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Impact of Different Thawing Methods on Physico-chemical Characteristics, Electrophoretic Profile and Sensory Evaluation of Frozen Beef *Longissimus dorsi* Muscle

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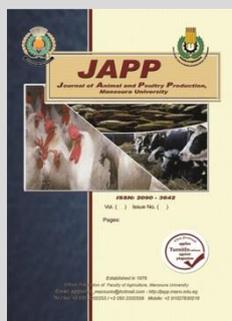


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ABSTRACT

In this study, the quality characteristics which include (physicochemical, structural characteristics and sensory evaluation) of beef loin submitted to fast freezing and thawed by five different methods (Refrigerator, Oven, Microwave, tap water and Room temperature) were examined. The beef samples were defrosted to the point of reaching a core temperature of 0°C, followed several parameters were examined which include physical characteristics [(drip loss, thawing loss, cooking loss, and water holding capacity (WHC)]. In addition to the chemical characteristics [(moisture content, protein solubility, thiobarbituric acid (TBA), myoglobin concentration (Mb) and met myoglobin formation (Met-Mb)]. As well as polyacrylamide gel electrophoresis profile (SDS-PAGE) and sensory evaluation were performed. The findings suggested that, beef sample thawed by tap water revealed lower drip, thawing and cooking loss, also leads to higher moisture content and WHC than the other thawing methods. Moreover, this thawing method (tap water) prevent lipid oxidation by decreased TBA value and percent of Met-Mb formation as well as increasing Mb. It was also evident from the findings that the thawing techniques influenced the electrophoretic patterns of the beef samples. It was found that the samples with microwave thawing method have a higher band counts and greater intensity, showing higher damage to the proteins of the meat, making fragmentation, structure loss and liberation of small peptides, which affecting the nutritional value and functionality of the meat. However, the data of this experiment have shown that beef sample thawed by tap water protect meat quality and consistency efficiently and will help the meat manufacture and those who consuming the products. It was concluded that the characteristics of meat quality are connected not only to the freezing process, but also to the system and conditions of thawing used.

Keywords: beef *longissimus dorsi*, frozen meat, thawing method, physico-chemical, SDS-page, sensory evaluation



INTRODUCTION

As of late, meat consumption has raised consistently because of its improved taste and nutritional value (Zhao *et al.*, 2019). Even though meat is rich in vitamins, minerals and essential amino acids, it is an extremely perishable food product (Choi *et al.*, 2017 and Leygonie *et al.*, 2012). Frozen storage has been used widely to increase the shelf life and to preserve the quality of meat. (Leygonie *et al.*, 2012). Until consumption or extra processing, frozen meat must be defrosted (Taher *et al.*, 2001 and Jia *et al.*, 2019). While one of the most un-forceful protection techniques is freezing, it actually brings about some food changes, particularly the formation of ice crystals (Leygonie *et al.*, 2012), which are influencing output characteristics. The degree of such mischief is straightforwardly linked to the freezing speed (Xia *et al.*, 2012). The defrosting cycle is viewed as a fundamental stage for frozen food before additional handling or eating, for example, cutting, dicing, and possible cooking. (Wen *et al.*, 2015).

The process of thawing typically occurs more slowly than the process of freezing, changing in physical and chemical properties, including changes in taste (Jia *et al.*, 2019), texture (James *et al.*, 2017), color (Alvarenga *et al.*,

2019), degeneration and protein aggregation, can be caused by a longer thawing period (Jia *et al.*, 2018). Defrosted meat loses other practical properties, such as water holding capacity (WHC) and proteins, in addition to water loss, which can affect the gelling process of the finished product containing the crude material (Olivo and Olivo, 2006; He *et al.*, 2013 and Xia *et al.*, 2012). Many variables influence the decline in meat quality during the defrosting period, including defrosting time (Qian *et al.*, 2019), defrosting temperature, and defrosting techniques (Choi *et al.*, 2017). Contrasted with customary defrosting procedures, for example water (Choi *et al.*, 2017), cooler (Qian *et al.*, 2019), and air defrosting (Li *et al.*, 2019). Some innovative defrosting technique for frozen meat have been widely used involving radio frequency (Bedane *et al.*, 2017), microwave (Choi *et al.*, 2017), vacuum (Cai *et al.*, 2018), and ultrasonic defrosting (Shi *et al.*, 2019).

