peptone water (PW Oxoid Ltd., Hampshire, England) for 2 min. Ten fold serial dilutions were carried out using the same diluents and from the appropriate dilution aliquots of 1 mL were used for microbiological investigations using officially recommended techniques as follows:

Total aerobic plate count (APC) was determined by pour plate technique using plate count agar (PCA) (Oxoid) as described by Maturin and Peeler (US FDA, 2001). Inoculated plates were incubated at 30 °C for 72 h before developed colonies were counted.

Coliforms were enumerated using violet red bile agar (VRBA) as described by ISO 4832 (2006). Colony count was performed after the incubation period of 48 h at a temperature of 37 °C was completed.

*Escherichia coli* count was determined by most probable number technique (MPN), from a previously prepared 10 fold serial dilutions, one mL aliquot of each dilution was transferred aseptically into triplicate tubes of Lauryl Sulfate Tryptose (LST) broth containing inverted Durham tubes. After incubation at 35 °C for 48 h, gas production was noted (Feng *et al.*, 2002). The MPN of *E. coli* was calculated on the bases of confirmed positive LST tubes for 3 consecutive dilutions by using MPN tables.

*E. coli* O157:H7 was isolated from samples using a selective enrichment technique (ISO, 16654). In brief the procedure involved blending 10 g of sample with 90 mL of sterile LST for 2 min to get 1:10 homogenate of the meat. This was incubated at 37 °C for 24 h before 0.1 mL of

appropriately diluted LST was streaked onto Tellurite Cefixime Sorbitol-MacConkey Agar (TCSMAC) plates. These plates were incubated at 35-37 °C for 18-24 h. Further confirmation of the isolates was carried out by inoculating colonies on to Trypticase Soy Agar with yeast extract (TSAYE) plates and then allowing the developed colonies to grow onto Levine's Eosin-Methylene Blue (L-EMB) agar and *E. coli* agar with MUG (EC with MUG). Final identification of indole positive colonies was performed using Remel's RapIDTM ONE System and *E. coli* 0157 latex agglutination test.

*Salmonella* species isolated from samples was carried out in accordance with method described by Andrews *et al.* (2011). Sterile lactose broth was used to blend the sample, followed by inoculation into Rappaport-Vassiliadis (RV) medium and Tetrathionate (TT) Broth for enrichment purposes. These enrichment cultures were incubated for 24 h at 42 °C for RV medium, and at 43 °C for TT Broth. Further isolation was performed using Bismuth Sulfite (BS), Xylose Lysine Desoxycholate (XLD) and Hektoen Enteric (HE) Agar plates. Isolates were confirmed as Salmonella by using Remel's RapID ONE System and Salmonella Latex agglutination Test.

*Staphylococcus aureus* was estimated on Baird-Parker Agar using the method given by Bennett and Lancette (2001).

*Clostridium perfringens* count was determined by pour plate technique using sporulation of *Clostridium perfringens* agar (SPA) as recommended by ISO 7937 (2004): One mL of the previously prepared decimal dilutions was transferred aseptically into two separate sterile Petri dishes to which approximately 15 mL of sterile molten SPA were added. After thorough mixing, the inoculated plates were allowed to solidify before being incubated at 35-37 °C for 48 h under anaerobic conditions (using anaerobic jar with CO<sub>2</sub> generation packet). Media used throughout this work were from (Oxoid- England).

## RESULTS

The microbiological specification of broiler meat as stipulated by the Jordanian Institute of Standards and Metrology is given in Table 1. Results obtained in this study were interpreted in accordance with these specifications. Table 2 shows the country of origin and category of chicken meat imported in relation to the number of tested lots. It is clear that by far, Brazil is the major exporter of broiler meat to Jordan with MDM being the most imported category.

Total aerobic plate count detected in each imported lot is shown in Table 3. It is evident from this table that total count varied from < 10<sup>3</sup>/g to > 10<sup>9</sup>/g. The same table demonstrates that MDM was the category that harbored the highest total aerobic plate count whereas; in the other categories this count never exceeded 10<sup>3</sup> CFU/g. It is important to note that on the bases of total count only one lot failed to pass the microbiological criteria of the quality standards followed in this investigation. Total coliform count for all tested lots is given in Table 4. This count was by far the highest in MDM where the majority (38 lots)

