EFFECT OF MULTIPLE FREEZE-THAW CYCLES ON THE QUALITY OF LOCAL AND IMPORTED BROILERS CHICKEN MEAT

By

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ABSTRACT

Although freezing is a good preserving technique that keep the meat quality for up to one year, nevertheless, repeating of freeze -thaw process represent a significant problem that affect the sensory, chemical and microbiological quality of meat. The objective of this study was to explore the effect of repeated freeze-thaw cycles (F-T cycle) of frozen chicken meat on the physical, chemical and microbiological quality of imported and local frozen chicken. Frozen broilers were left in refrigerator at (4-7°C) until complete thawing then it were examined for the different parameters. Refreezing of thawed broiler carcasses was done in deep freezer at (-18 °C) for one week. As the number of F-T cycle increased the overall sensory characteristics declined significantly in raw and cooked broilers and the drip loss decreased significantly (P < 0.05), although the total drip loss increased. Shear force increased then decreased by the 3rd cycles in imported broilers, but in local broilers it decreased by the 2nd cycle. The sensory characteristics of imported broilers were better than that of local broilers, which deemed as spoiled by the second F-T cycle. PH increased significantly (P<0.05) among the cycles. Moreover, repeated F-T cycles resulted in a greater degree of lipid and protein oxidation, as TVB-N, TBARS values increased greatly. Aerobic plate count (mesophilic and, psychotropic) and total yeast-mold count were significantly (P<0.05) higher in local broilers than that of imported one and markedly increased by repeating F-T cycles. According to the Egyptian standards, the local broilers were considered unfit for human consumption from the second F-T cycle, meanwhile the imported broilers in most of the examination were within the permissible limits until the 3rd F-T cycle.

<u>Keywords:</u>

Freeze-thaw, chicken broilers, TVB-N, TBARS, sensory and microbiological analysis.

INTRODUCTION

Freezing has been an excellent preserving technique in which meat and meat products can be preserved in a condition similar to that of normal state and can be kept satisfactory for six months to one year (Rahman *et al.*, 2014). Nonetheless, the consequences of freezing and thawing on the quality of meat remain a significant problem (Leygonie *et al.*, 2012a). Freezing and thawing are complex processes that involve heat transfer as well as a series of physical and chemical changes, which can affect the quality of the meat and meat products (Bing *et al.*, 2002).

Storage or transportation of frozen food in temperature above freezing point is a bad practice applied in many retail outlets, restaurant and home, which result in thawing of the food then refreezing and repeating of this cycle. Temperature fluctuation and multiple freeze-thaw (F-T) cycles result in some quality deterioration, especially with regard to flavor, texture, and color, due to protein denaturation, osmotic removal of water, mechanical damage, and protein aggregation and cross-linking (Xia *et al.*, 2009) and (Leygonie *et al.*, 2012a).

Microbial spoilage is effectively terminated during freezing, as the microbes become dormant. Unfortunately, they regain their activity during thawing, as thawing are a much slower process as freezing and is less uniform, certain areas of the meat will be exposed to more favorable temperature conditions for microbial growth. There is an increase in moisture and nutrients available to microbes post freeze-thaw due to exudate formation. The moisture lost during thawing is rich in proteins, vitamins and minerals derived from the structural disarray caused by the freezing process, which consequently provides an excellent medium for microbial growth. The reactivation of microbial activity upon thawing coupled with cellular damage resulting from frozen storage because more rapid spoilage of frozen-thawed meat compared to fresh meat (Leygonie *et al.*, 2012a).

Therefore, the objective of the current study was to assess the effects of multiple freeze - thaw cycles on physical, chemical and microbial changes in chicken meat.

MATERIAL AND METHODS

Two lots of frozen broilers carcasses (25 each) were transported to the lab in an icebox. Broilers carcasses were purchased from Cairo market (local imported) at the first third of their shelf life and were kept at (-18 °C) deep freezer, until examined.

