Quality and sensory attributes of tumbled or marinated beef jerky

by

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Abstract

Various methods are utilized by meat processors to make beef jerky. The most common methods utilized are tumbling or marinating meat. Each of these processing methods may influence jerky quality or sensory attributes. More research needs to be conducted to understand how these processing methods may affect the finished product. The objective of this research was to evaluate the quality and sensory characteristics of vacuum-packaged shelf stable beef jerky produced using tumbling or marination.

Twelve USDA Select beef inside top rounds (*semimembranosus*) were held at 2°C for 10 - 14 d before processing. On each processing day, whole rounds were trimmed, pH was measured, and the weights were recorded. Trimmed rounds were manually cut in half and each half was allocated to a tumbled or marinated treatment group. Before the treatments were applied, the beef round halves were sliced and weighed. Pieces from each half were held for moisture, fat, and protein determination, with separate samples allocated for transmission electron microscopy (TEM) and light microscopy (LM) to evaluate structural changes, sarcomere length (SL), and myofiber diameter (MD). After tumbling or marination, percent pickup was measured, and a piece from each half of tumbled or marinated rounds was held for sodium chloride content (SCC) analysis. Additionally, structural changes, SL, and MD were measured.

After thermal processing, samples were sorted into groups based on storage periods of day 0, 3 months, or 6 months as well as calculating the percent cook yield. Analysis on day 0 consisted of measuring the pH, moisture, and protein content, moisture protein ratio (MPR), water activity (a_w), instrumental color, shear force (SF), trained sensory evaluation, SCC, structural changes, SL, and MD. Furthermore, vacuum-packaged jerky treatments stored for 3
and 6 months were analyzed for instrumental color, trained sensory evaluation, aw, SF, SCC, moisture content, structural changes, SL, and MD.

Data was analyzed using a 3×2 factorial design where the fixed effects were the tumbled and marination processing methods of raw, raw tumbled, and raw marinated with storage periods at day 0, 3 months, and 6 months with individual rounds used as a random blocking factor. Sarcomere length and myofiber diameter results were analyzed separately based on the main effects of the treatment method.

There were similar (P>0.05) results for the percent cook yield while the percent pickup was higher (P<0.05) for the tumbling process versus the marination process. Analysis of the moisture, fat, and protein content showed no differences (P>0.05) among treatment methods within raw top-round samples. After the tumbling and marinating process, there was no difference (P>0.05) in the SCC among the raw top-round slice samples. After the beef jerky was thermally processed, there were no differences (P>0.05) for pH, aw, protein content, MPR, or SCC. However, the moisture content was higher (P<0.05) in tumbled jerky than in marinated jerky which could be related to the increase in percent pickup during the tumbling process.

The instrumental color values showed that L* values for tumbled beef jerky were lighter (P<0.05) in color and more red (P<0.05) in color for a* value than marinated jerky. Overall, jerky became darker (P<0.05) in color and more red (P<0.05) during storage. Shear force values were lower (P<0.05) in tumbled jerky with no change (P>0.05) over time. Sensory panelists found tumbled jerky to be less brittle (P<0.05), less chewy (P<0.05), and more flavorful (P<0.05) at day 0 and up to 6 months of storage at 20℃ compared to marinated jerky. Changes in structural integrity were observed due to processing methods and storage time. However, there was no difference between treatments or storage time for SL and MD. During storage, there was
a decrease ($P<0.05$) in SCC over time and an interaction ($P<0.05$) with process treatment and storage time for $b^*$ value. All the other variables of $a_w$, SF, moisture, and MPR were similar ($P>0.05$) for processing method and storage time. Overall, tumbling produced a jerky product that was more tender, less brittle, and more flavorful during 6 months of storage compared to marination as a processing method. Although tumbling yielded a higher percent pickup and jerky was darker in color than marinated jerky, the processing method did not influence SCC, $a_w$, or MPR of beef jerky.
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**Green Double Arrowed Line:** Cross-section diameter of myofiber

**Red Single Arrowed Line:** Endomysium

**White Single Arrowed Line:** Glycogen granules

**Yellow Single Arrowed Line:** Z-disc

**Red Double Arrowed Line:** Perimysium

**Red Circle:** Cellular debris

**Orange Double Arrowed Line:** M-line

**Black Double Arrowed Line:** I-band

**White Double Arrowed Line:** Distance of sarcomere

**Blue Double Arrowed Line:** Myofiber and myofibril tears

**White Closed Point:** Thick filament

**White Open Point:** Thin filament

**Pink Double Arrowed Line:** Myofiber and myofibril breakdown
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Dedication

I would like to dedicate this thesis to my parents Mike and Susannah. I am very thankful to have such loving, hardworking, supportive, and dedicated parents who have been there for me every step of the way. They have encouraged me to pursue my dreams despite the many obstacles that I have encountered in the last several years. If it wasn’t for them I would not be who I am today with the values that I instill within my everyday life. It has been a long road to get to where I am today but I am thankful to have parents who I can look to when I need support or advice. Thank you, Mom and Dad, for always pushing me to be the best version of myself in all that I do.
Chapter 1 - Literature Review

Introduction

Jerky is a product that can be made from any species of meat. It is a method that has been utilized for centuries, dating back to Egypt when humans needed a way to preserve meat from larger animals (USDA FSIS, 2016). The term “jerky” comes from the Spanish word “charque” which means meat that is dried (USDA FSIS, 2016). The USDA Food Data Central (2021) evaluated a nutrition facts panel on a beef jerky product and concluded that the jerky product had over 30 g of protein and contained other nutritional components such as calcium, carbohydrates, and iron. This beef jerky item had high protein which was associated with the Food and Drug Administration (FDA) Code of Federal Regulations (CFR) which states that for a product to carry a “high protein” claim, it must contain at least 20% or more of the recommended dietary intake (RDI) (FDA, 2023). The current Dietary Guidelines for America (DGA) set by the USDA DGA (2020) states that females at ages 19 or older, should consume around 46 g of protein a day. Likewise, men ages 19 or older should consume 56 g of protein a day. Since the 30 g of protein for the beef jerky is at least 20% of the recommended intake of protein it can be considered a high protein product.

There has been limited research evaluating how beef jerky production methods influence final product quality and sensory characteristics. However, a study by Choe et al. (2007) analyzed cured pork meat and the sensory properties of pork jerky. This study showed that tumbling resulted in a higher percent pickup than marination the longer the product was tumbled, but the authors did not find any differences among flavor, texture, juiciness, and overall acceptability for sensory panels. They found that $L^*$, $a^*$, and $b^*$ decreased over time the longer the samples were immersed or tumbled resulting in a darker product. This suggests that
differences can occur in jerky products due to production methods, impacting final product quality.

**Standards for Beef Jerky**

USDA FSIS has a standard of identity for the moisture protein ratio (MPR) for jerky. Jerky must be labeled by species or kind for ground and formed or whole muscle products (USDA FSIS, 2005). The MPR defines the moisture and protein content within a product and for a product to be labeled jerky it must have an MPR of 0.75 to 1.0 or less (USDA FSIS, 2005). Jerky compliance guidelines have been established to identify the key steps to make beef jerky, ensure the safety of the product, and provide scientific support for understanding the significance of the process (USDA FSIS, 2014).

Whole muscle beef jerky was commonly manufactured by being soaked in a marinade, tumbled in a tumbler, or a needle used to inject a marinade solution into the meat. Jerky products can be cured using nitrite or other curing agents, uncured, smoked using a smokehouse, or simply using an oven to cook and dry the product without smoke being introduced. If binders are used, the product name must contain the name of the binder if it is above 3.5% (USDA FSIS, 2005). However, if a binder is less than 3.5% in a formulation, the binder only needs to be listed in the ingredient statement and not within the product name (USDA FSIS, 2005).

**Economics of Beef Jerky**

Beef jerky is a product that has increased in popularity for consumers as a convenient on-the-go snack. From 2020 to 2022 there has been an increase of 1.4% in the U.S. in beef jerky consumption with projections that the consumption of beef jerky will continue to grow (Jerky Brands, 2023). This increase in consumption is projected to increase to 132.58 million people in the U.S. consuming beef jerky by 2024 with original flavored beef jerky, which is a standardized
flavor across the industry, being the number one preference for consumers (Jerky Brands, 2023). Research shows that for beef consumption in 2023, on average each household in the U.S. consumed 25 kg of beef products which included beef jerky (Jerky Brands, 2023) with the U.S. being the leading producer of beef jerky followed by Brazil and China (Berry, 2016).

Some consumers are focusing on what foods they consume when snacking, and choosing items that they consider healthy for them (Berry, 2023). According to Berry (2023), 18% of consumer respondents viewed jerky as a high-protein snack and 17% of consumer respondents viewed jerky as a healthy option. A study done by Convenience Store Decisions which is a trade publication found that 79% of consumers who participated in the study wanted to be able to better understand what ingredients are on the package (Joncheff, 2021). This relates to why some consumers are shifting towards wanting a healthier high protein option since they believe that if they can better understand what is in the product then they associate that as being a healthy option (Joncheff, 2021).

**Process of Tumbling**

A tumbler is a stainless steel barrel that rotates circularly for a defined period based on the product that is being utilized in the machine (Toldrá and Nollet, 2017). Tumblers are used in the production of beef jerky to accelerate protein extraction and increase the speed of curing if a curing agent such as sodium nitrite is used in the formulation. Most tumblers are designed to pull a vacuum to improve product yield by reducing the amount of air in the product. This allows the marinade to absorb within the product more easily to increase the flavor profile and binding of functional ingredients (Hui, 2012).

There are two main types of tumblers used in the meat industry for tumbling meat products. One type is a side load impact tumbler (Pearson and Gillett, 1996). A side-loaded impact tumbler
has baffles and a look similar to a cement mixer. The tumbler is angled and provides the advantage of limiting the amount of product that can fall once it gets to the top of the tumbler during rotation to reduce abrasion and structural damage (Hui, 2012). When each piece of meat in the tumbler moves, it interacts with other pieces of meat causing friction to be applied. The friction creates kinetic energy which is short mechanical compressions that result in structural impact on each of the pieces. This structural impact can start the breakdown process of myofibers and myofibrils within the cellular matrix (Toldrá and Nollet, 2017).

Another type of tumbler used in the meat industry is a multiple batch tumbler. This tumbler is automated and utilizes an inverted system to drop the meat when it gets to the top of the tumbler. It is recommended to have about a 0.30 m drop within the tumbler to cause the protein matrix to accumulate on the meat surface (Toldrá and Nollet, 2017). This results in a more pliable meat product allowing the brine to penetrate the cell membranes more easily (Toldrá and Nollet, 2017). Pearson and Gillett (1996) suggested that the multiple batch tumbler is the most efficient option to utilize for a streamlined and efficient process. The tumbler can speed up processing and result in more muscle-to-muscle interaction while still having rest periods where no tumbling occurs and the product sits idle, producing products with a stronger structural integrity which results in a less tender product (Pearson and Gillett, 1996).