**Table 1:** Jordan microbiological specifications of chicken meat.

| Microbiological parameter          | CFU/g                 |
|------------------------------------|-----------------------|
| Total aerobic plate count          | < 10 <sup>6</sup>     |
| Total <i>Staphylococcus aureus</i> | < 5 x 10 <sup>2</sup> |
| Total <i>Escherichia coli</i>      | < 5 x 10 <sup>2</sup> |
| Total Coliform                     | < 5 x 10 <sup>3</sup> |
| <i>Streptococcus faecalis</i>      | < 5 x 10 <sup>2</sup> |
| <i>Clostridium perfringens</i>     | < 10 <sup>2</sup>     |

**Table 2:** Number of tested lots of chicken imported to Jordan in relation to country of origin and category of the meat.

| Country | MDM | Chicken carcass | Off bones | Thighs | Total |
|---------|-----|-----------------|-----------|--------|-------|
| Brazil  | 24  | 20              | 21        | 0      | 65    |
| Turkey  | 12  | 0               | 0         | 0      | 12    |
| USA     | 0   | 0               | 0         | 10     | 10    |
| Ukraine | 0   | 9               | 0         | 0      | 9     |
| Norway  | 4   | 0               | 0         | 0      | 4     |
| Total   | 40  | 29              | 21        | 10     | 100   |

**Table 3:** Ranges of total aerobic plate count detected in each imported chicken meat lot

| Category of chicken meat | Ranges of total CFU/ g |                                      |                                     |                                     |                  |
|--------------------------|------------------------|--------------------------------------|-------------------------------------|-------------------------------------|------------------|
|                          | < 10 <sup>3</sup>      | >10 <sup>3</sup> - < 10 <sup>4</sup> | >10 <sup>4</sup> - <10 <sup>5</sup> | >10 <sup>5</sup> - <10 <sup>6</sup> | >10 <sup>6</sup> |
| MDM                      | 4                      | 10                                   | 15                                  | 10                                  | 1                |
| Full Carcass             | 29                     | 0                                    | 0                                   | 0                                   | 0                |
| Off Bones                | 21                     | 0                                    | 0                                   | 0                                   | 0                |
| Thighs                   | 10                     | 0                                    | 0                                   | 0                                   | 0                |

**Table 4:** Ranges of total coliforms count detected in each imported chicken meat lot.

| category of chicken meat | ranges of total coliforms plate count/g |                         |                                     |                  |
|--------------------------|---|-------------------------|-------------------------------------|------------------|
|                          | < 10                                    | >10 - < 10 <sup>2</sup> | >10 <sup>2</sup> - <10 <sup>5</sup> | >10 <sup>5</sup> |
| MDM                      | 0                                       | 28                      | 1                                   | 1                |
| Full Carcass             | 29                                      | 0                       | 0                                   | 0                |
| Off Bones                | 21                                      | 0                       | 0                                   | 0                |
| Thighs                   | 10                                      | 0                       | 0                                   | 0                |

contained counts between >10 but less than 10<sup>2</sup> coliform/g. Two lots harbored high numbers; one contained 9 x 10<sup>2</sup> and the other 5 x 10<sup>4</sup> coliform/g.

Therefore, these two lots failed to pass the Jordanian criterion for coliforms count. It is worth mentioning that one of these lots was the one which failed to pass the test for total aerobic plate count as it was more than 10<sup>6</sup> CFU/g, while the other was with high aerobic count but passed that particular test. In regard to the other microorganisms it was found that all imported lots shared almost the same identical results; Total *E. coli* was always < 10 CFU/g, total *S. aureus* < 10 CFU/g and *Clostridium perfringens* < 10 CFU/g. Throughout this work *E. coli* 0157 and *Salmonella* species were never isolated.

## DISCUSSION

Most published work pertinent to the microbiological quality of broiler meat has dealt with samples collected from the market place (Cohen *et al.*, 2007) and in certain instances these samples were found to be of unacceptable quality (Cahaba, 2007). This work reports on the microbial load of broilers meat lots imported to Jordan and results presented herein were detrimental in the decision making process to permit or reject the distribution of these lots into the market. This work does not provide any guarantee that the microbiological quality of broiler meat described in this investigation would remain the same as a result of handling in the market place or at homes.

The microbiological quality of broiler meat is an indication to the overall hygienic conditions followed in the slaughter houses, cleaning procedures, packaging, storage and distribution. Any irregularity in the hygienic standards during these production steps would be reflected on the CFU/g obtained using total aerobic plate count and the isolation of certain indicator organisms. Results of this investigation indicated that two out of 100 imported lots were out of specifications with the MDM being with the highest microbial content.

