Effects of Marination with Different Salt Concentrations on Some Quality Criteria of Beef Meat

Buket ER DEMİRHAN*, Burak DEMİRHAN

Faculty of Pharmacy, Gazi University, Department of Pharmaceutical Basic Sciences, 06330, Ankara/Turkey.
*Corresponding author: B. Er Demirhan E-mail: erbuket@gazi.edu.tr

RESEARCH ARTICLE

Abstract
In the present study, the effect of different salt concentrations on some quality properties such as the total number of coliform bacteria, texture, cooking loss, total salt content, metmyoglobin, pH, the water activity of steaks prepared from longissimus dorsi muscle of fresh beef was investigated. For this purpose, steaks prepared from longissimus dorsi muscle were dipped in marination brine in four different concentrations; 0% (control group), 1.5% NaCl, 2% NaCl and 2.5% NaCl, and marinated for 24 hours at +4 °C. The initial crude protein, crude fat, pH, moisture, and ash contents of beef were determined as 20.50%, 5.80%, 6.13, 72.63%, 1.07%, respectively. In the cooked meat samples, the hardness, chewiness and gumminess of the samples marinated with 1.5% and 2% NaCl was found to be higher than the control group. Metmyoglobin content of the meat samples marinated with NaCl is lower than the control group. The results revealed that the total number of coliform bacteria decreased drastically in parallel with the increase in salt concentration. As a result, the changes in the quality of the bovine longissimus dorsi muscle with different salt concentrations and marination were investigated, and positive effects on the textural properties and coliform bacterial load of the meat were observed.

Keywords: beef meat; marination; salt; quality.

INTRODUCTION
Red meat has an important role in healthy and balanced nutrition. Meat is one of the main sources of protein for the nutrition of human in the world and contains valuable nutrients such as vitamin B12 and other B vitamins, iron, zinc, selenium and phosphorus with high biological value (Pereira and Vicente, 2013; Zerabruk et al., 2019). In addition, fresh meat can spoil as a result of contamination from physical, microbial and chemical hazards (Soyiri et al., 2008; Okoronkwo et al., 2014). Meat and meat products provide an appropriate growth environment for bacteria, yeast and molds. Along with the hygiene and the acidity of the meat, storage temperature, and the structure of the muscle tissue are also factors that affect the deterioration of meat (Dave and Ghaly, 2011). Microorganisms that cause spoilage can reduce the quality of meat, a valuable source of protein, making it tasteless or making it a source of foodborne infection (Sofos, 1994; Okoronkwo et al., 2014). Therefore, various processing and preservation methods are applied to inactivate or inhibit microbial growth to lengthen the product’s shelf life while maintaining flavor and safety (Sofos, 1994). Sodium chloride (NaCl or salt) is among the most important food additives widely used in the production of meat products due to its positive effect on water holding capacity, suppression of microbial growth, reduction of water activity, increased solubility of some proteins and typical salt taste (Sebranek and Fox, 1991; Sebranek, 2009; Kunová et al., 2015; Mariutti and Bragagnolo, 2017; Bae et al., 2018). Also, salt has an
effect on physicochemical properties and sensory properties (Aaslyng et al., 2014; Kunová et al., 2015). Salt is considered as a safe and sterile food additive that is widely used in the world (Biango-Daniels et al, 2018). Traditionally, salt has been evaluated as a food preservative additive that improves human health by limiting the growth of foodborne pathogens and spoilage microorganisms or by killing these microorganisms (Gordon and Barbát, 1989). In order to prevent the development of pathogenic microorganisms such as *Escherichia coli*, *Clostridium botulinum*, *Salmonella* and *Listeria monocytogenes*, salt is added to foodstuffs to reduce the water activity of foods (Sofos, 1984; Wirth, 1989; Kim et al, 2018). For these reasons, salt is one of the common and important components used in the marination of meat and meat products (Yusop et al, 2011). One of the common ways to add salt to meat and meat products is to marinate meat, and salt marination is increasingly used in the meat industry (Björkroth, 2005; Perisic et al, 2013).

The aim of the present study was to evaluate the effect of different salt concentrations on some physical, chemical properties and total coliform bacteria counts of steaks prepared from *longissimus dorsi* muscle of fresh beef.