404

Examined Freeze-Thaw (F-T) cycles were performed as follow:

Immediately after transportation to lab (3) carcasses from each lot were left in refrigerator until complete thawing then examined for the different parameters. Refreezing of thawed broilers was done in deep freezer (-18 °C) for one week, thawing was done in the refrigerator at (4°C) Weekly F-T cycles were continued until the sensory examination deems the samples spoiled.

I.Sensory analysis:

The sensory attributes of raw (color, odor, consistency and overall score) and cooked (flavor, juiciness, taste and overall score) broiler samples were analyzed by a trained panelists team consist of 9 members of ¹ Animal Health Research Institute, Agriculture Research Centre assessing the sensory quality of thawed samples. Using a five-point scale as follows: 5 for "excellent", 4 for "very good", 3 for "good", 2 for "fair" and 1 for "poor" (**Rahman** *et al.*, **2012**). An overall score of one or less was deemed as spoiled.

II.Physico-chemical characteristics.

1.Shear force;

Chicken meat samples were analyzed for shear force (kgf/cm³) at Cairo University Research Park (CURP), Faculty of Agriculture. Steaks of 2x2x2 cm were prepared from each breast and thigh chicken meat cooked at core temperature (72°C) and six core samples of 1.3cm diameter were removed parallel with the muscle fiber direction using hand- held coring device. Each core was sheared once with Warner-Bratzler sheare force (WBSF) device attached to an instron Universal Testing machine (Model 2519-105, USA) with a 55-kg tension/compression load cell and a crosshead speed of 200 mm/min. An average shear force value was calculated and recorded for each sample.

2. Determination of pH:

PH, as a mean of three measurements was done using pH meter (Jenway, 3310), according to **Harold** *et al.* (1981).

3.<u>Drip loss:</u>

Drip loss was determined according to Li *et al.* (2014). After complete thawing of the sample, it was weighed as a whole (W1), then weighed again after blotting dry (W2) with tissue paper. Drip loss was calculated as the percentage of weight lost as follows:

Drip loss % =
$$\frac{W_1 - W_2}{W_1} \times 100$$

4.DeterminationTotal Volatile Basic Nitrogen (TVB-N) (mg/100g) according toEU (1995).by using distillation method.

Calculation:

$$TVB - N = \frac{(V1 - V2) \times 0.14 \times 2 \times 100}{M}$$

V1 = Volume of 0.01 M HCl in ml for sample

V2 = Volume of 0.01 M HCl in ml for blank

M = Weight of sample in g.

5.Thiobarbituric Acid Reactive Substances (TBARS) mg malonaldehyde (MDA)/kg were determined according to **Robles-Martinez** *et al.* (1982).

Calculation:

$TBARS = \mathbf{R} \times 7.8$

Where: R = absorbance of the sample against the blank.

III.Microbiological examination.

The technique recommended by **APHA (2001)** was applied for determination of aerobic plate count (Mesophiles) at 35 ° C for 48 h and psychrotrophs at 7 ° C for 7-8 days and mold and yeast count at 25 ° C for 5 days.

IV.Statistical analysis.

The experiment was conducted in three repetitions. Data were analyzed using SPSS software (release 20, IBM CO). Means were compared using T-test, and significance was tested at $\alpha = 0.05$

RESULTS AND DISCUSSION

Imported and local frozen broilers carcasses were subjected for three F-T cycles (one week each), and examined after each cycle, the local broilers samples were deemed spoiled by sensory examination after the 3rd F-T cycle, so it was examined chemically and microbiologically after the 1st and 2nd cycles only.

Sensory examination:

The mean values of overall sensory score Fig. (1) in raw imported broilers after the first F-T cycle was significantly (P<0.05) higher than that of raw local broilers, which continued by the 2^{nd} F-T cycle, and the same at the 3^{rd} cycle where, the local broilers showed spoilage sensory

406

j. Egypt. act. med. Assac 78, no 3. 403 - 417/2018/

EFFECT OF MULTIPLE FREEZE-THAW CYCLES ON THE

attributes. Sensory scores were gradually declined as affected by repeating F-T cycle to

indicate deterioration of overall sensory score. These results were in agreement with those reported by Xia *et al.* (2009), Leygonie *et al.* (2011), Leygonie *et al.* (2012 b), KAkhtar *et al.* (2013), Rahman *et al.* (2014) and Ali *et al.* (2015).