**Process of Marinating**

Marination typically includes various ingredients and spices to create a brine that has different flavor profiles and an increase in tenderness. Marination is defined as “the act of submerging or keeping in a liquid for thorough wetting and softening for hydrotherapy” (Hui, 2012). The overall process is simple but provides less impact on the muscle structure than
tumbling, and can allow for improved tenderness, juiciness, and an increased flavor profile of the product (Hui, 2012).

An important factor to consider is the percentage of salt (NaCl) that is used within a marinade formulation that is added to the meat to ensure the strength of the brine penetrates the meat during immersion (Toldrá and Nollet, 2017). This immersion process is created by osmotic pressure that occurs between the marinade solution and the meat product allowing for a transfer of mass with the ingredients in the brine to migrate into the meat (Xu et al., 2023). A study by Xu et al. (2023) analyzed pork ham slices with various Gallic Acid (GA) levels to determine the effect of the chloride concentration on migration into the meat at various intervals throughout a 6 h period. The rate at which the GA penetrated the meat increased with more osmotic pressure being applied which eventually slowed down when the chloride level reached equilibrium (Xu et al., 2023).

Marination can increase product yield and moisture retention depending on the product. This immersion period can vary from a few hours up to a few days depending on the product. Gault (1985) found that products could be influenced by an acidic condition such as marination and affect the water-holding capacity of the product. They found that the lower pH would lower the level of tenderness in a product since it was not able to retain as much moisture. Additionally, when consistency is achieved within a product, marination can yield a more tender product, have more flavor, and be darker in color (Hui, 2012).

**Jerky Production Methods**

There are a variety of methods that processors use to produce whole muscle or ground and formed beef jerky (Lonnecker et al., 2010). Tumbling and marination are production methods used singly or in combination with dry or wet ingredients to add flavor and uniqueness to jerky
products. To ensure the safe production of jerky, USDA FSIS (2014) established compliance guidelines that ensure safety through the main critical operating parameter (COP) water activity.

Testing can be conducted to determine if jerky meets the criteria to be shelf stable. To be shelf stable, jerky should have a water activity of \( \leq 0.85 \text{ a}_w \) (anaerobic conditions – vacuum-packaged) or \( \leq 0.70 \text{ a}_w \) (aerobic conditions – not vacuum-packaged) (USDA FSIS, 2005). There are intervention methods that can be used during the production of beef jerky that aid in reducing the risk of pathogenic growth during storage. Preheating jerky strips in marinade to 71°C to reduce the risk of Salmonella growth, dipping the product in 5% acetic acid for 10 min, or using a mixture of calcium sulfate at a 1:2 or 1:3 ratios in water for 30 sec to reduce pathogenic growth during jerky production (USDA FSIS, 2014).

The jerky compliance guidelines outline specific production steps to ensure an understanding of the process to safely produce quality jerky. The top round (semitendinosus) is a common cut of meat used for beef jerky production that is either sliced into strips or ground using a grinder to later become a chopped and formed product. The meat that is used for a chopped and formed application is ground and then formed into strips (USDA FSIS, 2014). These meat pieces or strips are soaked in a marinade mixture or tumbled in a tumbler for a specific period of time to enhance the quality of the product (USDA FSIS, 2014).

The pieces are put onto racks before being put into a smokehouse for thermal processing. The initial cooking cycle starts with a surface preparation step lasting around 30 min where the strips are heated to a lower temperature to achieve a tacky surface (USDA FSIS, 2014). This allows for the smoke to adhere more easily to the product and improve product texture and color. There must be enough humidity within the smokehouse during this step because if the product dries out too quickly, the pathogens on the product can become more heat resistant (USDA FSIS, 2014).
The cooking step is considered the period beginning from when the product is put into the smokehouse, including the surface preparation step of the product becoming tacky until it reaches the end of cooking to ensure the product is thoroughly cooked. The COPs consist of a combination of relative humidity and product time temperature. Relative humidity relates to the dry bulb and wet bulb temperatures where the dry bulb indicates the air temperature within the processing unit. A wet bulb indicates the amount of cooling that occurs as moisture is removed from a surface. When there is 100% relative humidity in a smokehouse, the wet bulb temperature is equal to the dry bulb temperature. These temperatures are only accurate if the thermal processing unit is sealed with the dampers closed to prevent moisture loss during the cycle (USDA FSIS, 2014).

During thermal processing, a 5 log_{10} reduction of *Salmonella spp.* and Shiga toxin-producing *Escherichia coli* (*E. coli*) must be achieved during the lethality step (USDA FSIS, 2014). The goal of the lethality treatment is to kill pathogenic microorganisms utilizing critical operating parameters that utilize parameters such as water activity, relative humidity, and time temperature to ensure effective interventions have been applied to the product.

A post-dry heat step is utilized to increase the reduction in jerky pathogenic growth. Usually, it is dried for 10 min at 135°C to achieve a 2 log_{10} reduction (USDA FSIS, 2014). After ensuring the safety of the product, storage, and handling are important to ensure that no cross-contamination occurs. The product can be sealed in a vacuum-packaged bag or nitrogen-flushed sealed bag and put in boxes on shelves at 20°C for storage (USDA FSIS, 2014).
Impacts on Quality and Sensory

Shelf Life

Shelf life can be defined by the time and storage conditions where food remains safe to yield desired quality parameters for sensory, chemical, physical, and microbiological characteristics that meet specifications outlined by the company (Manzocco et al., 2010). The main factors that can influence the shelf life of jerky are oxidation and microbial safety (Manzocco et al., 2010). Beef jerky is not a high safety risk providing the water activity is \( \leq 0.85 \) and COPs were met during production (USDA FSIS, 2005). There is the potential that beef jerky can harbor \textit{Staphylococcus aureus}, \textit{Clostridium perfringens}, \textit{Listeria monocytogenes}, and \textit{Bacillus subtilis} (USDA FSIS, 2005). Holley (1985), found that the ideal temperature for the storage of thermally processed beef jerky to prevent pathogenic growth is 20°C.

Another factor that aids in extending the shelf life of beef jerky is the packaging utilized to store the product. Packaging has the primary purpose of being able to provide a physical barrier that protects the product from the environment while reducing the risk of microbial contamination or growth, oxidation, or color change to extend the shelf life of a product (Rosa, 2019). According to the USDA FSIS (2016), the estimated shelf life of beef jerky is 12 months based on storage at 20°C, and lipid oxidation that exceeds the threshold that consumers or trained panelists deem acceptable through sensory analysis.

Vacuum-packaging is commonly used for the storage of beef jerky with the addition of an oxygen scavenger to reduce the amount of oxygen present in the package and prevent other environmental factors from coming in contact with the product during storage. Since oxygen can speed up lipid oxidation, having as low of level as possible aids in a slower oxidation reaction and a higher quality beef jerky product (Li et al., 2022).
Yield

Yield is an important factor for the finished product to determine the weight change that occurs in a product after it has been thermally processed to maximize value and ensure consistency (Berry, 2016). Liu et al. (2017) analyzed the influence of dry time on beef jerky yield at 1 h increments up to 5 h. This data showed that the most optimal time for the highest yield was at 4 h. Since the heating process allowed for the product to dry out due to loss of moisture, it caused the myofibrils to weaken and lose water from the surface. The reaction decreased moisture and overall structural integrity to weaken the meat’s water-holding capacity (Lui et al., 2017). This is important because various ingredients such as salt can retain more moisture and affect the product yield when in beef jerky it should be relatively low since it is a dried product (Berry, 2016).

Proximate Analysis (moisture, fat, protein)

Typically, the moisture content for a raw beef top round is 72 - 73% with a fat content of 2.5 - 3.5% and the protein content is 23 - 24% (Acheson et al., 2015). Skaar and Boyle (2011) found the moisture content of thermally processed beef jerky to be 16 - 18%, protein content of 55 - 58%, and moisture protein ratio of 0.30. Yang et al. (2009) showed that moisture content in beef jerky tends to stay the same during storage and packaging such as vacuum-packaging which can play a role in retaining moisture content. Additionally, Miller et al. (1988) analyzed beef top round, beef heart, and beef tongue jerky in a comparative study where the top round had the highest moisture at 81.6%, and was in between the heart and tongue at 3.6% fat.

pH

The pH of beef jerky is 4.72 - 6.73 according to Jose et al. (1994). Typically, it is optimal to have it be around 5.4 - 5.8 for beef jerky to be considered shelf stable to reduce the risk of
microbial growth during storage (Yang et al., 2009). Okonkwo et al. (1992) showed that although the pH value tended to stay the same during extended periods of storage there was a slight decrease that could occur in beef products. This consistency within the pH related to the water activity during storage is critical for safety in terms of microbial growth of the product for human consumption (Yang et al., 2009).

**Water Activity**

Water activity is a measurement of the concentration of water and availability of water based on the total concentration of dissolved substances in a product due to its ability to bind to water for pathogens, bacteria, or microorganisms to grow usually ranging from around 0.65 \( a_w \) to 0.90 \( a_w \) and should be \( \leq 0.85 a_w \) to be considered vacuum-packaged shelf stable beef jerky (USDA FSIS, 2014). This water activity level is based on the growth limits for *Staphylococcus aureus*, and whether oxygen is present (ICMSF, 1996). According to ICMSF (1996), the water activity limit for *Staphylococcus aureus* growth is 0.83 \( a_w \) under aerobic conditions and 0.90 \( a_w \) under anaerobic conditions for optimal growth conditions. However, a lower water activity decreases the risk of pathogen growth. Functional ingredients such as salt compete with available water to increase the safety and functionality of the product.

**Sodium Chloride**

Sodium chloride is utilized as an important method to preserve food products due to its antimicrobial properties (Petran et al., 1989). It has been shown that high levels of SCC which contain 20% of the daily value for human consumption or more have anti-listeria properties to dehydrate and lower the water activity of beef jerky products (FDA, 2021). Some researchers think that it is not due to dehydration, but instead is due to the interference of substrates to stop the cellular function in the product (Woods and Wood, 1982; Erecinska and Deutsch, 1985;
Smith et al., 1987; Csonka, 1989). A study done by Parfentjev and Catelli (1964) showed that unless a product contains 10% or more SCC, there may not be an effect of the SCC to reduce the level of *Staphylococcus aureus* presence in the sample. There is more work that could be done as the results in the study by Parfentjev and Catelli (1964) proved to be variable.