**MATERIALS AND METHODS**

**Materials**

Fresh beef meats (*longissimus dorsi*) were obtained from a local market (Ankara, Turkey). Excess fat and connective tissue of the meat were separated and cut into slices about 1 cm thick. The meat sample was transferred to the laboratory under the cold chain. The marinade formulations contained different concentrations of salt (0-control group, 1.5% NaCl, 2% NaCl and 2.5% NaCl). Meat slices were dipped into marinating solutions and marination was carried out at 4°C for 24 hours. Then, marinated meat samples were used for physicochemical, texture and microbiological analyses.

**Proximate analysis**

Proximate composition (crude protein, crude fat, moisture and ash content) was determined according to AOAC methods. The ash content in the initial sample was determined using the AOAC method (AOAC, 2005a). Meat samples were weighed between 3-5 g in ash crucibles. Ash crucibles containing meat samples were then placed in the muffle furnace and burned until a light gray ash color or constant weight was obtained after the temperature reached 550 °C by operating the muffle furnace. After the incineration process, the ash crucibles were cooled in a desiccator and the samples were weighed and the ash contents were calculated. Total nitrogen measurement of initial sample was conducted based on the Kjeldahl method (AOAC, 2005d). Protein nitrogen content was obtained by multiplying total nitrogen by 6.25. Total fat of initial sample was extracted with petroleum ether (40–60°) following the AOAC, method (AOAC, 2005b). The moisture of initial sample was determined according to AOAC method (AOAC, 2005c).

**pH measurement**

pH value in meat samples was determined according to AOAC method. Briefly, 10 grams of meat samples were weighed and 100 mL of distilled water was added and homogenized with Ultra Turrax® T25 (IKA Labortechnik, Germany). After homogenization, the pH measurements of samples were carried out with the calibrated pH-meter (Hanna HI 221, Romania) until they were stable.

**NaCl analysis**

Briefly, the actual salt contents (NaCl) of samples were determined using a titrimetric method. Meat samples were weighed approximately 10 g and placed in Erlenmeyer flasks. Weighed samples were dissolved in 100 mL of distilled water. 2 mL of K₂CrO₄ was added to the solution and the solution was titrated with AgNO₃ until the first permanent color of red Ag₂CrO₄ was observed. The amount of salt in the sample was calculated from the amount of silver nitrate used in the titration (Nagy et al., 2015).

**Water activity**

The water activity (a_w) of samples was determined directly using an AquaLab Series 3 water activity meter (Decagon Devices Inc., Pullman, WA, USA) (Fernández-Salguero et al., 1993).

**Microbiological analysis**

All the media for microbial analysis were prepared according to the manufacturer’s instructions. Four groups of beef samples from each salt level were analyzed at each sampling time. For microbiological analysis, samples were
weighed 10 g under sterile conditions and homogenized with 90 mL Maximum Recovery Diluent (Merck 1.12535) and stomacher (Bagmixer-400, France) for 1 minute. In the analysis, dilution was performed in serial dilutions with the maximum recovery diluent by calculating the ratio of 1/9. Total coliform bacteria were grown on Violet Red Bile Agar (Merck 1.01406) at 37 °C for 24 h (Halkman, 2005). The counting results obtained were expressed as log10 CFU (colony forming unit) g⁻¹.

Texture analysis
Texture profile analysis of the raw and cooked samples was conducted with a TA.XT2 texture analyzer (Surrey, England). Hardness, springiness, gumminess, cohesiveness and chewiness values of raw and cooked samples were determined by making an average of 3 measurements from each group.
The speed of the test: 1.0 mm s⁻¹
Waiting time between the two compression cycles: 1 s
Application time: 10 s
Trigger force: 0.01 N
The sample compression: 23.0%.

Metmyoglobin
Metmyoglobin was determined using the spectrophotometric method (Fu et al., 2017). A 2 grams of meat sample was weighed, then 20 mL of 0.04 M phosphate buffer solution (pH 6.8) was added and homogenized with ultraturrax at 12000 rpm for 20 seconds. The mixture was then taken into 50 mL falcon flask and centrifuged at 10000 g for 30 minutes at 4°C. The supernatant in the upper part was filtered through whatman no:1 filter paper. The filtrate was filtered again through a 0.22 µm PTFE filter and absorbance were measured by UV-Visible Spectrophotometer (Specord 50 Plus, Analytik Jena GmbH, Germany) at 525, 545, 565 and 572 nm.
Metmyoglobin content was calculated with the following formula;
MetMb (%) = (-2.541R1 + 0.777R2 + 0.800R3 + 1.098) * 100
R1: A572nm/A525nm
R2: A565nm/A525nm
R3: A545nm/A525nm