These modern thawing techniques have many benefits, for example abridged defrosting time, decreased oxidation process, and improved meat quality. Therefore, to preserve meat quality and reduce losses, an appropriate thawing technique must be considered. Conventional strategies and modern technology were utilized to defrost meat in earlier research studies (Choi *et al.*, 2017 and He *et*

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al., 2016). Notwithstanding, air, water and cooling defrosting all outcome in helpless meat quality because of delayed defrosting and wide temperature ranges between the outer and inner meat layers, (Chakanya *et al.*, 2017; Chandirasekaran and Thulasi, 2010). Microwave, ultrasonic, radio frequency, high pressure, and ohmics were recommended and can significantly accelerate the defrosting of meat (Manios and Skandamis, 2015; Beauchamp *et al.*, 2010).

Be that as it may, every method has its own shortcomings. Every one of them can cause lopsided defrosting, denaturation of protein, and changes in conformation (Wu *et al.*, 2017). It is therefore important to establish a fast defrosting procedure that mitigates unwanted changes in frozen meat and preserves meat quality. Therefore, the motivation behind this study is to assessment the impact of five thawing techniques on the physico-chemical, electrophoretic pattern and sensory evaluation of beef longissimus dorsi muscle submitted to fast freezing.

MATERIALS AND METHODS

Sample preparation

The experiment was carried out at the laboratories of the animal science department, college of agricultural engineering sciences, University of sulaimani. In this study, *Longissimus dorsi* muscle was purchased from a local abattoir during the 24 h post-mortem period from five beefs aged approximately 30 months at the time of death. The samples were stored on ice and transferred to the laboratory. Both subcutaneous fat and connective tissues were aseptically removed prior to the experiment. A beef samples were randomly divided into five different experimental groups wrapped in moisture-impermeable poly-ethylene bags and individually frozen at the processing plant (2 hours of stored freezing to -36°C) and then stored in a freezer at (-8°C) before study.

Thawing methods

Experimental frozen samples are thawing by five different techniques. (Refrigeration in a domestic refrigerator at approximately (5±1°C) for 22 hours, in a microwave for 5 minutes, in an oven at ± 40°C for 2 hours and 30 minutes, wrapped in polyethylene bags of low density and immersed in tap water at approximately 20°C for 2 hours 30 minutes, and at room temperature at approximately 25°C for 2 hours and 30 minutes). When temperature of deep muscle exceeds 10°C in all techniques, thawing is terminated. Followed several characteristics were examined.

Physical characteristics

Water holding capacity (WHC)

Water holding capacity was estimated by the technique of (Zhou *et al.*, 2018) with some modification. Beef defrosted samples (10g) were wrapped in filter paper, put in a centrifuge tube, and centrifuge at 5000 rpm and 4°C for 10 minutes. The water holding capacity is calculated as follows:

$$\text{WHC (\%)} = \left[\frac{\text{weight of meat before centrifugation (g)} - \text{weight of meat after centrifugation (g)}}{\text{weight of meat before pressing (g)}} \right] \times 100$$

Cooking loss

Cooking loss is calculated in accordance with the procedure of (Xia *et al.*, 2012) with slight modification.

Briefly, (10 g) of thaw samples are placed in heat-stable foil paper and held at 80°C for 30 minutes in the water bath until the center temperature has reached 70°C. To measure cooking loss, the samples was weighing before and after cooking.

Drip loss

Drip loss is determined by weight of thaw samples (10 g) and the values were measured. The samples were packaged in polyethylene containers, suspend at 4°C for 24 h, and weighed again. The new values were measured again. Drip loss is calculated as the weight loss proportion.

Thaw loss

Thaw loss is measured by measuring each sample before freezing and again after defrosting and dry with filter paper. Thaw loss is calculated as a proportion of the initial weight.

Chemical characteristics

Moisture content

The moisture percent was measured in accordance with the official procedure (AOAC, 2000).

Protein solubility

The protein solubility (mg/gm meat) in thaw sample is determine according to the procedure of (Lowry *et al.*, 1951), with standard of bovine serum albumin.

Thiobarbituric acid (TBA) value

To calculate the TBA value a solution containing 20% trichloroacetic acid (TCA) in 2M phosphoric acid was used according to the procedure mentioned by (Witte *et al.*, 1970). The absorbance was read by means of a spectrophotometer at 530 nm. [mg malondialdehyde (MDA)/kg muscle] was expressed as the TBA value. The results were determined by multiplying the absorbance by a factor of 5.2.