**Instrumental Color**

Instrumental color can be measured using a colorimeter which measures tristimulus values, also known as International Commission on Illumination (CIE) \( L^* \), \( a^* \), and \( b^* \) (AMSA, 2012). The \( L^* \) value measures a spectrum of 0-100 with black being zero and white being 100, \( a^* \) value measures a spectrum of negative to positive with green being negative and red being positive. The \( b^* \) value measures a spectrum of negative to positive with blue being negative and yellow being positive. These values can be used to calculate hue which has deeper saturation and lightness properties to display the brightness or darkness of color. Chroma has a lower saturation level with colors that are more pale or dull (AMSA, 2012; Mancini et al., 2020). It is important to keep the aperture size consistent within a study and utilize the largest size to obtain the most accurate and widespread readings (AMSA, 2012). Additionally, a study by Han et al. (2023) analyzed beef jerky storage after high hydrostatic pressure processing to see the effects on the jerky after 60 d. The data showed that in terms of color, the \( L^* \) value decreased the longer the product was stored which was similar for the \( a^* \) and \( b^* \) values.

**Shear Force**

The shear force (SF) method was developed by Shackelford et al. (1999) to analyze tenderness and is considered a more efficient alternative to Warner-Bratzler shear force (WBSF) as it was implemented on production lines to guarantee tenderness of a product before it was shipped to a consumer. Nevertheless, Yang et al. (2009) showed that during storage the shear
force values change over time in beef jerky to become lower resulting in a more tender product. Additionally, finished product tenderness of bovine muscles can be affected due to cooking, quality grade, amount of connective tissue, or the type of muscle (Belew et al., 2003).

**Sensory Analysis**

Research has shown that beef tenderness is the most significant factor consumers consider when eating meat (Savell et al., 1987). This means that consumers are willing to spend more money to make sure that the products they are consuming will meet the satisfaction they desire in terms of tenderness (Miller et al., 2001). Additionally, Wheeler et al. (1994) found that the tenderness factor has to do with the level of marbling in the product. Since the top round (semimembranosus) has a lower level of marbling and beef jerky is cooked and dried to a more significant degree than a steak it causes the tenderness factor to be vastly different. This idea was solidified by the research done by He et al. (2018), Gatellier et al. (2010), and Sante-Lhoutellier et al. (2008) where meat increases in toughness as it cooks when a higher heating method is used such as a smokehouse. A study by Żochowska-Kujawska et al. (2017) conducted a sensory evaluation on home style beef jerky which found that utilizing the technique of marination yielded a higher tenderness, chewiness, juiciness, and connective tissue perception was perceived by trained panelists.

A factor that is important in terms of eating experience for beef jerky is flavor. According to Legako et al. (2015) and Kerth et al. (2015), it is one of the most complex palatability traits since it has volatile compounds and consumers utilize retro-nasal olfaction during their eating and cooking experience. The volatile compounds are developed through either thermal lipid degradation or Maillard browning where non-enzymatic is more prevalent since it is visual on the outer surface and occurs after lipid degradation (Kerth et al., 2015). Another study by Miller
et al. (1988) focused on beef jerky made out of top rounds, beef heart, or tongue that was assessed by consumers which found that top rounds provided the best flavor desirability and visual appearance.

**Skeletal Muscle Structure**

Skeletal muscle is morphologically characterized as nucleated; elongated cells that are arranged in parallel arrays. These elongated cells are known as muscle fibers where each muscle fiber is surrounded by connective tissue called the endomysium (Mense and Gerwin, 2010). The endomysium contains capillaries with small nucleated cells called satellite cells. Satellite cells are classified as the adult stem cells of the skeletal muscle (Mense and Gerwin, 2010). Satellite cells are found between the sarcolemma and basal lamina to assist with the process of repairing, growing, and regenerating muscles (Frontera and Ochala, 2015). Muscle fibers are surrounded by the lipid trilaminar structure, as observed by Transmission Electron Microscopy (TEM), known as the plasma membrane. A muscle fiber contains the sarcoplasmic reticulum, T-tubules, mitochondria, glycogen granules, and myofibrils (Brooks et al., 2023).

A sarcomere is a functional contractile unit within myofibrils. Sarcomeres contain both thick (myosin, 2 heavy, and 4 light chains) and thin (actin, tropomyosin, and troponin) filaments that are arranged in distinct bands (Brooks et al., 2023). These bands (A, I, and H) are illustrated in Figure 1. The A-band contains overlapping thick and thin filaments. The I-bands only contain thin filaments and are bisected by the Z-disc. The Z-disc are electron-dense bands that run perpendicular to the long axis of myofibrils and help anchor the thin filaments in place. Alpha-actinin is one of the proteins found in the Z-disc that anchors thin filaments (Brooks et al., 2023). The H-band contains only thick filaments and spans over the M-line which is in the middle of the sarcomere. M-lines are composed of proteins such as myomyosin that anchor thick filaments in
place (Frontera and Ochala, 2015). The Z-disc defines the end of each sarcomere within the myofibrils. During muscle stretching or relaxation I-bands and H-bands are increased in length which corresponds with increased length of sarcomeres. During contraction, sarcomere lengths are shortened resulting in shorter I-bands and H-bands (Frontera and Ochala, 2015).

**Figure 1.** The skeletal muscle structure and sarcomere structure of meat (College, 2017).

**Meat Structure**

Light microscopy (LM) and (TEM) are imaging techniques that can be used to assess structural and ultra-structural changes that might occur during the processing and storage of beef jerky. These techniques can be used to analyze a variety of samples such as raw, tumbled, marinated, and beef jerky during storage. The use of LM and TEM can assess if structural changes have occurred over time and what type of treatment was applied to the product. It has been shown by Bhat et al. (2018) that tumbling has more effect on the myofiber structure than the marination process due to constant friction during processing on muscle proteins that aid in disruption and solubilization. The impact that additional storage has on myofibrillar and stromal
proteins can aid in the promotion of degradation when tumbling is added and no brine is utilized. This results in a longer storage period to attempt to degrade the myofibrillar proteins when a brine is not introduced (Nondorf and Kim, 2022, Tuell et al. 2021, Tuell et al. 2022).

Nondorf et al. (2022) observed only slight structural changes of disrupted muscle structures in tumbled samples as compared to non-tumbled samples which showed more disruption to the muscle structures. They attributed the muscle structural changes to the storage of loins from cull cows (M. longissimus lumborum) due to tumbling for 90 min with or without a spike mat which utilizes spikes around the wall of the tumbler. This was attributed to how the tumbling caused fragmentation of myofibrils and connective tissue was not affected for intramuscular. Loins (5 d postmortem) were stored for up to 0 d or 14 d in clear vacuum-packaged bags. Proteolysis and tenderness were associated more with meat storage than the tumbling process (Nondorf et al., 2022).

Feng et al. (2020) did a study on fresh meat from brown bull calves to assess structural changes during the storage of the semitendinosus, longissimus thoracis, rhomboideus, gastrocnemius, infraspinatus, psoas major, and biceps femoris. They found that fresh samples seemed to hold the integrity of their structures, but once meat was stored for up to 7 d in polyolefin vacuum-packaged bags, the myofibrils diminished. Feng et al. (2020) stated it was difficult to see the integrity of the A-band, Z-disc, or I-band as they were more disordered but undamaged after the 14 d storage period due to the cracking of muscle fibers and shortening of sarcomeres. There is limited research on structural changes in jerky stored for longer than 30–60 d that could be further explored to understand the structural changes that occur during storage.
Conclusion

Beef jerky is a product that is rising in popularity and has increased in consumption each year. However, there is a lack of research that assesses the diversity of processing methods used for jerky manufacturing to better understand the impact on quality and sensory attributes. This leads to the need for further research to be done on common beef jerky processing methods to understand if differences among treatments occur and if so what are those differences. Therefore, the objective of this research was to evaluate the quality and sensory characteristics of vacuum-packaged shelf stable beef jerky produced using tumbling or marination.
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Chapter 2 - Quality and Sensory Attributes of Tumbled or Marinated Beef Jerky

Abstract

Twelve USDA Select beef inside top rounds (*semimembranosus*) were held at 2°C for 10 - 14 d before processing. Each of the 12 rounds was trimmed and weighed, and pH was measured before being manually cut in half. Once halved they were divided into two treatments of tumble or marinade before being sliced into 3 mm slices using a slicer and weighed. These raw samples were later analyzed for moisture, fat, and protein content. Transmission electron microscopy (TEM) and light microscopy (LM) were used to assess structural changes, sarcomere length (SL), and myofiber diameter (MD). After tumbling or marination, percent pickup, sodium chloride content (SCC), structural changes, SL, and MD were determined. After thermal processing, percent cook yield, water activity (a_w), pH, moisture and protein content, moisture protein ratio (MPR), color, shear force (SF), sensory, SCC, SL, and MD were measured. Vacuum-packaged jerky was stored at 20°C and evaluated after 3 and 6 months of storage for color, sensory, a_w, SF, SCC, moisture, structural changes, SL, and MD.

There was no difference (*P* > 0.05) in pH, moisture, fat, or protein content among raw top round samples, as well as no difference (*P* > 0.05) in SL and MD. After tumbling and marination, there was no difference (*P* > 0.05) in the SCC, SL, or MD samples. The percent pickup was higher (*P* < 0.05) with an increase of 58.4% in tumbled versus marinated slices. After thermal processing (day 0), there was no difference (*P* > 0.05) between treatments for percent cook yield, pH, a_w, SF, protein, MPR, or SCC, SL, and MD. The moisture content was higher (*P* < 0.05) with an increase of 51.52% in jerky produced using tumbling. Jerky produced using tumbling had lower (*P* > 0.05) *L* with a 6.71% change and higher (*P* > 0.05) *a* with a 23.79% increase compared to marinated
beef jerky. Regardless of the processing method, after 6 months of storage beef jerky was darker (P<0.05) with a 17.45% decrease and became redder (P<0.05) with a 17.45% increase compared to jerky at day 0 and 3 months of storage. Shear force values were lower (P<0.05) with a 18.16% decrease in tumbled versus marinated beef jerky. The trained panelists found that tumbled jerky was 20.75% more tender (P>0.05), 15.3% less brittle (P>0.05), and 14.62% more flavorful (P>0.05) than marinated jerky. Tenderness and flavor declined (P<0.05) at 3 months of storage and texture became more brittle (P<0.05) at 6 months of storage. There were visual differences and a decrease (P>0.05) in muscle structural integrity in raw top rounds, raw marinated or tumbled slices, and jerky from day 0 to 6 months of storage. There was a 20.70% decrease (P<0.05) in SCC during storage. The b* value decreased (P<0.05) by 37.03% for tumbling during storage time but did not change (P<0.05) for marination. All the other variables of a_w, SF, moisture, and MPR were similar (P>0.05) for the processing method and storage period.

Overall, tumbling produced a jerky product that was more tender, less brittle, and more flavorful during 6 months of storage compared to marination as a processing method. Although tumbling yielded a higher percent pickup and was darker in color than jerky produced using marination, the processing method did not influence the SCC, a_w, or MPR of jerky produced.