The cooking loss
Cooking loss in meat samples was made according to the method stated by Zahid et al. (2018). Meat samples were weighed before cooking and then it was cooked in boiling water for 30 min. After cooking, the percentage of cooking loss was calculated.
Cooking loss (%) = [(W1–W2)/W1] * 100
W1 is the weight of the raw meat sample
W2 is the weight of cooked meat sample

Statistical analysis
Statistical evaluation of the results was done by using SPSS statistical program. Data were presented as mean ± standard error. One-way analysis of variance (ANOVA) was used for the statistical evaluation of data, followed by Duncan’s and LSD post hoc test at p< 0.05 (Daniel, 1991).

RESULTS AND DISCUSSIONS
Proximate analysis, pH, NaCl and water activity
The moisture, crude protein, crude fat and ash values of raw meat were found as 72.63%, 20.50%, 5.80% and 1.07%, respectively. Proximate values obtained from raw longissimus dorsi are in approximate agreement with the literature (Önenç et al., 2004; Underwood et al., 2008; Arse et al., 2013; Ilavarasan et al., 2016; Dagne et al., 2021). It is stated that the protein content of M. longissimus dorsi varies between 21.23 and 22.34 g and 100 g⁻¹ (Kunová et al., 2015).

The quality of meat can be defined as a combination of many different characteristics such as color, water holding capacity, textural feature and acceptability by the consumer. The changes that occur in the muscle after slaughter are effective on these features. pH is an important factor in the quality of meat, and these biochemical changes are also affected by pH (Kunová et al., 2015). The initial pH value of the raw meat was found 6.13. The pH value of the control group was 6.50 and the pH values of the meat samples marinated with 1.5, 2 and 2.5% salt were determined as 6.28, 6.17 and 6.16 respectively. The salt used in marination may decrease the isoelectric point of the proteins in the meat and cause a decrease in pH value (Elias et al., 2020).
The salt content of the raw material was 0.37% at the beginning and after marination it was 0.26 in the control group. The salt ratio of the meat samples marinated with 1.5, 2 and 2.5% NaCl were determined as 1.32%, 1.54% and 2.14%, respectively.

The water activity values of the marinated meat samples were found to be low compared to the control group (Figure 2). It is stated that the salt used in meat products causes a decrease in water activity (Elias et al., 2020). In our study, it is thought that the water activity values of the samples marinated with NaCl were lower than the control group since the salt content of the marination groups was higher than the control group.

![Figure 1](image1.png)

**Figure 1.** The mean NaCl content of the sample group. Error bars represent ±standard deviations of the sample groups.

![Figure 2](image2.png)

**Figure 2.** The aw values of sample groups.

**Microbiological analysis**

The initial total coliform bacterial load was found to be 3.59 log CFU g\(^{-1}\). After marination, total coliform bacteria growth was observed only in the control group (3.97 log CFU g\(^{-1}\)). The total number of coliform bacteria in salt-marinated meat samples was considered to be less than 10\(^2\) CFU g\(^{-1}\).

**Texture profile analysis**

Meat texture, which is associated with edibility, mouthfeel, tenderness, and product yield, has an important place in determining the quality of meat. (Chang et al., 2010). Texture profile analysis (TPA) was performed on both raw and cooked samples.

The hardness is the maximum force obtained in the first compression (Chang et al., 2010). While the hardness of the control group was the highest in the raw samples, the hardness of the samples marinated with 1.5% NaCl was
found to be the highest in the cooked samples (Figure 3). The difference in the hardness values of the raw samples is not statistically significant \( (p > 0.05) \). In the cooked meat samples, the hardness of the samples marinated with 1.5% and 2% NaCl was found to be higher than the control group \( (p < 0.05) \). The hardness of the meat sample marinated with 2.5% NaCl was found to be lower than the control group \( (p < 0.05) \).

Cohesiveness shows how long the foodstuff can withstand a second deformation relative to its resistance under the first deformation. It is found by dividing the working area in the second compression sequence into the working area during the first compression (Nishinari et al., 2013).

The cohesiveness values of the groups marinated with NaCl in raw samples were found to be higher than the control group \( (p < 0.05) \). The resistance of marination groups to a secondary deformation in raw samples is higher than the control group (Figure 4).