The mean values of overall sensory score Fig. (2) in cooked imported samples after the first F-T cycle was significantly (P<0.05) higher than that of cooked local broilers. This difference continued through the 2nd and the 3rd F-T cycle, where spoilage signs were seen as local broilers. It is obvious that overall sensory score of cooked imported and local broilers decreased from the 1st F-T cycle to the 2nd F-T cycle and continued to in the 3rd F-T cycle. **Xiong (2000), Mancini and Hunt (2005), Yu,** *et al.* **(2005) Luciano** *et al.* **(2009) and Leygonie** *et al.* **(2012b), in agreement with those report these results.**



 Fig. (1): Overall sensory score for raw chicken
 Fig.(2):Overall sensory score for cooked chickenen

 meat during 3 freeze-thaw cycles.
 meat during 3 freeze-thaw cycles.



Fig.(3):Shear force mean values for breast chicken meatduring 3freeze-thaw cycles.



There are sig. diff. (P<0.05) between points having the same letter.

Physico-chemical characteristics.

Shear force:

Shear force is used as a measurement of meat tenderness. Fig. (3, 4) illustrates the mean values of shear force for imported and local examined samples (kgf/cm³). Concerning the breast samples, the mean values of shear force Fig. (3) in imported broilers was significantly (P<0.05) lower than that of local broilers after the 1st F-T cycle, which continued by the 2nd F-T cycle. These results are in relation well with sensory score for cooked samples Fig. (2). concerning the thigh samples, the mean values of shear force Fig. (4) in imported broilers was significantly (P<0.05) lower than that of local broilers. Nevertheless, by the 2nd F-T cycle there was no significant difference (P>0.05) between imported and local broilers after the 1st F-T cycle. Regarding the effect of F-T cycle, it is obvious that shear force values of even thigh or breast increased in the 2nd F-T cycle, then decreased again by the 3rd F-T cycle, in imported broilers. On the other hand, shear force values decreased from the 1st F-T cycle in local broilers. These results are in agreement with those reported by Lagerstedt *et al.* (2008), Xia *et al.* (2009) and Lui *et al.* (2010).

408

j. Egypt. net. med. Assac 78, no 3. 403 - 417 (2018)

<u>PH:</u>

The pH values of both thigh and breast increased by repeating of F-T cycles in both of imported and local broilers.

Concerning the pH of breast and thigh samples, as illustrated in Fig. (5,6) there was no significant difference (P>0.05) between imported and local broilers at the 1st cycle. However, by the 2nd F-T cycle the mean values of pH in imported broilers was significantly lower (P<0.05) than that of local broilers, which indicate a good keeping quality of imported broilers than the local one. These results are in agreement with those reported by **Rahman** *et al.* (2015). On the contrary, **Ersoy** *et al.* (2008), Leygonie *et al.* (2012b) and Zhang *et al.* (2017) reported a slight decrease in pH after freezing and thawing process. the increase or decrease of meat pH from the initial value influences the rate of oxidation as well as the microbial shelf life and drip loss and vice versa (**Rahman** *et al.*, 2015).



Fig. (5): pH mean values for breast chicken meat during 3 freeze-thaw cycles.



Drip loss:

The mean values of drip % Fig. (7) In imported broilers was significantly (P<0.05) higher than that of local broilers in the 1st F-T cycle, but by the 2nd F-T cycle, these values were nearly the same for imported and local broilers. The drip loss values decreased as freeze-thaw

j.Egypt.net.med.Assac 78, no 3, 403 - 417/2018/

cycles increased for both imported and local broilers as it exceeded the permissible limit for drip loss stated by the Egyptian standard (ES, 1090\2005) by (5%) from the 1st F-T cycle, and the cumulative drip loss in the 2nd F-T cycle reached 14.5% and 12% for imported and local broilers, respectively, Meanwhile it reached 19.3% by the 3rd F-T cycle in the imported broilers. The increased drip loss may be due to dripping of broilers with water for prolonged chilling during slaughtering process without enough time for dripping before freezing increase carcasses weight. The obtained results in this study agree with those reported by Xia *et al.* (2009), Rahman *et al.* (2014), Rahman *et al.* (2015) and Zhang *et al.* (2017) who stated that drip % decreased as the number of freeze-thaw cycles increased.