Determination of myoglobin concentration (Mb) and percent of met-myoglobin (M-Mb)

Myoglobin and Met-myoglobin were determined in accordance with (Krzywicki, 1982). To assess the myoglobin concentration in each sample, five gm of minced meat was used. The samples were extracted with a cold buffer of phosphate (0.04 M, pH 6.8) by 1:10 ratio of sample to buffer. Samples were homogenized for 15 seconds, and then the solution were centrifuged at 5000 rpm for 30 minutes. At 525, 572 and 700 nm, the absorbance of the filtered supernatant was read. Mb was measured by using the formula of Krzywicki (1982).

$\text{Mb (mg/kg)} = (A_{525} - A_{700}) \times 2,303 \times 5 (\text{dilution factor}) \times 1000$
and percentage M-Mb was measured using the formula of (Krzywicki, 1982).

$$\text{MMb (\%)} = 1.395 - ((A_{572} - A_{700}) / (A_{525} - A_{700})) \times 100$$

During the assay, samples were kept on ice at all points.

Sodium dodecyl sulfate- polyacrylamide Gel electrophoresis (SDS-Page)

Thaw beef samples (3 g) are homogenized for 60 seconds in 5% SDS (27 ml) at 11,000 rpm. The mixtures are incubated for 1 h at 85°C and then centrifuged for 10 minutes at 6100 x g. The protein content of the supernatant is measured using the biuret method and modified to 18 mg/ml using the method described in the literature. (Fritz *et al.*, 1989 and Laemmli, 1970). The supernatants are dissolved with a 1:1 (v/v) ratio of SDS-PAGE sample buffer containing [0.125 mol/L Tris-HCl pH 6.8, 4% (w/v) SDS,

10% (v/v) β-mercaptoethanol, 20% (v/v) glycerol and 0.005% (w/v) bromophenol blue] and boiled for 5 minutes. The samples (10 μl, 9 mg/mL) are load onto the 4% stacking and 12% separating gels and then subject to electrophoresis using a mini vertical apparatus. The gels are stained after electrophoresis with 0.125 percent (w/v) coomassie brilliant blue R-250 in 50 percent (v/v) methanol and 10 percent (v/v) acetic acid, and then discolored for 2-3 h with 50 percent methanol and 10 percent acetic acid until the background is clear.

Sensory evaluation

The pieces of beef muscle samples removed from frozen storage and thawing under various thawing methods before sensory evaluation. Then cooked for 20 minutes in an oven at 180°C to an internal temperature was reached to 75±1°C using a probe thermometer and served warm to several expert panel and meat scientists with previous experience. Assessment of sensory traits was conducted by means of the 5-unit descriptive scale used to assess color, flavor and aroma, tenderness, juiciness and overall acceptability. Panelists were expected to clean their palate with drinking water between samples. (Keeton, 1983).

Statistical analysis

The data from this experiment were analyzed according to analysis of variance (ANOVA) using the General Linear Model (GLM) within the statistical program Complete Randomized Design (CRD) procedures of XL-stat. (Addinsoft, version.5.03, 2016) in one-way ANOVA, and its significance was verified at the level of 5% using Duncan's multiple range test.

RESULTS AND DISCUSSION

Moisture content, water holding capacity (WHC) and cooking loss

Table 1. Shows the results of moisture content, WHC and cooking loss percent of beef sample, Which is thawed after freezing by five thawing technique. All the evaluated parameters significantly (p <0.05) were influenced by thawing methods. Tap water thawing obtained the highest moisture content and WHC by the rate (76.708% and 39.117%) respectively, followed by refrigerator thawing (76.676 % and 34.625%) respectively for moisture and WHC. While, microwave thawing recorded the lowest moisture content and WHC respectively by the rate (69.195% and 27.070%). Connection with cooking loss, also tap water thawing significantly (p<0.05) had the lowest value (15.784%), while the microwave thawing methods had the highest cooking loss value (39.324%). In this experiment, moisture content and WHC of the beef sample after thawing by tap water were comparatively higher than those of other thawing methods as a result of drip loss, thus with thawing temperature, thawing loss appeared to increase. However, short-term microwave thawing causes comparatively higher losses as it heats product quickly and evaporation is increases.

In the previous study by Kim *et al.* (2013) found that the reasons of drip loss and reduced moisture content, cooking loss, tenderness, lightness and redness of the meat may be related to the pork freeze-thawing. However, Pires *et al.* (2002) didn't observed any variation in moisture percent for samples of pork loin kept at frozen for 15 days

and defrosted at 7 ° C or 25 ° C. The highest loss of cooking by microwave thawing method may be related to the increment of protein denaturation, which caused water loss. Functional properties such as water holding capacity and proteins are lost by defrosted meat in addition to losing moisture, which can weaken the gelling of final meat-containing products (Olivo and Olivo, 2006). The data in this study are consistency with the results stated by Bustamante-Vargas *et al.* (2016) when evaluated the exudate loss of breast chicken meat submitted to various thawing technique, the results of the study observed that the greatest loss of exudate was observed of the microwave thawing method. Moreover, Zhu *et al.* (2020) demonstrated that tap water thawing recorded higher WHC comparatively with other thawing methods.