**Introduction**

In 2022, the gross income from beef jerky sales was $1.21 billion in the U.S. and sales are expected to have a 1.2% increase over the next three years (Hiner, 2021). USDA FSIS (2005) has regulations requiring a product that is labeled as jerky to have a moisture protein ratio (MPR) of 0.75:1 or less. If the product meets that criterion, processors should also follow the jerky compliance guidelines set by USDA FSIS (2014) on how to manufacture jerky so it is safe to
consume. Typically, if jerky is produced following the guidelines, and has a water activity value of 0.85 or less, it can have a shelf life of up to 12 months (USDA FSIS, 2016).

A survey by Lonnecker et al. (2010) of 37 Midwestern small plants that produced beef, pork, turkey, or buffalo jerky found that 56% of the respondents made whole muscle jerky and 44% of the respondents made ground and formed jerky. Jerky samples were provided by processors to Lonnecker et al. (2010) and were measured for sodium chloride content. These authors found that the salt content of these samples ranged from 2.10 – 11.54%. Additionally, the marinade incorporation ranged from 1-85% and percent pickup ranged from 3-100%. A smokehouse was used by 92% of respondents while the remainder used a commercial oven or an oven in combination with a smokehouse (Lonnecker et al., 2010). During thermal processing, 35% of respondents used dampers to control relative humidity while the remainder used steam injection, direct addition of water, or a combination of these methods. Vacuum-packaging was used to package beef jerky by 78% of respondents with the rest using no vacuum or gas-flushed plastic bags. For storage of the packages, 32% refrigerated jerky, 38% kept jerky at 20°C and 3% froze their jerky (Lonnecker et al., 2010). The overall conclusion from this survey was that processors exhibited diversity in the processing of products which caused variability in production. This presented the need for understanding what differences looked like in terms of quality and sensory attributes when different processing methods such as tumbling or marinating were used with a lethality step to produce jerky.

A study by Skaar and Boyle (2011) analyzed whole-muscle beef jerky produced using tumbling or marination. Beef strips were marinated for 24 h or vacuum-tumbled for 20 min. They found that the tumbling process was able to increase the flavor intensity of the product, and
tumbled jerky had a lower protein content and a lower water activity (a_w) than jerky produced using marination.

A recent study by Nondorf et al. (2022) demonstrated changes in fresh meat structure after tumbling, not tumbling, or tumbling with a spike mat which consisted of having a layer of spikes adhered to the inside walls of a tumbler to aid in tenderization of the beef strip loins (M. longissimus lumborum) from 12 Holstein cows. This study found that there were interactions between the different tumbling methods and storage periods suggesting that the utilization of tumbling on fresh beef could be a streamlined and effective method to improve the tenderness of the product.

Overall, it is important to understand that since there is a rise in the demand for beef jerky processors should follow the guidelines outlined by the USDA FSIS to create a safe and quality product. The processing methods may impact the overall structural component, safety, and quality parameters of the product. The objective of this research was to evaluate the quality and sensory characteristics of vacuum-packaged shelf stable beef jerky produced using tumbling or marination.

**Materials and Methods**

**Experimental Design**

This study used 12 USDA Select beef inside top rounds (semimembranosus) that were stored in a non-barrier shrink bag held at 2°C for 10 - 14 d before processing. On each processing day, whole rounds were trimmed, pH was measured and the weights were obtained before and after trimming. Trimmed rounds were manually cut in half, and each half was allocated to a tumbled or marinated treatment. Before the processing treatments were applied, each beef round half was sliced using a slicer (Treif Puma Slicer, Shelton, CT) into 3 mm slices and then
weighed. Pieces from each half were collected for determination of structural analysis, sarcomere length (SL), and myofiber diameter (MD) using transmission electron microscopy (TEM) and light microscopy (LM) and for proximate analysis. After tumbling or marinating, percent pickup was measured following a 5 min rest period, and a sample from each half of tumbled or marinated rounds was held to measure sodium chloride content (SCC), structural analysis, SL, and MD.

After thermal processing, samples from each treatment were vacuum-packaged and sampled initially on day 0 and after 3 and 6 months at 20°C. Cook yield was determined after thermal processing. On day 0, the pH, moisture, and protein content, $a_w$, instrumental color, shear force (SF), sensory evaluation, SCC, structural analysis, SL, and MD were measured. After 3 and 6 months of storage instrumental color, sensory evaluation, $a_w$, SF, SCC, moisture, structural analysis, SL, and MD were measured.

**Beef Jerky Production**

Beef rounds were trimmed and weighed and pH was collected using a pH probe (Hanna Instruments, Smithfield, RI) before being manually cut in half, and sliced using a slicer (Treif Puma Slicer, Shelton, CT) parallel to the muscle fiber orientation into 3 mm slices. Once the top round was sliced, raw samples were allocated for structural analysis, SL, and MD.

The tumbling process consisted of slices from one-half of each of the 12 rounds put into 16 cm × 24 cm heat-sealed bags (Uline, Pleasant Prairie, WI). A proprietary marinade mixture with a pH of 4.70 was added at a ratio of 1,361 g marinade per 3,175 g of meat. This mixture consisted of soy sauce, Worcestershire sauce, water, and spices. After the addition of marinade, the bags were heat sealed and put in a vacuum LT-15 tumbler (Lance, Hartford, Wisconsin). A
vacuum was pulled between 10 – 15 Hg. Tumbling was continuous for a 30 min period at 20 rpm in a refrigerated room at 2°C.

The marination soaking process consisted of slices from one half of each round placed into lugs. An individual lug contained slices from one-half of a given round. Each lug contained the same marinade as the tumbling process. Slices were completely submerged in the marinade for 24 h at 2°C a day before the tumbling process so all samples could be thermally processed at the same time.

After tumbling and marination, the slices were removed from the bags and laid on metal trays with metal racks to drain for 5 min before being weighed to calculate percent pickup. Samples that were allocated for structural analysis, SL, and MD were pulled after collecting percent pickup.

The slices were placed on large metal racks on a smokehouse cart with slices from each round and treatment segregated by rack. Treatments were thermally processed in a smokehouse (Maurer-Atmos, Reichenau, Germany) for a 3 h proprietary cooking and drying cycle. Following thermal processing racks were removed, and treatments were cooled at 2°C on the rack for less than 2 h.

After cooling, treatments were put in 1 mil gusseted poly bags (Uline, Pleasant Prairie, WI) and tied for storage at 2°C overnight. The next day, treatments were weighed to determine cook yield and were then packaged into a 7.5 cm × 14.0 cm impermeable 3 mil thick clear pouches (water vapor transmission rate < 7.0 g / m², 90% relative humidity at 38°C, oxygen transmission rate < 70.0 cm³ / m², 65% relative humidity at 23°C, Sealed Transparent Vacuum Pouch, Jayshri Propack, Changodar, India), with two oxygen absorbers (Sorb-oo, Walton’s,
Wichita, KS), and sealed using a vacuum-packager (Multivac C500, Skokie, IL). The vacuum-packaged beef jerky was stored at 20°C in closed cardboard boxes for up to 6 months.

The samples that were used for proximate analysis for the raw, raw tumbled, raw marinated, and day 0, 3 months, and 6 months of storage were cut into small pieces, frozen in liquid nitrogen, and then put into a one-liter stainless container (Waring, Stamford, CT) that was cooled using liquid nitrogen to keep the product from sticking on a one-liter single speed base (Waring, Stamford, CT) to powder the samples. The powdered samples were then put into 7 ½ cm × 18 ¼ cm whirl pack bags (Uline, Pleasant Prairie, WI) and placed in a -80°C freezer (Fisherbrand, Waltham, MA) until analysis was completed within 2 weeks or less of each sample period. Additionally, jerky samples that were allocated for SF and sensory analysis at day 0, 3 months, and 6 months were collected at each storage period and stored in a -40°C freezer for up to 6 months until evaluated.

Proximate Analysis

Moisture Content

Moisture content was determined using the AOAC 950.46 oven drying method (AOAC International, 2000a). A 5 g powdered sample from raw, raw tumbled, raw marinated, and jerky treatments stored for day 0, and for 3 and 6 months were weighed in duplicate onto labeled pre-dried aluminum heating cups. The cups were placed in a drying oven (Fisherbrand, Waltham, MA) for 24 h at 100°C. After drying, samples were put into a desiccator to cool and reweighed. Moisture percentage was calculated using the following equation:

$$\text{Moisture} \% = \left( \frac{\text{wet weight} (g) - \text{dry weight} (g)}{\text{wet weight} (g)} \right) \times 100$$
Fat Content

Using the Folch method (Folch et al., 1957) for fat analysis, the powdered samples were weighed in duplicate into 15 ml sterile tornado tubes (Midsci, Fenton, MO) and 1.6 ml Milli-Q (MQ) water was added. Next, 4 ml of chloroform (CHCl₃, Fisher Chemical, Waltham, MA) and 4 ml of methanol (CH₄O, Fisher Chemical, Waltham, MA) were added to the samples. The tubes were capped and vortexed (Vortex-Genie 2, Fisher Scientific, Waltham, MA) for 5 sec before being shaken on a wrist action shaker (Burrell, Pittsburgh, PA) for 10 min. After shaking, 2 ml of 0.74% potassium chloride (KCl, Fisher Chemical, Waltham, MA) was added to each tube and vortexed for about 5 sec until fully mixed. The samples were centrifuged (Centrifuge 5810 R, Eppendorf, Hamburg, Germany) at 1,000 × g for 5 min. Next, 1 ml of the bottom chloroform layer was pipetted from the tube into pre-labeled 12 × 75 mm glass culture tubes (Fisherbrand, Waltham, MA). The tubes were then placed in a Reacti-Vap Nitrogen Evaporator (Thermo Fisher Scientific, Waltham, MA) under the flow of nitrogen. When the chloroform evaporated from the tube and no odor was present, tubes were put in a drying oven (Fisherbrand, Waltham, MA) for 30 min until completely dried. Tubes were weighed to obtain the final weight and the sample fat percentage was calculated using the following equation:

\[
\text{Fat} = \left( \frac{\text{weight of glass tube containing lipid} - \text{weight of empty glass tube}}{\text{weight of meat}} \times 4 \right) \times 100
\]

Protein Content

Protein content was determined using the TruMac N (Leco, St. Joseph, MO) following a modified version of AOAC method 992.15 (AOAC International, 2000b). The 0.5 g powdered samples were weighed out in duplicate into tared crucibles and loaded into the carousel to determine the percent nitrogen for raw top rounds and day 0 samples. The protein content was calculated using the following equation:
\[ \% \text{Nitrogen} \times 6.25 = \% \text{Protein} \]

**Moisture Protein Ratio (MPR)**

The moisture protein ratio (MPR) was calculated using moisture and protein values (Allen et al. 2007) for tumbled or marinated beef jerky using the following equation:

\[ \frac{\% \text{Moisture}}{\% \text{Protein}} = MPR \]

**pH Analysis**

The pH was determined in duplicate using powdered samples that were homogenized (Homogenizer 850, Fisher Scientific, Hampton, NH) in 50 ml MQ water at 10,000 rpm for 20 sec. The pH was measured with an In Lab Science Pro-ISM probe connected to a Seven Compact pH meter (Mettler Toledo, Columbus, OH) for day 0 samples. Additionally, the pH of each whole top round was manually measured in the middle of each top round using a pH probe (Hanna Instruments, Smithfield, RI) before fabrication with one measurement per round.