Fig. (7): Drip loss % of chicken meat duringFig. (8): TVB-N of chicken meat during 33 freeze-thaw cycles.freeze-thaw cycles.

There are sig. diff. (P<0.05) between points having the same letter.

410 j.Egypt.net.med. Assac 78, no 3. 403 - 417/2018/

TVB-N:

As shown in Fig. (8), the mean values of TVB-N (mg / 100 g) in imported broilers was significantly (P<0.05) lower than that of local broilers after the 1st F-T cycle, which continued by the 2nd F-T cycle. As the F-T cycles increased, it is obvious that TVB-N values increased for both imported and local broilers. According to the Egyptian standards (ES,1090\2005), the imported broilers were within the permissible limit (20 mg /100 g) even after the 2nd F-T cycle. Moreover, the local broilers exceeded this limit by the 2nd F-T cycle, which indicate the beginning of decomposition. These results could be explained and correlated with microbial load of both imported &local broiler carcasses examined. The obtained results are in agreement with those reported by Xiong (2000), Rowe *et al.* (2004), Sriket *et al.* (2007), Xia *et al.* (2012), KAkhtar *et al.* (2013), Zhao *et al.* (2015) and Zhang *et al.* (2017) who stated that there was a significantly increase in TVB-N (P<0.05) in meat subjected to multiple F-T cycles.

TBARS:

TBARS (mg malonaldehyde /kg) mean values Fig. (9) in the 1st F-T cycle for imported broilers was similar (P>0.05) to that of local broilers, which continued through the 2nd F-T cycle. Regarding the effect of F-T cycles, it is obvious that as the F-T cycles increase, TBARS values of both imported and local broilers increased. However, these values were below the permissible limit stated by the Egyptian standard (0.9), which was exceeded by the 2nd F-T cycle (ES, 1090\2005). The increased TBARS values indicate the increased fat oxidation due to fluctuation of temperature and bad storage conditions. These results agreed with those obtained by Benjakul and Bauer (2001), Morcuende *et al.* (2003), Jin *et al.*, (2009) Jin-Ping *et al.* (2012), Xia *et al.* (2012), Rahman *et al.* (2014) and Rahman *et al.*, (2015) who reported that TBARS value significantly (P<0.05) increased as the number of repeated freeze-thaw cycles increased. These could be attributed to microbial growth, enzymatic, oxidative rancidity.

Marwa A. H. Abbas et el



Fig. (9):TBA means values for chicken meat Fig. during 3 freeze-thaw cycles.



There are sig. diff. (P<0.05) between points having the same letter.

Microbiological analysis:

The mean values of APC Fig. (10), psychrotrophic Fig. (11) and mold and yeast Fig. (12) counts (log cfu/g) for imported broilers were significantly (P<0.05) lower than that of local broilers after the 1st F-T cycle. This result continued through the 2nd F-T cycle. Concerning the APC and psychrotrophic count of imported broilers. They gradually increased by repeated F-T cycles and reached to just above the permissible limit (5 log cfu/g) stated by the Egyptian standard **(ES, 1090\ 2005)** by the 3rd F-T cycle. Meanwhile mold and yeast count reached 3.39 log cuf/g by 3rd cycle. In case of local broilers, these counts were much higher as they began with counts near the maximum permissible limit and by the 2nd F-T cycle they were more than this limit by about 1.8 log cfu/g. In the same way, mold and yeast count in local broilers began with high count (3.07) and reached to 4.13 log cfu/g by the 2nd F-T cycle. It is worth to mention that these results indicated the good keeping quality of imported broilers comparing to the local one. The obtained results agree with those reported by **(Nollet, 2012)**. The increase in APC could be attributed to handling contamination during thawing process, bad storage condition and thawing that lead to prepare a suitable condition for microbial growth **(Leygonie** *et al., 2012a and Rahman et al., 2014)***.**

412 j. Egypt net med. Assac 78, no 3. 403 - 417/2018/

EFFECT OF MULTIPLE FREEZE-THAW CYCLES ON THE



Fig. (11): Psychotropic count mean log CFU/g for chicken meat during 3 freezethaw cycles.