Table 1. Moisture content, water holding capacity (WHC) and cooking loss percent of beef loin samples subjected to different thawing methods

Thawing methods	Moisture %	WHC %	Cooking loss %
Refrigerator	76.676 ±0.209	34.625 ±0.599	27.882 ±0.546
Oven	75.454 ±0.24	30.202 ±0.52	36.418 ±0.669
Microwave	69.195 ±0.159	27.070 ±0.585	39.324 ±0.528
Tap water	76.708 ±0.357	39.117 ±0.664	15.784 ±0.547
Room temperature	75.419 ±0.199	31.970 ±0.038	34.489 ±0.783

Means with different superscripts in the same column are significantly different (p<0.05)

On the other hand, Ku *et al.* (2014) detected the highest cooking loss percent in microwave thawing method when evaluated the quality of ham freezing by electro-magnetic freezing and thawing by different methods. Meanwhile, Ambrosiadis *et al.* (1994) explained that fast defrosting by water immersion reduced exudate losses, while, resulted in higher losses by microwave thawing for 35 minutes to reach 0°C. The current study demonstrated that, tap water and refrigerator thawing techniques resulted of lower drip, thawing and cooking loss exudates, that was compatible with literature, which revealed the techniques that cause less damage to meat and lower losses of exudate using low temperatures and shorter cycles.

Drip loss and thawing loss

Drip loss and thawing loss alterations in the beef sample after thawing according to various thawing technique are seen in Figs.1. Based on thawing techniques, drip loss and loss of thawing after freezing presented significant (p<0.05) differences. Drip loss and thawing loss from beef loin with tap water thawing method revealed the lowest rate with (0.888% and 3.527%) respectively. While, microwave thawing obtained the highest drip loss and thawing loss rate with (2.202% and 6.164%) respectively. There were no significant difference (p<0.05) in the drip loss of beef samples with refrigerator (1.819%), microwave (2.202%), oven (2.003%) and room (1.894%) thawing methods. In terms of thawing loss, the values ranged from (6.164%) for microwave followed by room temperature (5.432%), oven (4.302%), refrigerator (3.527%) and tap water (3.527%) thawing methods respectively. This observation clarified that beef samples thawed by tap water

and refrigeration resulted in less thawing loss than other thawing methods.

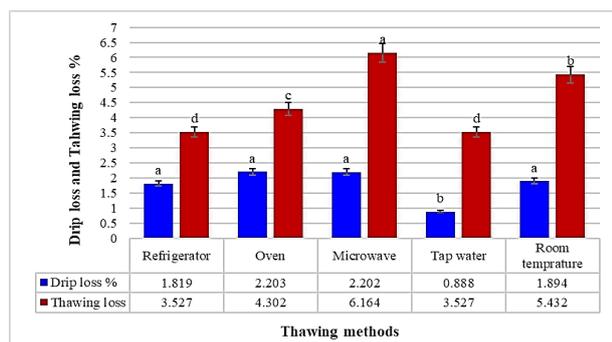


Figure 1. Drip loss and thawing loss percent of beef loin samples subjected to different thawing methods

High drop loss means that muscle fiber water-soluble losses which, indicates a loss of nutritional value. Therefore, we can show from the results of the current study if the meat thawed by the tap water, it will reduce drip loss from thawing, which thought to be lead to quality maintenance. The excessive release of liquid and, subsequently, nutrient loss, harming manufacturing profitability and deceiving customers are one of the major problems caused by meat thawing and cooking. On the other hand, the thawing process should be controlled because the raise in temperature stimulates the growth of microorganisms contained in meat, posing risks of food poisoning and increasing spoilage of the product. Due to the formation of large extracellular ice crystals by incomplete defrosting of frozen meat significantly induces quality degradation such as, lipid oxidation, protein oxidation, protein denaturation, and microbial growth during thawing (Xia *et al.*, 2012).