**Water Activity**

Water activity was measured using a modified version described by Harper et al. (2010) and an Aqualab 4TE water activity meter (Aqualab, Riverside, CA). Several small thick and thin pieces from each treatment were put into a plastic cup for \(a_w\) determination on day 0 and after 3 and 6 months of storage.

**Sodium Chloride Content**

To measure the sodium chloride content (SCC) of tumbled or marinated beef jerky, 1 g of powdered sample was measured in duplicate was placed into a 100 ml glass beaker. Next, 9 ml of boiled distilled water at 100°C was added and the mixture was stirred for 1 min, paused for 30 sec, and then stirred again for 1 min. The glass beaker containing the sample was cooled in a cold water bath until it reached 25°C. Once the sample cooled, a 9 cm P8 grade plain filter paper
The circle (Fisherbrand, Waltham, MA) was folded into a cone shape and placed into a plastic funnel in a 20 ml beaker. The liquid containing the sample was poured into the filter and at least 5 ml of filtered liquid was collected. A Quantab high-level sodium chloride strip (Hach, Loveland, CO) was placed into the glass beaker containing the filtered liquid and left to sit until a black line appeared on the top of the strip. Once the strip turned black it was read using the standard on the back of the container set by Hach (Hach, Loveland, CO).

\[
\frac{NaCl}{0.10} = \%NaCl
\]

**Instrumental Color**

The external color of tumbled or marinated jerky was measured using a Hunter Lab MiniScan™ EZ spectrophotometer (Model 4500, Hunterlab, Reston, VA) using setting D65 on day 0 and after 3 and 6 months of storage. One piece within each package was measured in 3 different locations on the surface of the jerky to collect \( L^* \), \( a^* \), and \( b^* \) values.

**Shear Force**

To determine the SF of tumbled or marinated beef jerky on day 0 and after 3 and 6 months of storage, a straight blade was used with an Instron Universal Testing Machine (Model 5569 Canton, MA). Two random pieces of beef jerky at 20°C were manually cut with a rectangular plastic guide that was 25.4 mm wide into 25.4 mm strips along the fiber orientation. Each strip was sheared 3 times perpendicular to the longitudinal axis muscle fiber orientation with the straight blade at a rate of 250 mm/min in a 100 kg load cell.

**Sensory Evaluation**

Before trained sensory panels were conducted, panelists were trained using commercial and study samples that consisted of whole muscle and ground and formed beef jerky that was held at 20°C. Products were whole muscle or ground and formed with a similar flavor profile to
allow panelists to evaluate differences between samples and develop an understanding of anchor points for tenderness, texture, and flavor. The level of oxidation as an off-flavor was evaluated during the training sessions but since no panelist was able to detect any level of oxidation it was not utilized in the trained panel sessions. Each panelist sampled six jerky products per training session and had distilled water, gala apple slices, and unsalted crackers available to cleanse their pallet between samples.

Tumbled or marinated beef jerky samples from day 0 and 3 and 6 months of storage were presented to eight trained sensory panelists during 12 different panel sessions. The samples were removed from a -40°C freezer after being stored in the freezer for up to 6 months and thawed at 2°C overnight before evaluation. Panelists sampled six treatments per panel session. For sensory evaluation, jerky was manually cut using a guide box into 19 mm strips along the fiber orientation and then cut again into 19 × 19 mm squares. Each panelist received two 19 × 19 mm pieces of each sample, including a warm-up sample. A random warm-up sample that was from any treatment and storage period was served and discussed at all 12 sessions before evaluating the treatments.

Panelists were provided with distilled water, gala apple slices, and unsalted crackers to cleanse the pallet between samples. Treatments were evaluated for tenderness, texture, flavor, and off-flavor using a 100-point line scale on electronic tablets (Lenovo TB-8505F, Morrisville, NC) which used Qualtrics (Version 2417833; Qualtrics Software, Provo, UT). A score of 0 meant that the sample was extremely tough/chewy for tenderness, extremely soft for texture, extremely bland for flavor, and none for off-flavor. A score of 100 represented extremely tender/non-chewy for tenderness, extremely brittle/hard for texture, and extremely intense for flavor and off-flavor.
Structural Analysis

A sample slice of a raw top round or beef jerky was cut using a 3 mm diameter Biopunch (Ted Pella, Redding, CA). Each sample had 3 holes that were cored from 3 different locations within the slice along the long axis of the muscle fibers. Samples were immersed in a 0.5 ml primary fixative containing 4% paraformaldehyde (Electron Microscopy Services (EMS), Hatfield, PA) and 2% glutaraldehyde (EMS, Hatfield, PA) in a 0.1 M sodium cacodylate buffer (pH 7.2 – 7.4) (EMS, Hatfield, PA) in a sterile 2 ml micro-centrifuge tube. Samples were fixed for 16 h at 20°C with constant rotation on a shaker platform. After fixation, samples were washed 3 times for 5 min in a 0.5 ml 0.1 M sodium cacodylate buffer (pH 7.2 – 7.4) at 20°C with constant rotation.

Samples were enblocked stained by immersion in 0.5 ml 2% uranyl acetate (EMS, Hatfield, PA) aqueous stain for 1 h at 20°C with constant rotation. Samples were light protected during staining by wrapping tubes with tin foil. After staining, samples were washed 3 times with distilled water for 5 min each at 20°C with constant rotation.

Samples were dehydrated using an ascending acetone (EMS, Hatfield, PA) series (50, 60, 70, 80, 90, 95, 100 %). Samples were immersed and held for 5 min at 20°C for each step of acetone. Once the samples were dehydrated, they were sent to The KSU Microscopy Facility at Kansas State University for the remaining steps of the procedure. Fixed dehydrated 3 mm cores were bisected before infiltration. Samples were infiltrated with resin which is a viscous substance that is solidified to protect the integrity of a sample that was immersed in a 1:1 acetone: resin ratio for 30 min at 20°C followed by 1:2 acetones: resin for 1 h and 100% resin for 10 min. The fresh resin was added and infiltrated for an additional 16 h at 20°C with constant rotation.
Infiltrated samples were embedded into flat molds and oriented for longitudinal and cross-sectioning. The resin was cured in a drying oven at 60°C for 24 – 48 h. Once cured, blocks were trimmed and semi-thin sections (250-300 µm thick, purple to green sections) for LM and thin sections (60-90 nm, thick, gold to silver sections) for TEM were cut. Sections were cut on a Leica EM UC7 M80 ultra-microtome (Leica, Deerfield, IL). Semi-thin sections were absorbed onto glass slides, stained with toluidine blue, and imaged by LM. Thin sections were absorbed onto 200 mesh copper grids (EMS, Hatfield, PA) and imaged by TEM.

TEM images were collected using CM-100 TEM (Philips/FEI Company) at 100 kV equipped with a Hamamatsu C8484 digital camera (AMT, Chazy, NY) using AMT V602.591n software. LM images were collected using a Lasar Scanning Microscopy (Carl Zeiss, White Plains, NY, USA) equipped with a Zeiss Axioplan 2 upright microscope with a Plan Neofluor objective of 40x/0.75, differential contrast interference (DIC) optics, on a LSM 5 Pascal Version 3.2 SP2 software program analyzed using Image J.JS online software to analyze SL and MD with 30 replications for raw, raw tumbled, raw marinated, day 0, and 3 and 6 months of storage.

Statistical Analysis

Results were analyzed using SAS (Version 9.4; SAS Inst., Inc., Cary, NC) PROC GLIMMIX. This study utilized a 3 × 2 factorial design where the fixed effects were processing method and storage time with individual rounds used as a random blocking factor. The usage of α ≤ 0.05 as the level of significance was utilized for this study. The Kenward-Roger's adjustment was used for all analyses. Sarcomere length and MD were analyzed separately based on the main effects of the treatment method.
Results and Discussion

Proximate Analysis

The moisture, fat, and protein content of raw top rounds used to manufacture beef jerky were similar ($P > 0.05$) (Table 1). This showed that all the top rounds started at a similar baseline for proximate analysis.

There was no difference ($P > 0.05$) among treatment methods for protein content with similar values ranging from 54.67 – 55.79% (Table 2). Skaar et al. (2011) showed a difference between the marinated or tumbled products where tumbled jerky had a higher protein content which contradicts this study where there was no difference. These authors found that marination increased protein content by about 30% (Skaar et al., 2011).

There was a difference ($P < 0.05$) for the main effect of moisture content based on the processing method (Table 2). Tumbled jerky had a higher ($P < 0.05$) moisture content at 8.26% than the marinated product at 5.46% with a 51.52% increase in the tumbled product. This means that there could be similarities between the tumbled product having a higher pickup percentage to retain more moisture. On the other hand, Skaar et al. (2011) showed that there was no difference in the moisture content of jerky based on the treatment methods of tumbling or marinating. This may be because Skaar et al. (2011) found no difference in the percent pickup which may be related to the moisture percentage.

The moisture content of jerky during storage was similar ($P > 0.05$) and ranged from 6.62 – 7.17 (Table 4). To sustain the product quality, it would be important that the moisture content remain stable during the storage period to minimize moisture migration out of the product (Miller et al., 1988).
**Moisture Protein Ratio**

The moisture protein ratio (MPR) was similar ($P>0.05$) for tumbled and marinated jerky with values ranging from 0.13 – 0.14 (Table 2). Since the USDA FSIS (2014) states that a beef jerky product must have an MPR of 0.75 or less to be labeled as jerky, these values of 0.13 – 0.14 meet the criteria to be considered beef jerky but are very low compared to what is required for this product.

**Marinade Pick Up**

There was a 58.4% increase ($P<0.05$) in percent pickup using tumbling compared to the marination of beef strips (Table 3). According to Pearson and Gillett (1996), tumblers accelerate the extraction of meat proteins which allows for the improvement of the amount of absorption by the product. This is due to the agitation that occurred during tumbling, loosening the muscle structure, and increasing the amount of retained marinade (Pearson and Gillett, 1996). Marination had less structural impact on muscle structure due to a lack of agitation during the marination process resulting in less marinade pickup. Additionally, a study by Boles and Shand (2002) on *semimembranosus* found that injecting before tumbling improved the percent pickup but there was no difference among continuous versus intermittent tumbling.