Mechanically Deboned Meat was defined by the Regulation (EC) 853/2004 as the product obtained by removing meat from flesh-bearing bones after boning using mechanical means and this process results in the loss or modification of the muscle fiber structure. Kumar *et al.* (1986) indicated that the large surface area, the release of cellular fluids and the heat generated during mechanical deboning will all enhance bacterial growth in this category of broiler meat. Due to the fear of high bacterial contamination of MDM many suggested the use of heat or chemical disinfectants during the extraction procedure to minimize level of contaminants (Bijker *et al.*, 1987; James and James, 2002; Pomykala and Michalski, 2008; Hecer and Ulusoy Sözen, 2011). Therefore, our findings regarding the high microbial content of MDM are consistent with the generally accepted literature.

With the exception of MDM, results of total aerobic plate count presented in this investigation were very much lower than those reported by others. Amara *et al.* (1994) found that TAPC in broiler meat varied between  $6 \times 10^6$  and  $10^7$  CFU/g, whereas Avadi and Safarmashaei (2011) calculated the mean of TAPC to be equivalent to  $10^5$  CFU/g. Similar results were also obtained from different countries such as Turkey, Pakistan and Morocco (Göksoy *et al.*, 2004; Chaudhry *et al.*, 2011; Cahaba *et al.*, 2007). The remarkable difference between our results and those of others could be attributed to one or both of the following factors: first, all lots analyzed were frozen and the process of freezing by itself is antagonistic against the growth of microorganisms; second, The Jordan FDA is very demanding in the registration of foreign companies that export food into the kingdom, compliance with the rules and regulations of HACCP is a *pri-requisite* for registration and at random these companies are subject to inspection by JFDA experts.

The isolation of *E. coli* in particular and the detection of high numbers of coliforms in general is an indication of fecal pollution which reflects the microbiological quality of the commodity under test (Capita *et al.*, 2002). Their presence in alarming levels may be attributed to a faulty evisceration and improper washing of the carcasses after this process. The use of contaminated water for washing purposes or cleaning of machines may also contribute to this contamination phenomenon. As shown in Table 4, MDM was the category of chicken meat which was highly contaminated with coliforms while chicken portions, off bones and whole carcasses harbored coliforms in counts below 10 CFU/g. but *E. coli* as well as *E. coli* O157 were either present in negligible number in case of the former organism (count < 10 cells/g) or not present at all in the

case of the latter organism. Upon comparison, counts of coliforms given in Table 4 were found to be a lot lower than those reported by Cahaba *et al.*, (2007) and well below the allowed limit in the Jordanian specifications. On the other hand, *E. coli* count was always detected in counts below 10 CFU/g as established by the MPN method.

Fries (2002) has pointed out the significance of poultry meat contamination with *Salmonella* species during slaughterhouse processing of broilers. Although *Salmonella* spp. were never detected in any of the broiler meat tested in the current work, it has been detected in 15.39% of chicken breast fillets and 10.53% of frozen ground chicken tested by Kozačinski *et al.* (2006). Quite high figures of *Salmonella* contamination was reported by Göksoy *et al.* (2004) who emphasized that such a contamination is affected by the procedures followed in the slaughter houses or the processing plants. These authors have highlighted the sources of contamination as follows: 1. the rapid rate of production keeps the birds in close proximity throughout processing; 2. limitations in the design of processing equipment, including that used in scalding, defeathering, and evisceration; 3. the difficulty of washing the abdominal cavity effectively after evisceration when the carcass remains as a full body; 4. retention of water by skin, which tends to entrap bacteria in the crevices and feather follicles. It is evident that contamination can be prevented or at least minimized if adequate processing procedures are followed and good hygienic conditions are maintained.

Cohen *et al.* (2007) demonstrated that Coagulase positive Staphylococci and *Clostridium perfringens* were respectively detected in 10.4% and 7.2% of commercially available poultry meat they tested. These results were in agreement with those reported by Wen and McClane (2004) who investigated levels of *C. perfringens* in American retail foods. In this context it is worth mentioning that *S. aureus* as well as *C. perfringens* are major causes of food borne diseases and poisoning. These organisms were detected in the broiler meat studied here in numbers well below the allowable limits in Jordan.

## CONCLUSION

This work indicated that imported frozen broiler meat to Jordan is with acceptable microbiological quality in relation to organisms tested in the current work. This is probably "in most part" due to the rules enforced by the Jordan FDA and the strict regulations followed in the registration of companies that export meat to the country.

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