Compared to thawing in a microwave oven, chicken breast meat thawed under refrigeration and room temperature showed the better meat quality results, as seen by less product disruption (Colpo *et al.*, 2012), as indicated by (Colla and Prentice-Hernandez, 2003). On the other side, Xia *et al.* (2012) noted that when samples of pork were thawed in a refrigerator (4°C), the physicochemical characteristics closest to fresh samples and the lowest quality losses relative to other thawing techniques (room temperature, water immersion or microwave oven) were seen. However, Oliveira *et al.* (2015) observed that the highest drip loss was found in microwave thawing method when studied the quality of breast chicken meat submitted to various thawing technique. Moreover, Yu *et al.* (2010) recorded that by increasing drip loss and cooking loss by thawing, myofibril was disrupted and affects the quality of beef with the higher thawing rate.

However, Kim *et al.* (2006) indicated that drip loss was reduced and WHC was raised by using ohmic thawing at 250W microwave power in frozen pork meat. Furthermore, Bailey and James (1974) recommended changing the airflow rate therefore the findings of this study showed that after freezing, meat defrosted with tap water would minimize the thawing loss resulting from freezing and thawing, which is assumed to contribute to quality maintenance.

Protein solubility

Protein solubility is an important metric for meat quality and is closely connected to many other physical and

functional properties (Marcos *et al.*, 2010). The effect of thawing techniques on the solubility of protein in beef samples have been investigated in Fig.2. Significant differences ($p > 0.05$) were found in sarcoplasmic, myofibrillar and total protein concentration among thawing methods. Refrigerator thawing obtained the higher myofibrillar (82.210 mg/gm meat) and total protein (108.048 mg/gm meat) concentration. While, microwave thawing recorded the lower myofibrillar (71.089 mg/gm meat) and total protein (83.520 mg/gm) concentration. Reduction of protein solubility is an indicator for degradation of muscle protein and it is related with increased hydrophobicity and exudation of surface (Zhang *et al.*, 2017). Connection with sarcoplasmic protein, room temperature thawing had a higher protein concentration (40.463 mg/gm meat). Meanwhile, microwave thawing had a lower protein concentration (12.431 mg/gm meat). Already microwave thawing recorded the lowest concentration of protein, this observation may be related to the variation in thawing time for microwave thawing were just 5 minutes and the remain were more than 2 hours and it can also be clarified by the influence of the various processes of thawing approved on the meat structure. Also to the differences in thawing time. However, the connection between protein concentrations and the exudate percent released from meat and losses by drip and thawing. The greatest losses of exudate and protein were possibly due to microwave thawing due to the degree of protein denaturation and structural destroyed (Leygonie *et al.*, 2012; Ali *et al.*, 2015). The denaturation contributes to the protein's loss of water holding capacity, which causes higher exudate losses, even with short thawing time 5 minutes. These results are corresponding with the result found by (Bustamante-Vargas *et al.*, 2016) who reported the lowest protein concentration by microwave thawing method when studied the electrophoretic profile of chicken breast exudate has been subjected to various thawing processes.

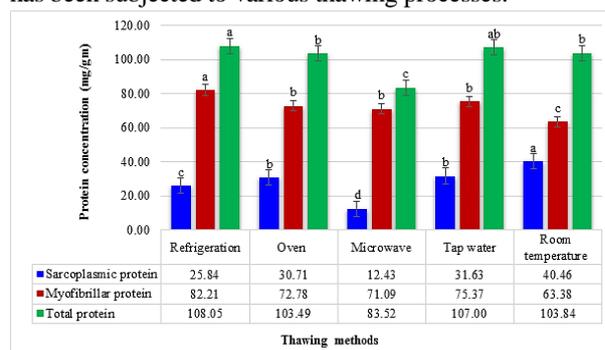


Figure 2. Sarcoplasmic, myofibrillar and total protein concentration (mg/gm meat) of beef loin samples subjected to different thawing methods.

Thiobarbituric acid (TBA), myoglobin concentration (Mb) and percentage of met-myoglobin formation (Met-Mb)

The influence of thawing techniques on TBA value, myoglobin concentration (Mb) and met myoglobin formation (Met-Mb) of beef samples are shown in Table 2. Significant changed ($p < 0.05$) were noticed among the samples submitted to various thawing methods. From the results clarified that significantly ($p < 0.05$) higher TBA

value (1.113 mg Malondialdehyde/kg meat) was measured for the samples thawed by microwave thawing while; the lowest value was obtained by tap water thawing (0.421 mg Malondialdehyde/kg meat). These observations are similar with the result by (Zhu *et al.*, 2020) who revealed that microwave thawing obtained higher TBA value in *longissimus dorsi* muscle of pork submitted to six microwave-based thawing technique.