**Cook Yield**

There was no difference ($P>0.05$) among processing methods for cook yield, with marinated jerky having a cook yield of 32.6% and tumbled jerky having a cook yield of 31.4% (Table 3). The results from a study on whole-muscle beef jerky by Sindelar et al. (2010) found higher cook yields of 44.1 - 45.5% for tumbled products. Sindelar et al. (2010) added sodium nitrite to some of the treatments and used a different thermal processing and drying schedule which may affect cook yield.
**pH**

The pH of tumbled or marinated jerky was similar \( (P > 0.05) \) and ranged from 5.45 - 5.56 (Table 2) which was similar to the recommended range of pH of 4.7 - 6.7 (Yang et al., 2002). A study by Lonnecker et al. (2010) had a similar average pH value of 5.86 based on all the small meat processors various processing methods.

**Water Activity**

The \( a_w \) of beef jerky should be \( \leq 0.85 \) after thermal processing to be considered shelf stable and safe to consume according to the FSIS Guidelines (USDA FSIS, 2014). All \( a_w \) values in this study were below the threshold of 0.85 with a range of 0.74 – 0.76. There was no difference in \( a_w (P > 0.05) \) due to the processing method (Table 2). Skaar et al. (2011) found lower values for \( a_w \), ranging from 0.54 - 0.62 with marinated jerky having a lower \( (P < 0.05) \) \( a_w \) than tumbled jerky. On the other hand, Lonnecker et al. (2010) found a 0.74 \( a_w \) value for beef jerky which was very comparable to the results in this study.

The \( a_w \) of jerky was similar \( (P > 0.05; \) Table 4 \) during storage and ranged from 0.76 – 0.74. Vacuum-packaging played a role in maintaining the quality of the product so the \( a_w \) remained consistent throughout the storage period and decreased the likelihood of contamination. Yang et al. (2009) looked at \( a_w \) of pork jerky and beef jerky with the beef jerky produced using a semimembranosus muscle. This study found no difference \( (P > 0.05) \) in \( a_w \) over a 30 d storage time.

**Sodium Chloride Content**

Although the tumbling process yielded a higher percent pickup than the marinating of raw beef strips, the SCC remained similar \( (P > 0.05) \) between the process treatments, at 2% which is a common SCC (Table 2). In contrast, Skaar et al. (2011) found that beef jerky strips
processed using marination had a higher SCC than tumbling. Skaar et al. (2011) hypothesized that the long soaking period of 24 h allowed for more marinade to penetrate and increase the SCC. A study on beef, emu, and turkey jerky by Carr et al. (1997) found no difference in SCC among the different species of jerky produced using tumbling. The SCC was similar ($P > 0.05$) based on the treatment method of thermally processed jerky with similar values that ranged from 3.3 – 3.4% (Table 3).

There was a difference ($P < 0.05$) in SCC due to vacuum-packaged storage for up to 6 months (Table 4). The SCC was the highest at 3.7% and similar ($P > 0.05$) at day 0 and 3 months, and then decreased ($P < 0.05$) at 6 months to 2.9%, resulting in a 20.70% decrease during the storage period. Sensory panelists also found the flavor of the jerky decreased during the storage period (Table 9).

**Instrumental Color**

The instrumental color values of $L^*$ and $a^*$ for tumbling or marination were different ($P < 0.05$) (Table 5). Tumbled jerky was lighter ($P < 0.05$) in color and less red ($P < 0.05$) than marinated jerky. Over the 6-month storage period, $L^*$ and $a^*$ values changed ($P < 0.05$; Table 6). From 3 to 6 months, $L^*$ became darker ($P < 0.05$) in color while $a^*$ became more red ($P < 0.05$). Sindelar et al. (2010) analyzed $L^*$ and $a^*$ values with whole muscle jerky at various storage periods including day 0. Within that study, it showed that there was no difference among treatments for the samples that did not have any nitrite added with jerky being darker in color when marinade was used.

Over the 6-month storage period, there was an interaction ($P < 0.05$) for process treatment and storage time for $b^*$ (Table 7). There was no change ($P > 0.05$) in $b^*$ for marinated jerky from day 0 to 6 months of storage. However, $b^*$ became less yellow ($P < 0.05$) for tumbled jerky
between day 0 to 3 months of storage, and then remained stable \((P>0.05)\) for the remainder of the storage period. Sindelar et al. (2010) found no change \((P>0.05)\) in \(b^*\) with beef jerky stored up to 56 d, except between d 0-7 for tumbled jerky.

**Shear Force**

For the tumbled and marinated jerky, the SF values differed \((P<0.05)\) with tumbled jerky being lower, requiring less force than marinated jerky. There was an 18.16% increase in tenderness for tumbled jerky compared to marinated jerky (Table 5). Panelists also found that tumbled jerky was more tender \((P<0.05)\), less brittle \((P<0.05)\), and had more flavor \((P<0.05)\).

Zochowska-Kujawska et al. (2017) looked at various types of marinades for home-style beef jerky and found there was no difference \((P>0.05)\) in SF among the different types of marinades; however, they did find differences \((P<0.05)\) in SF among the species of deer and boar jerky.

The SF of tumbled and marinated jerky stored for 0, 3, and 6 months was similar \((P<0.05; \text{Table 6})\). In contrast, the panelists found that the jerky texture became harder after 6 months of storage. Yang et al. (2009) found when comparing beef jerky (semimembranosus) and pork jerky (semimembranosus) that was marinated for 24 h, packaged in non-vacuumed sealed impermeable bags, and stored for up to 30 d, there was a decrease in SF during storage.

**Sensory Analysis**

Results for tenderness, texture, and flavor of tumbled and marinated beef jerky are shown in Table 8. Panelists found that tumbled jerky was more tender \((P<0.05)\) than marinated jerky. Marinated jerky was more brittle \((P<0.05)\) in texture than tumbled jerky, and tumbled jerky had a more intense \((P<0.05)\) flavor. Tumbled jerky might be more appealing to consumers in terms of tenderness being 40.14% more tender than marinated jerky, less brittle by 22.27%, and more flavorful by 7.43% compared to marinated jerky.
Carr et al. (1997) evaluated tenderness, flavor intensity, and chewiness in tumbled emu, beef, and turkey jerky. They found that beef jerky was the most flavorful, but towards the middle to lower end in terms of tenderness and chewiness. In the study by Skaar et al. (2011) trained panelists found chewiness and moisture to be similar between tumbled or marinated beef jerky, and marinated jerky was saltier and had a more intense flavor than tumbled jerky.

A difference ($P<0.05$) was observed in beef jerky for tenderness, texture, and flavor due to storage for up to 6 months (Table 9). Regardless of using a tumbling or marination process to manufacture beef jerky, jerky was less tender ($P<0.05$) after 6 months of storage compared to jerky at day 0, while tenderness at day 0 was similar ($P>0.05$) to jerky at 3 months of storage, and jerky at 3 months of storage was similar to ($P>0.05$) jerky at 6 months of storage. The jerky texture was more brittle ($P<0.05$) after 6 months of storage compared to day 0 and 3 months of storage. The jerky flavor was most intense ($P<0.05$) on day 0, and then declined by 11.6% by 3 months of storage. The flavor intensity of beef jerky was similar ($P>0.05$) between 3 and 6 months of storage. Kim et al. (2010) found that beef jerky irradiated by an electron beam had a decrease in appearance during storage, along with an initial decrease in texture.

**Raw, Tumbled, or Marinated Top Round**

The raw top rounds (*semimembranosus*) that were held in a cooler at 2°C for up to two weeks were imaged by LM (Figure 2 A, D) and TEM (Figure 3 A, D). There was some loss of skeletal muscle morphological structure during the two-week storage. Muscle fibers and satellite cell nuclei were not observed by LM or TEM. There was no difference ($P>0.05$) among process treatments for SL or MD (Table 10).

Remnants of the sarcoplasmic reticulum, T-tubules, and mitochondria were observed. The myofiber diameters (green double arrowed lines) were determined and endomysium (red
single arrowed line) spacing between myofibers was noted. Figure 2A indicated some capillaries (red single arrowed line) were still visible. Glycogen granules (white single arrowed line), myofiber diameter, endomysium, and perimysium (red double arrowed line) were represented in Figure 2D. Figure 3D represented the integrity of thick (white closed point) and thin filaments (white open point) in raw beef samples. Thick filaments were prominent while thin filaments were less prominent but still present.

A representative sarcomere length (white double arrowed line) and intact Z-disc (yellow single arrowed line) were indicated in Figure 3A. The I-band (black double arrowed line) indicated the distance between each A-band in the sarcomere. I-bands and M-lines showed signs of breakdown. Feng et al. (2020) showed that after 7 d of storage myofibril integrity diminished and by 14 d of storage, M-lines, and I-bands showed further degradation. There were no satellite cell nuclei observed by Feng et al. (2020) in postmortem meat which were also found in this study. The results shown in Figure 3A where cellular debris (red circle) was present in raw beef were also reported by Feng et al. (2020).

Representative tumbled raw top round samples were observed by LM (Figure 2 B, E) and TEM (Figure 3 B, E). The main difference between the raw top round (Figure 2 A, D) and the raw tumbled top is represented in Figure 2 (B, E). In raw tumbled samples, myofiber tears running perpendicular to the long axis of fibers were observed (blue double arrowed line). These tears had relatively large gaps filled with dispersed cellular debris compared to the jerky samples that showed reduced gaps and concentrated cellular debris (Figure 4 A, B, C). In addition, myofibril tears were observed by TEM running perpendicular to the long axis of the fibrils mainly in the I-bands. These structural differences observed in raw tumbled samples (tears with large gaps) might explain the finding that SF values were less in raw tumbled samples as
compared to raw top round and raw marinated samples. In addition, structural observation of large gaps between tears in raw tumbled samples taken together with the finding of significantly higher moisture content for raw tumbled samples supported that these gaps might be filled with marinade fluid.

Myofiber tears (blue double arrowed line) shown in Figure 2B occurred after 30 min of tumbling the raw top round at 20 rpm. The myofiber diameters, glycogen granules, and endomysium were still visible and similar to what was observed for raw top rounds. This was in direct contrast to that observed by N’Gatta et al. (2021) that reported 30 min of tumbling resulted in reduction in the distance between myofibers. In addition, N’Gatta et al. (2021) reported some cracks perpendicular to myofibers. They did not report or show data indicating major and numerous myofiber tears running perpendicular to the long axis of fibers. In direct opposition to that reported by N’Gatta et al. (2021), a greater variation in endomysium spacing between myofibers was observed (Figure 2 E). Since tears in myofibers were not observed in raw or marinated samples, these tears were most likely due to mechanical damage and shearing of myofibers and myofibrils.