There are sig. diff. (P<0.05) between points having the same letter.

[CONCLUSION

The objectives of this study were to know the effect of repeated freeze-thaw cycles of imported and local frozen chicken meat on the sensory, physicochemical quality and microbiological assessment. From the results of the present study, it can be concluded that, repeated freeze-thaw cycles affected the sensory, physicochemical and microbiological quality of chicken meat causing the deterioration of meat quality. It may also be concluded that avoiding repeated freezing and thawing of broilers was the best. The results of this study indicated that,the sensory characteristics, physicochemical characteristics of imported chicken were better than that of local broilers that deemed as spoiled by the beginning of the 3rd F-T cycle. This may be attributed to the low hygiene practice during slaughtering and preparation of local broilers.

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EFFECT OF MULTIPLE FREEZE-THAW CYCLES ON THE

تأثير دورات التجميد والاذابه المتعددة على جودة لحوم الدواجن المحلية والمستورده

1مروه عبد الحميد عباس ²خالد الخواص ³جيهان محمد عبد العزيز قاسم ¹طبيب بيطرى أول - إدارة التفتيش على اللحوم – مديرية طب بيطرى الجيزة ² رئيس بحوث - قسم الرقابة الصحية على اللحوم - معهد بحوث صحة الحيوان - الدقى ³ أستاذ الرقابة الصحية على اللحوم - كلية الطب البيطري - جامعة القاهرة

الملخص العربي

علي الرغم ان التجميد يعد أحد أفضل وسائل الحفاظ علي جودة اللحوم الي ما يزيد عن عام ألا ان تكرار عمليات الاذابه واعادة التجميد تعتبر أحد اكبر المشاكل التي تؤثر سلبا علي جودة اللحوم والدواجن من حيث خواص اللحوم الطبيعيه من اللون والملمس والرائحة وايضا الخصائص الكيميائية والميكروبيولوجيا. ان الهدف من الدراسة الحالية هو معرفة تاثير تكرار دورات الاذابة والتجميد علي لحوم الدواجن المجمدة المستوردة والمحلية من حيث التأثير علي الخواص الطبيعيه والكيميائية والميكروبيولوجيا. تمت اذابة عينات الدواجن المجمدة في تلاجات التبريد حتي يتم اذابتها بالكامل ثم اجراء التجارب المعملية عليها و اعادة تجميدها مره أخري في الفريزر لمدة اسبوع.

كلما زاد عدد دورات الاذابة والتجميد ادي ذلك الي زيادة في التغير السلبي في الخواص الحسية في لحوم الدواجن الغير مطهية والمطهية من حيث تغير اللون الواضح والملمس والرائحة بزيادة دورات الاذابة والتجميد و كذلك السائل الانفصالي الذي يقل كلما زاد عدد دورات الاذابة و التجميد وكذلك التأثير علي طراوه اللحوم االتي تقل بعد الدورة الاولي بشكل ملحوظ. أيضا زيادة دورات الاذابة و التجميد تؤدي الي زيادة تأكسد الدهون ووتكسير البروتينات الموجودة في لحوم الدواجن . مما يؤدي الي تزرنخها وتغير ملحوظ في الطعم والرائحة .كما يزيد عدد البكتيريا الهوائية وعدد البكتيريا المحبه البروده وكذلك الفطريات عن الحد المسموح بة في المواصفات القياسية المصريه لتصبح بذلك غير صالحة للاستهلاك الادمي.