For meat and meat products, lipid oxidation is an important quality parameter since it can lead to rancidity (Jin *et al.*, 2009; Nolsøe and Undeland, 2009). Microbial spoiling can be delayed when beef and beef products are processed under frozen conditions, but fat oxidation occurs and the beef components can be oxidized. Many researchers have identified that during the frozen and chilled storage of meat and meat products a general pattern of TBA value was increased (Devatkal *et al.*, 2004 and Rajkumar *et al.*, 2004). In frozen meats, fat oxidation continues at a slower rate than in refrigerated meats, according to (Tan and Shelief, 2002), and the TBA values presented only minor variations throughout frozen storage at -20°C for 69 days.

Table 2. Thiobarbituric acid (mg Malondialdehyde/kg meat) TBA value, myoglobin concentration (mg/gm meat) and percentage of met-myoglobin formation of beef loin samples subjected to different thawing methods

Thawing type	TBA	M-b	Met-Mb
Refrigeration	c 0.445 ±0.033	abc 3.962 ±0.061	ab 36.418 ±1.403
Oven	c 0.481 ±0.023	bc 3.913 ±0.047	b 33.043 ±1.09
Microwave	a 1.113 ±0.044	c 3.766 ±0.158	a 37.486 ±1.297
Tap water	c 0.421 ±0.014	a 4.218 ±0.05	c 23.828 ±0.887
Room temperature	b 0.843 ±0.025	ab 4.181 ±0.042	c 26.899 ±1.071

Means with different superscripts in the same column are significantly different (p<0.05)

The TBA value in this study ranges from 0.421 to 1.113 (mg Malondialdehyde/kg meat), so in this study, TBA values remained lower than the acceptable rancidity amount. Several factors affecting the oxidative stability of meat including anti and pro-oxidant balance and the composition of oxidation substrates of polyunsaturated fatty acids (PUFA), cholesterol, proteins and pigments (Bertelsen *et al.*, 2000). A rich source of such components is beef. In this experiment, microwave thawing showed the highest TBA value, since heating could influence several factors that are involved in oxidizing lipids. Heat interferes with the muscle cell structure and inactivates antioxidant enzymes, freeing oxygen from oxymyoglobin. The high temperature reduces the energy for activation oxidation and breaks down hydroperoxides into free radicals that proliferate lipid peroxidation. For the pre-frozen meat samples, heating appeared to be very pro-oxidative as assessed by the high TBA values.

Previous studies have documented that frozen storage and defrosting could lead to auto-oxidation of polyunsaturated fats under freezing conditions (Cheng *et al.*, 2019). TBA was much more affected by the thawing conditions than the freezing conditions. (Chun *et al.*, 2016). Thus, their higher temperatures of thawing and microbial action can clarify the relatively higher TBA values

measured for microwave thawing. The variations in meat color are used to determine meat quality during frozen storage and thawing. There were significant changes (p<0.05) in the myoglobin concentration and met-myoglobin formation were detected among thawing methods of beef samples. As shown in Table 2. Beef samples subjected to tap water obtained the highest myoglobin concentration (4.218 mg/gm meat) and lowest met-myoglobin formation (23.828 %). In contrast, microwave thawing recorded the lowest myoglobin concentration (3.766 mg/gm meat) and highest met-myoglobin formation (37.468 %).

Hughes *et al.* (2014) revealed that reductions in water holding capacity decreased the reflectivity of meat pigments, which in turn reduced the concentration of myoglobin. However, lipid and protein oxidation, microstructural changes, and drip loss can change quickly during thawing (Llave *et al.*, 2014). Conversely, significant changes in the meat color of microwave-thawed pork loin caused by non-uniform heating have been reported (Choi *et al.*, 2017).

SDS-page

The SDS-page electrophoresis analysis was performed to estimate the alteration in the protein composition of beef samples under various thawing technique. The results are demonstrated in Fig. 3. For the various thawing technique, significant differences (p<0.05) in the protein content of beef loin were observed between the bands. Several protein bands have steadily become thinner, lighter or even vanished. For the different thawing methods, high or low intensity and difference in bands were found, suggesting a variance in the size of proteins expressed in the samples from the various thawing technique (Refrigerator, Oven, Microwave, Tap water and Room temperature). The best findings were those showing the least number of bands and the lower intensity which, found with tap water and refrigerator thawing techniques. Meanwhile, the microwave thawing showed the worst results.