The longitudinal and cross-sectional images of thick and thin filaments were not as easily defined by TEM observation (Figure 3 B, E), especially the thin filaments in Figure 3E compared to the raw top round sample in 3D. I-bands and M-lines showed signs of breakdown as with the raw samples. There was more inter-myofibrillar cellular debris observed as compared to raw samples. This was similar to what was found by N’Gatta et al. (2021) who showed disorganization of myofiber ultrastructure and fragmentation that occurred to the sarcomeres within the myofibrils after tumbling occurred. N’Gatta et al. (2021) reported that the extent of
fragmentation and damage to the structural integrity of the myofibers and myofibrils increased the more meat pieces were tumbled.

The raw top round after it had soaked in the marinade for 24 h is represented in Figure 2 (C, F). Differences in structural changes were observed for the tumbled product (Figure 2 B, E) versus the marinated product (Figure 2 C, F). One of the main differences that occurred was that raw marinated samples did not show myofiber tears running perpendicular to the long axis of fibers, but did show splits in myofibers and myofibrils. Degradation of myofiber integrity was observed in raw marinated samples as compared to raw top round samples. In most areas, the spacing between myofibers was similar to the raw top round, but in some areas, the myofiber spacing was greater than that observed in the raw tumbled sample (Figure 2 C, F). As shown in Figure 2C the increased spacing was filled with cellular debris. Since the pH of the marinate was 4.70 these structural changes were probably due to acid hydrolysis of proteins. Sharedeh et al. (2011) reported similarly that spacing between myofibers increased as the pH of marinade decreased from 6.5 to 4.3. In addition, the salt concentrations of 2%, which was similar to what was found in Table 3, were associated with greater spacing between myofibers. They concluded that this phenomenon wasn’t the result of myofibril swelling due to marinade uptake.

Xu et al. (2023) analyzed lean pork ham and Unal et al. (2023) analyzed beef Holstein (Longissimus lumborum) with scanning electron microscopy (SEM). Both studies showed similar results to Figure 2D where the controls showed structural integrity and limited spacing before the marinade was introduced to the product. Once the marinade was introduced, the structural integrity started to breakdown and more spacing was present within the samples which was comparable to Figure 2C where spacing and cellular debris started to develop.
In Figure 2C, there was a breakdown that occurred within the myofibers (pink double arrowed line) where the split within the myofibers was not very well defined compared to Figure 2B where tears occurred. As indicated in Figure 3C these myofiber splits were due to degradation of the thin filaments found in I-bands of myofibrils. Thick filaments were observed by TEM, but thin filaments showed degradation to the point that thin filaments were difficult to observe if at all in cross-sections of myofibril A-bands (Figure 3F) as compared to raw top round and raw tumbled samples. Raw marinated samples showed greater degradation of M-lines with similar Z-disc to raw top round and raw tumbled samples, supporting the conclusion that the pH of the marinate (4.70) resulted in these further structural changes due to acid hydrolysis of proteins.

Latif (2010) analyzed marination effects on chicken breast meat where the 24 h marinated samples showed similar results to this study in Figure 3C. The results indicated that after the marination process, the Z-line, I-bands, and parts of the structural integrity of the sarcomere were still visible but degradation had started to occur which was similar to what was shown in Figure 3C.
**Figure 2.** Structural evaluation of raw top round, compared to raw tumbled or marinated top round. Representative LM (A–F). Images show longitudinal (A–C) and cross-section images (D–F). Refer to page xi for the Legend for Microscopy Images that has a description of all markups in the figures.

**Figure 3.** Structural evaluation of raw top round, compared to raw tumbled or marinated top round. Representative LM (A–F). Images show longitudinal (A–C) and cross-section images (D–F). Refer to page xi for the Legend for Microscopy Images that has a description of all markups in the figures.
Day 0, 3, and 6 Months – Tumbled or Marinated Beef Jerky

As represented in Figure 4 (A, B, C) myofiber tears were present after cooking in tumbled samples. The major differences were observed by LM between raw and cooked products were gaps that had become compressed and cellular debris that became concentrated. This observation supports that raw tumbled sample gaps might be filled with marinade fluid. Structural observations of reduced gap distances with a concentration of cellular debris support that cooking resulted in marinade loss and reduction in the gaps. There were no major structural changes ($P>0.05$) that were observed from LM and TEM between day 0, 3, and 6 months of storage for vacuum-packaged jerky stored at 20°C (Table 11).

Marinated jerky samples at day 0 and stored for 3 and 6 months under vacuum at 20°C were represented in Figure 4 (D, E, F). The splits in myofibers that were observed by LM in raw marinated samples were prominent and the concentration of cellular debris was greater in cooked/jerky samples. The concentration of cellular debris was observed between splits and between myofibers. There were no major structural changes that were observed by LM between day 0, 3, and 6 months of vacuum-packaged storage at 20°C.

Kim et al. (2022) analyzed beef jerky that was injected and tumbled with various brine levels and dried for different periods. The results were similar to Figure 4A with the 30% water/70% beef combination that was cooked for 3 h which showed a decrease in the structural integrity and myofiber diameters.

Ultrastructural analysis by TEM of tumbled jerky samples at day 0 and stored for 3 and 6 months under vacuum at 20°C was represented in Figure 5 (A, B, C). Major ultrastructural changes were observed between tumbled jerky versus tumbled raw samples. At day 0 (Figure 5 A), massive degradation of I-bands, M-lines, Z-disc, and A-bands was observed. Thin filaments
were no longer distinguishable. There was also an increased amount of cellular debris between myofibrils. Thick filaments were difficult to discern. Remnants of the sarcoplasmic reticulum, T-tubules, and mitochondria were no longer observed. At 3 months of storage (Figure 5 B), fragmented Z-disc, an electron-dense remnant of A-bands, and cellular debris were observed (Figure 5 B). At 6 months of storage (Figure 5 C), an electron-dense remnant of A-bands, with decreased cellular debris, and increased spacing between remnant myofibrils was observed.

Ultrastructural analysis by TEM of marinated jerky samples at day 0 and stored for 3 and 6 months under vacuum at 20°C were represented in Figure 5 (D, E, F). Major ultrastructural changes were observed between marinated jerky verses marinated raw samples. At day zero (Figure 5 A), massive degradation of I-bands, M-lines, Z-disc, and A-bands was observed. This degradation was greater than what was observed with tumbled jerky. Thin filaments were no longer distinguishable. There was an increased amount of cellular debris between myofibrils. Thick filaments were difficult to define. Remnants of the sarcoplasmic reticulum, T-tubules, and mitochondria were no longer observed. At 3 months of storage (Figure 5 B), fragmented Z-disc, an electron-dense remnant of A-bands, and cellular debris were observed (Figure 5 E). Thick filaments were no longer visible. At 6 months of storage (Figure 5 F), an electron-dense remnant of A-bands, with decreased cellular debris, and increased spacing between remnant myofibrils was observed. The increased space was greater than what was observed with tumble jerky stored for 6 months.

Comparing raw tumbled and marinated samples with cooked tumbled and marinated samples indicates that cooking increased protein degradation most likely due to heat-induced protein denaturation and increased protein aggregation. Increased storage time resulted in increased loss of meat structure. Which would need to be confirmed through further analysis.
The results for day 0 demonstrate that for the sensory and SF analysis, the tumbled samples were more tender due to the impact of the structural integrity of the product. Sensory analysis on the tumbled samples resulted in a higher score for tenderness, texture, and flavor attributes.

Based on the results, the impact that occurred on the tumbled and marinated product during the storage period showed that overall the beef jerky samples decreased in tenderness, texture, and flavor over time. Żochowska-Kujawska et al. (2017) found similar results in the study on shear force and sensory properties of home-style jerky more specifically in beef where the samples decreased in the structural integrity over time. However, further work could be done on extended storage periods of beef jerky as limited research was available.

**Figure 4.** Structural evaluation of day 0 beef jerky compared to beef jerky at 3 or 6 months of storage that was tumbled or marinated. Representative LM (A – F). Images show longitudinal images (A – F). Refer to page xi for the Legend for Microscopy Images that has a description of all markups in the figures.
Figure 5. Structural evaluation of day 0 beef jerky compared to beef jerky at 3 or 6 months of storage that was tumbled or marinated. Representative TEM (A–F). Images show cross-section images (A–F). Refer to page xi for the Legend for Microscopy Images that has a description of all markups in the figures.

Conclusion

Tumbling produced a jerky product that was more tender, less brittle, and more flavorful during 6 months of storage compared to marination as a processing method. Although tumbling yielded a higher percent pickup and was darker in color than jerky produced using marination, the processing method did not influence the SCC, $a_w$, or MPR of jerky produced. If given the opportunity to choose which method to utilize for their facility, it is recommended that processors utilize tumbling to yield the highest quality and sensory product. Additional research is needed to assess a combination of various processing methods or extended periods of storage to see the potential effects processing methods have on the quality and sensory attributes of jerky.
References


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Tables

Table 1. Least squares means (LSmeans) of moisture, fat, and protein on raw top round slices.

<table>
<thead>
<tr>
<th>Process</th>
<th>Moisture (%)</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marinade</td>
<td>73.27</td>
<td>2.14</td>
<td>22.95</td>
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<tr>
<td>Tumble</td>
<td>73.05</td>
<td>2.56</td>
<td>22.40</td>
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<tr>
<td>P-value</td>
<td>0.3919</td>
<td>0.2306</td>
<td>0.0906</td>
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<tr>
<td>SEM²</td>
<td>0.2284</td>
<td>0.2679</td>
<td>0.2368</td>
</tr>
</tbody>
</table>

¹MPR: Moisture Protein Ratio.
²Standard error of the least squares means.

Table 2. Least squares means (LSmeans) of protein, pH, aw, moisture, and sodium chloride on raw tumbled or raw marinated top rounds.

<table>
<thead>
<tr>
<th>Process</th>
<th>Protein (%)</th>
<th>pH</th>
<th>aw¹</th>
<th>Moisture (%)</th>
<th>Sodium Chloride (%)</th>
<th>MPR²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marinade</td>
<td>55.79</td>
<td>5.56</td>
<td>0.7430</td>
<td>5.4575b</td>
<td>3.410</td>
<td>0.13</td>
</tr>
<tr>
<td>Tumble</td>
<td>54.67</td>
<td>5.45</td>
<td>0.7571</td>
<td>8.2694a</td>
<td>3.336</td>
<td>0.14</td>
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<tr>
<td>P-value</td>
<td>0.2792</td>
<td>0.0529</td>
<td>0.2385</td>
<td>&lt;.0001</td>
<td>0.527</td>
<td>0.6425</td>
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<tr>
<td>SEM³</td>
<td>0.7796</td>
<td>0.0522</td>
<td>0.0104</td>
<td>0.5843</td>
<td>0.009</td>
<td>0.0202</td>
</tr>
</tbody>
</table>

¹aw: Water Activity.
²MPR: Moisture Protein Ratio.
³Standard error of the least squares means.