![Figure 3. The mean hardness value of raw and cooked sample groups. Error bars represent ± standard errors of the sample groups.](image3)

![Figure 4. The mean cohesiveness values of the raw and cooked sample groups. Error bars represent ± standard errors of the sample groups.](image4)

The energy required for a semi-solid food to break down until it is swallowed is called gumminess. It is calculated by multiplying cohesiveness and hardness (Trinh and Glasgow, 2012). The difference between the gumminess values in raw samples is not statistically significant \( (p > 0.05) \). The gumminess value of the groups marinated with 1.5% and 2% NaCl in the cooked samples was higher than the control group, and the gumminess value of the group marinated with 2.5% NaCl was lower than the control group \( (p < 0.05) \) (Figure 5). It is understood that the meat samples marinated with 2.5% NaCl require lower energy during chewing after cooking compared to the control group.
Springiness is the degree to which the food sample returns to its original height after compression. It is also a rate or percentage of the original height of a product or food. Springiness refers to the height at which the food can recover at the end of the first bite and the beginning of the second bite. Springiness is the rate at which a deformed food returns to its undeformed state after the force causing the deformation is removed (De Huidobro et al., 2005). Springiness values of raw and cooked meat samples marinated with NaCl were found higher than the control group (Figure 6). The difference between the group marinated with 2.5% NaCl only in cooked samples and the control group was found to be statistically significant \((p < 0.05)\).

The chewiness is the amount of energy required to chew solid foods up to ingestion (De Huidobro et al., 2005). It is stated that the chewiness value should decrease in cooked foods (Latoch, 2020). In the analysis of cooked meat samples, the chewiness value of the group marinated with 2.5% NaCl was found to be lower than the control group \((p > 0.05)\) (Figure 7). The chewiness values of the cooked meat samples marinated with 1.5% and 2% NaCl were found to be higher compared to the control group \((p < 0.05)\). The difference between the chewiness values of the raw samples was not found to be statistically significant \((p > 0.05)\).
Metmyoglobin

For consumers, the important criterion of quality characteristics for fresh meat is color and it shows that red meat is fresh (Jeong et al., 2009). The meats color is related to the three forms of myoglobin, the purple red color is due to deoxymyoglobin, the bright red color (bright cherry red) is due to oxymyoglobin and the brown color is due to metmyoglobin (Dobbelstein, 2005; Fu et al., 2017). The metmyoglobin value of the control group was 32.62 and the metmyoglobin values of the meat samples marinated with 1.5%, 2% and 2.5% NaCl were determined as 14.45, 13.60 and 15.00, respectively (Figure 8).

The cooking loss

The cooking loss of the control group is 48.30%, and it is similar to the cooking loss value found by Aktaş et al. (2003) in the beef longissimus dorsi muscle in the control group. Similar to our study, Aşcioğlu and Şevik (2019) found the cooking loss as 42.76% as a result of boiling the beef longissimus dorsi muscle. The cooking loss of the meat samples marinated with 1.5%, 2% and 2.5% NaCl was 39.39%, 39.45% and 43.86%, respectively. Cooking loss was found to be lower in the meat marinated with NaCl compared to the control group. It is stated that NaCl increases the water holding capacity of the meat and therefore causes lower cooking losses (Aktaş et al., 2003).

CONCLUSIONS

Different salt concentrations used in the marination of meats have an inhibitory effect on the growth of microorganisms. Therefore, it can have positive effects on the quality and shelf life of the meat product. The use of NaCl, which is thought to be effective in preventing the growth of bacteria and pathogenic bacteria that cause deterioration in meat and meat products, is quite common. In addition to the antimicrobial effect of NaCl, when the
results of our study and other studies in the literature are evaluated, it is seen that the textural properties of meat are affected in different ways when the appropriate NaCl concentration is used to extend the microbial shelf life.

**Author Contributions:** B.E.D. Conceived, designed and performed the analysis, wrote the paper; B.D. Performed the analysis, collected the data and wrote the paper.

**Conflicts of Interest**
The authors declare that they do not have any conflict of interest.

**REFERENCES**


43. Yusop SM, O’Sullivan MG, Kerry JP. Marinating and enhancement of the nutritional content of processed meat products. Processed Meats, 2011; 421-49. https://doi.org/10.1533/9780857092946.3.421