The bands which has larger intensity of the bands means the greater presence of several stronger bands, showed the higher decay to the defrosted meat, which prompts more prominent loss of myofibrillar and sarcoplasmic proteins, which can leads and liberation tiny peptides into the exudate. Myosin in the range below 205 kDa (Xia *et al.*, 2012), actin in the 45 kDa range, tropomyosin and troponin in the 40 to 45 kDa range, and short-chain myosin in the 25 kDa range are the bands that characterize myofibrillary proteins (Ali *et al.*, 2015). The sarcoplasmic protein characteristic bands have a wide distribution range of between 19 kDa and 110 kDa and are basically enzymes (Ali *et al.*, 2015). In the range between 30 kDa and 60 kDa, all defrosting techniques had greater or lower strength bands. The presence of actin was shown by the 45 kDa band and it indicated the presence of tropomyosin and troponin T between 40 and 45 kDa. Overall, weaker bands and a smaller number of bands were seen in the tap water and refrigerator thawing methods, suggesting less protein loss and less degradation due to the thawing process. The present study showed that the alteration in texture and structure of the network were linked to the protein degradation under various thawing methods. The findings of this study were compliant with those found by (Bustamante-Vargas *et al.*, 2016).

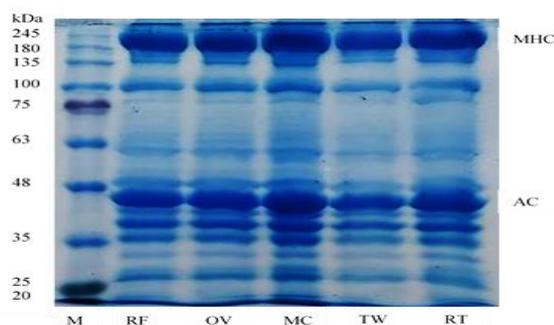


Figure 3. Changes in protein SDS-PAGE profiles of beef loin samples subjected to different thawing methods

M: marker, RF: refrigerator, OV: oven, MC: microwave, TW: tap water, RT: room temperature, MHC: myosin heavy chain, AC: actin

In the previous study by Bustamante-Vargas *et al.* (2016) demonstrated that when electrophoretic profile was performed on the chicken breast meat exudate, refrigerator and cold water thawing by fewer bands obtained the best results, and of lower intensity, in other words, the worst results were shown by microwave and oven thawing techniques. Several experiments were performed on the SDS-page by earlier researchers. After various thawing methods, Xia *et al.* (2012) conducted the electrophoresis pattern of pork sample and clarified no induced of protein aggregation and fragmentation resulting from thawing. However, in another study by the mentioned researcher, the electrophoretic profile showed variations in various freeze-thaw cycles that suggested aggregation of proteins and fragmentation. However, Ali *et al.* (2015) stated no variations between the bands corresponding to myosin (205 kDa), but several bands between 231 and 250 kDa were visible, and the size of the bands between 130 kDa and 86 kDa decreased in the size as the number of cycles increased. When conducted the electrophoresis pattern of the extract in chicken breast protein, which, has been subjected to freezing for one week at -20°C , and then six cycles of freeze-thawing for 12 hours at 4°C .

Sensory evaluation

Sensory evaluation was conducted according to the thawing techniques (refrigeration, oven, microwave, tap water and room temperature) for thawed beef samples after freezing to analyze the differences between thawing methods. The sensory evaluation of loin beef chops was determined for color, flavor, tenderness, juiciness and overall acceptability as shown in (Table 3). The findings revealed that there were no significant differences ($p < 0.05$) among thawing methods in sensory evaluation scores for color, tenderness and overall acceptability. Beef loin chops thawed by room temperature significantly ($p < 0.05$) recorded the highest score for flavor and aroma (4.143), however tap water thawing significantly ($p < 0.05$) indicated the highest score for juiciness (3.714). While, oven thawing method recorded the lowest score for flavor and juiciness (3.00 and 2.429) respectively. Although no significant differences ($p < 0.05$) were found in the sensory evaluation for the color, tenderness and overall acceptability by thawing methods which used. However, flavor and juiciness were higher with room temperature thawing. The higher juiciness score of tap water than other thawing methods may be related to the higher WHC and lower cooking loss in tap water thawing as compared to other thawing methods.