Table 3. Least squares means (LSmeans) of raw product pickup, cooked product yield, and sodium chloride percentages of tumbled or marinated top rounds and beef jerky.

<table>
<thead>
<tr>
<th>Process</th>
<th>Pickup (%)</th>
<th>Cook Yield (%)</th>
<th>Sodium Chloride (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marinade</td>
<td>17.3b</td>
<td>32.6</td>
<td>2.026</td>
</tr>
<tr>
<td>Tumble</td>
<td>27.4a</td>
<td>31.4</td>
<td>2.020</td>
</tr>
<tr>
<td>P-value</td>
<td>0.0004</td>
<td>0.1181</td>
<td>0.969</td>
</tr>
<tr>
<td>SEM¹</td>
<td>0.0170</td>
<td>0.0052</td>
<td>0.012</td>
</tr>
</tbody>
</table>

¹Means within a column without a common superscript differ (P < 0.05).
²Means within a column with different superscripts differ (P < 0.05).
³Standard error of the least squares means.

Table 4. Least squares means (LSmeans) of protein, pH, aw, moisture, and sodium chloride on tumbled or marinated vacuum-packaged beef jerky that was stored for up to 6 months at 20°C.

<table>
<thead>
<tr>
<th>Storage Time</th>
<th>aw¹</th>
<th>Moisture (%)</th>
<th>Sodium Chloride (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 Day</td>
<td>0.7394</td>
<td>7.17</td>
<td>3.695a</td>
</tr>
<tr>
<td>3 Months</td>
<td>0.7492</td>
<td>6.80</td>
<td>3.519a</td>
</tr>
<tr>
<td>6 Months</td>
<td>0.7614</td>
<td>6.62</td>
<td>2.906b</td>
</tr>
<tr>
<td>P-value</td>
<td>0.3196</td>
<td>0.7697</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>SEM²</td>
<td>0.0120</td>
<td>0.6621</td>
<td>0.0143</td>
</tr>
</tbody>
</table>

¹Means within a column without a common superscript differ (P < 0.05).
²aw: Water Activity.
³Standard error of the least squares means.
Table 5. Least squares means (LSmeans) $b^{*1}$ on tumbled or marinated vacuum-packaged beef jerky that was stored for up to 6 months at 20°C.

<table>
<thead>
<tr>
<th>Process × Storage</th>
<th>Day 0</th>
<th>3 Months</th>
<th>6 Months</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumble</td>
<td>11.22$^a$</td>
<td>8.07$^{bc}$</td>
<td>9.16$^b$</td>
</tr>
<tr>
<td>Marinade</td>
<td>6.69$^c$</td>
<td>6.95$^c$</td>
<td>7.71$^{bc}$</td>
</tr>
<tr>
<td>$P$ - value</td>
<td>0.0396</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SEM$^2$</td>
<td>0.8309</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

$^{abc}$Means within a row or column without a common superscript differ ($P < 0.05$).

$^1b^{*} = -60 =$ blue, $60 =$ yellow.

$^2$Standard error of the least squares means.

Table 6. Least squares means (LSmeans) of instrumental color and shear force on vacuum-packaged beef jerky produced using tumbling or marination.

<table>
<thead>
<tr>
<th>Process Treatment</th>
<th>$L^{*1}$</th>
<th>$a^{*2}$</th>
<th>Shear Force (kgf)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marinade</td>
<td>24.30$^b$</td>
<td>3.32$^b$</td>
<td>38.235$^a$</td>
</tr>
<tr>
<td>Tumble</td>
<td>25.93$^a$</td>
<td>4.11$^a$</td>
<td>32.360$^b$</td>
</tr>
<tr>
<td>$P$ - value</td>
<td>0.0032</td>
<td>0.0017</td>
<td>0.0039</td>
</tr>
<tr>
<td>SEM$^3$</td>
<td>0.6738</td>
<td>0.2325</td>
<td>2.1653</td>
</tr>
</tbody>
</table>

$^{ab}$Means within a column without a common superscript differ ($P < 0.05$).

$^1L^{*} = 0 =$ black, $100 =$ white.

$^2a^{*} = -60 =$ green, $60 =$ red.

$^3$Standard error of the least squares means.

Table 7. Least squares means (LSmeans) of instrumental color and shear force on tumbled or marinated vacuum-packaged beef jerky that was stored for up to 6 months at 20°C.

<table>
<thead>
<tr>
<th>Storage Time</th>
<th>$L^{*1}$</th>
<th>$a^{*2}$</th>
<th>Shear Force (kgf)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 Day</td>
<td>26.47$^a$</td>
<td>3.30$^b$</td>
<td>36.244</td>
</tr>
<tr>
<td>3 Months</td>
<td>25.17$^a$</td>
<td>3.61$^b$</td>
<td>34.462</td>
</tr>
<tr>
<td>6 Months</td>
<td>23.70$^b$</td>
<td>4.24$^a$</td>
<td>35.185</td>
</tr>
<tr>
<td>$P$ - value</td>
<td>0.0004</td>
<td>0.0073</td>
<td>0.3744</td>
</tr>
<tr>
<td>SEM$^3$</td>
<td>0.7243</td>
<td>0.2614</td>
<td>2.3747</td>
</tr>
</tbody>
</table>

$^{ab}$Means within a column without a common superscript differ ($P < 0.05$).

$^1L^{*} = 0 =$ black, $100 =$ white.

$^2a^{*} = -60 =$ green, $60 =$ red.

$^3$Standard error of the least squares means.
Table 8. Least squares means (LSmeans) for trained sensory evaluation of palatability characteristics of tumbled or marinated vacuum-packaged beef jerky.

<table>
<thead>
<tr>
<th>Process Treatment</th>
<th>Tenderness&lt;sup&gt;1&lt;/sup&gt;</th>
<th>Texture&lt;sup&gt;2&lt;/sup&gt;</th>
<th>Flavor&lt;sup&gt;3&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marinade</td>
<td>25.66&lt;sup&gt;b&lt;/sup&gt;</td>
<td>74.78&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.60&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Tumble</td>
<td>35.96&lt;sup&gt;a&lt;/sup&gt;</td>
<td>61.16&lt;sup&gt;b&lt;/sup&gt;</td>
<td>37.17&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>P - value</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>0.0455</td>
</tr>
<tr>
<td>SEM&lt;sup&gt;4&lt;/sup&gt;</td>
<td>1.8668</td>
<td>2.2130</td>
<td>1.0461</td>
</tr>
</tbody>
</table>

<sup>ab</sup>Means within a column with different superscripts differ (P < 0.05).
<sup>1</sup>Tenderness: 0 = extremely tough/chewy, 50 = neither tough/chewy nor tender/non chewy, 100 = extremely tender/non chewy.
<sup>2</sup>Texture: 0 = extremely soft, 50 = neither soft nor brittle/hard, 100 = extremely brittle/hard.
<sup>3</sup>Flavor: 0 = extremely bland, 100 = extremely intense.
<sup>4</sup>Standard error of the least squares means.

Table 9. Least squares means (LSmeans) for trained sensory evaluation of palatability characteristics<sup>1</sup> of tumbled or marinated vacuum-packaged beef jerky stored up to 6 months at 20°C.

<table>
<thead>
<tr>
<th>Storage Time</th>
<th>Tenderness&lt;sup&gt;1&lt;/sup&gt;</th>
<th>Texture&lt;sup&gt;2&lt;/sup&gt;</th>
<th>Flavor&lt;sup&gt;3&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 Day</td>
<td>33.00&lt;sup&gt;a&lt;/sup&gt;</td>
<td>63.45&lt;sup&gt;b&lt;/sup&gt;</td>
<td>39.05&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3 Months</td>
<td>32.10&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>67.30&lt;sup&gt;b&lt;/sup&gt;</td>
<td>34.53&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>6 Months</td>
<td>27.33&lt;sup&gt;b&lt;/sup&gt;</td>
<td>73.16&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.07&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>P - value</td>
<td>0.0485</td>
<td>0.0057</td>
<td>0.0032</td>
</tr>
<tr>
<td>SEM&lt;sup&gt;4&lt;/sup&gt;</td>
<td>2.1067</td>
<td>2.5063</td>
<td>1.2207</td>
</tr>
</tbody>
</table>

<sup>ab</sup>Means within a column with different superscripts differ (P < 0.05).
<sup>1</sup>Tenderness: 0 = extremely tough/chewy, 50 = neither tough/chewy nor tender/non chewy, 100 = extremely tender/non chewy.
<sup>2</sup>Texture: 0 = extremely soft, 50 = neither soft nor brittle/hard, 100 = extremely brittle/hard.
<sup>3</sup>Flavor: 0 = extremely bland, 100 = extremely intense.
<sup>4</sup>Standard error of the least squares means.

Table 10. Least squares means (LSmeans) of microscopy for treatments of raw, raw marinated, or raw tumbled treatments, or tumbled or marinated vacuum-packaged beef jerky.

<table>
<thead>
<tr>
<th>Process Treatment</th>
<th>SL&lt;sup&gt;1&lt;/sup&gt;</th>
<th>MD&lt;sup&gt;2&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw</td>
<td>1.3315</td>
<td>45.9362</td>
</tr>
<tr>
<td>Marinade</td>
<td>1.3471</td>
<td>46.1465</td>
</tr>
<tr>
<td>Tumble</td>
<td>1.1561</td>
<td>46.7733</td>
</tr>
<tr>
<td>P - value</td>
<td>0.5452</td>
<td>0.9946</td>
</tr>
<tr>
<td>SEM&lt;sup&gt;4&lt;/sup&gt;</td>
<td>241.80</td>
<td>9.7682</td>
</tr>
</tbody>
</table>

<sup>1</sup>SL = Sarcomere Length (µm).
<sup>2</sup>MD = Myofiber Diameter (µm).
<sup>3</sup>n=1.
<sup>4</sup>Standard error of the least squares means.
Table 11. Least squares means (LSmeans) of microscopy for treatments of tumbled or marinated vacuum-packaged beef jerky stored up to 6 months at 20°C.

<table>
<thead>
<tr>
<th>Process Treatment</th>
<th>SL(^1)</th>
<th>MD(^2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 0</td>
<td>1.2167</td>
<td>42.632</td>
</tr>
<tr>
<td>3 Months</td>
<td>1.3005</td>
<td>44.978</td>
</tr>
<tr>
<td>6 Months</td>
<td>1.0930</td>
<td>39.885</td>
</tr>
<tr>
<td>(P) - value</td>
<td>0.8798</td>
<td>0.4747</td>
</tr>
<tr>
<td>SEM(^4)</td>
<td>217.17</td>
<td>11.776</td>
</tr>
</tbody>
</table>

\(^1\) SL = Sarcomere Length (µm).
\(^2\) MD = Myofiber Diameter (µm).
\(^3\) n=1.
\(^4\) Standard error of the least squares means.