Table 3. Sensory evaluation of beef loin samples subjected to different thawing methods

Thawing method	Color	Flavor	Tenderness	Juiciness	Overall acceptability
Refrigerator	a 2.857 ± 0.459	ab 3.429 ± 0.369	a 2.857 ± 0.404	ab 3.000 ± 0.378	a 3.000 ± 0.488
Oven	a 2.429 ± 0.369	b 3.000 ± 0.218	a 3.000 ± 0.535	b 2.429 ± 0.202	a 3.286 ± 0.474
Microwave	a 2.857 ± 0.34	ab 3.286 ± 0.522	a 2.571 ± 0.369	ab 2.571 ± 0.481	a 3.000 ± 0.378
Tap water	a 2.857 ± 0.143	ab 3.571 ± 0.369	a 3.143 ± 0.34	a 3.714 ± 0.421	a 2.571 ± 0.369
Room temperature	a 3.286 ± 0.286	a 4.143 ± 0.143	a 3.714 ± 0.36	ab 3.143 ± 0.404	a 3.857 ± 0.34

Means with different superscripts in the same column are significantly different ($p < 0.05$)

CONCLUSION

It can be concluded that from the findings of the present study, all the methods of thawing have significantly affected the quality of beef meat. Generally, the best results were given by the tap water thawing technique, which resulted in lower drop losse, thawing and cooking losse with higher WHC and moisture content due to less damage to the cell structure. As well as this method, (tap water) decline fat oxidation in beef sample as a result of lowest TBA value and obtained better meat color than other thawing methods. The application of electrophoresis has proven to be a viable technique for assessing meat proteins. In addition, it was found that the samples with higher band counts, and greater intensity might be related to the higher losses, this suggests that the meat proteins suffered more damage from those samples, leading to fragmentation, structural loss and release of small peptides, affecting the nutritional quality and functionality of the meat. The preferred thawing technique is tap water as it retained sample consistency and quality; on the other hand, during freezing and thawing, it is the best way to improve the quality of beef meat and could be appropriate for the frozen meat industry as well as for those who consume its products.

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تأثير طرق الذوبان المختلفة على الخصائص الفيزيائية والكيميائية ونمط الترحيل الكهربائي والتقييم الحسي للعضلة الطولية الظهرية المجمدة لماشية اللحم

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في هذه الدراسة تم تقييم الخصائص النوعية والتي تشمل (الخواص الكيميائية والفيزيائية، الخصائص التركيبية والتقييم الحسي) للعضلة الطولية الظهرية لماشية اللحم التي تم تجديدها ومن ثم اذابتها بخمسة طرق اذابة مختلفة والتي تشمل (التبريد، الفرن، الميكروويف، ماء الحنفية وتحت درجة حرارة الغرفة). تم اذابة العينات الى درجة تصل فيها درجة الحرارة الداخلية للحم الى صفر مئوية، بعد ذلك تم تقدير عدة قياسات للحم والتي تتضمن القياسات الفيزيائية (الفقد اثناء التبريد، الفقد اثناء التجميد، الفقد اثناء الطبخ وقابلية اللحم على حمل الماء). بالإضافة الى الخصائص الكيميائية (محتوى الرطوبة، ذائبية البروتين وحمض الثيوباربيتوريك (TBA)، تركيز الميوغلوبين (Mb) وتكوين الميت ميوغلوبين (Met-Mb). علاوة على ذلك الترحيل الكهربائي لبروتينات اللحم والتقييم الحسي. أشارت النتائج الى أن عينة لحم الماشية التي تمت اذابتها بواسطة ماء الحنفية انتجت ادنى نسبة فقد اثناء التبريد والتجميد والطبخ، كما انتجت ايضاً اعلى محتوى للرطوبة وقابلية اللحم على حمل الماء WHC مقارنة بطرق الذوبان الأخرى. علاوة على ذلك، فإن طريقة الذوبان هذه (ماء الحنفية) تمنع أكسدة الدهون عن طريق تقليل قيمة TBA ونسبة تكوين الميت ميوغلوبين Met-Mb بالإضافة الى زيادة تركيز الميوغلوبين Mb. كذلك بينت نتائج الدراسة الى ان طرق الذوبان المختلفة اثرت معنوياً على نتائج الترحيل الكهربائي لعينات لحم الماشية، حيث وجد ان استخدام مايكروويف لاذابة اللحم ادى الى زيادة عدد الحزم وزيادة كثافة الحزم التي تظهر تلف بروتينات اللحم مما يؤدي الى تفتت اللحم وفقدان التركيب كذلك تحرم الحزم الصغيرة مما يؤثر على القيمة الغذائية ووظائف اللحوم. اضافة الى ذلك، فقد أظهرت بيانات هذه التجربة أن عينة لحم الماشية المذابة بالماء الحنفية تحافظ على جودة اللحوم وتماسكها بكفاءة وستساعد صناعة اللحوم وأولئك الذين يستهلكون هذه المنتجات. لذلك يمكن الاستنتاج الى أن خصائص جودة اللحوم مرتبطة ليس فقط بعملية التجميد، ولكن أيضاً بنظام وشروط الذوبان المستخدمة.