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Influence of harvest method on proximate composition, texture profile and protein oxidation of beef from grain-fed Nellore cattle

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ABSTRACT. The influence of harvest method on proximate composition, protein oxidation, and texture profile of beef from Nellore cattle were investigated. *Longissimus lumborum* (LL) muscles were obtained from twelve grain-fed Nellore carcasses. The animals were slaughtered after stunning (STU; n=6) or without stunning (WST; n=6) and after 24 hours postmortem, the LL was sliced into 2.54-cm steaks, packed under aerobic conditions, and stored at 4°C for nine days. Proximate composition was analyzed on day 0, whereas protein oxidation (carbonyl content) and texture profile (hardness, chewiness, cohesiveness, and springiness) were evaluated on days 0, 3, 6, and 9. STU and WST steaks exhibited similar proximate composition, hardness, and chewiness ($p > 0.05$). WST steaks exhibited greater protein oxidation and lower cohesiveness than their STU counterparts on day 0 ($p < 0.05$). Concerning the storage period, WST steaks demonstrated a decrease in springiness and a more pronounced increase in cohesiveness than their STU counterparts ($p < 0.05$). These results indicated that the harvest method influenced LL protein oxidation and texture profile from *Bos indicus* cattle.

Keywords: stunning; ritual slaughter; oxidative stability; storage; texture profile.

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Introduction

Brazil is a great beef producer and occupies the top position in beef exportation since 2015 (United States Department of Agriculture [USDA], 2022). Brazilian beef is destined for many countries, including where slaughter without stunning is required (USDA, 2022). In Brazil, two slaughter methods are usually applied to harvest cattle (Brasil, 2017). The first and most common method is the harvest with stunning, in which the animal became senseless before the time of slaughter. The second method is defined as a ritual or religious slaughter and comprises halal and kosher practices (Farouk, 2013). In these last methods, cattle have their throats slit and bleed to death without prior stunning (Farouk et al., 2014). In this sense, Brazilian regulation allowed the slaughter of animals under religious precepts (without stunning) since their products are destined for international trade with countries that make this requirement for consumption by a religious community (Brasil, 2017).

Oxidative processes are the main non-microbiological cause of meat quality deterioration during refrigerated storage. Meat quality is determined by color, texture, and oxidative susceptibility (Suman, Hunt, Nair, & Rentfrow, 2014), which can be affected by slaughter without stunning associated with pre-slaughter practices (Farouk, 2013). Although the color of fresh beef is the primary quality attribute governing the consumers' purchase decisions (Mancini & Hunt, 2005; Suman et al., 2014), texture also strongly contributes to this. Both are influenced by intrinsic and extrinsic factors (Neethling, Sigge, Hoffman, Suman, & Hunt, 2017). Concerning intrinsic factors, Nellore (*Bos indicus*) cattle are characterized by their excitable temperament, which influences antemortem management practices, contributing to increase the meat oxidation (Francisco et al., 2015). Furthermore, protein oxidation is known to promote conformational and functional changes in proteins, which can influence the texture profile (Cichoski et al., 2019).

Among extrinsic factors, the harvest method plays an important role in color (Önenç & Kaya, 2004; D'Agata, Russo, & Preziuso, 2009; Salim et al., 2019) and in the susceptibility of meat to lipid oxidation (Linares, Berruga,

Bornez, &Vergara, 2007; Salim et al., 2019). Recently, Salim et al. (2019) evaluated the influence of harvest on beef quality traits in the *longissimus lumborum* muscle from *Bos indicus* and observed that steaks from non-stunned cattle exhibited lower redness and color stability, and greater lipid oxidation than their stunned counterparts. Önenç and Kaya (2004) documented greater redness (a^*) in longissimus muscles from stunned *Bos taurus* cattle than in muscles from non-stunned animals. Moreover, Linares et al. (2007) reported an increase in lipid oxidation in longissimus muscle from non-stunned lambs than in their stunned counterparts.

While the influence of stunning on color stability and lipid oxidation of beef has been documented, the influence of harvest on proximate composition, protein oxidation and texture profile of beef from Nellore (*Bos indicus*) cattle is yet to be investigated. Therefore, the present study examined the proximate composition, protein oxidation, and instrumental texture profile of *longissimus lumborum* muscle from stunned and non-stunned Nellore (*Bos indicus*) stored under refrigeration.

Material and methods

Experimental design and beef steak preparation

Twelve carcasses of Nellore (*Bos indicus;* 24-36 months-of-age) bulls were used in this experiment. Animals were grain-fed and raised under similar conditions in a farm of Nanuque, state of Minas Gerais and slaughtered after stunning (STU; n=6) or without stunning (WST; n=6). Samples of beef longissimus were purchased from a commercial packing plant. Therefore, Institutional Animal Care and Use Committee approval was not obtained. Cattle were harvested at a commercial facility in Nanuque, Minas Gerais, Brazil, certified by the Brazilian federal inspection (SIF) under the following conditions: six (n=6) animals were slaughtered after stunning with a percussive captive bolt pistol (STU), and the remaining six (n=6) cattle were slaughtered without stunning (WST).

After 24 hours, the average cold carcass weight of STU was 282.48 kg, whereas that of WST was 281.15 kg. LL muscles were taken from the right carcasses, individually vacuum packaged, and shipped under refrigeration to the meat laboratory at Universidade Federal Fluminense (Niteroi, Rio de Janeiro, Brazil). LL muscles were sliced into eight 2.54-cm thick steaks, and their pH was directly measured using a portable pH meter (Hanna Instruments, Woonsocket, RI) equipped with an insertion probe (González-Fuentes, Hamedy, von Borell, Luecker, & Riehn, 2014), obtaining the pH values of 5.47 ± 0.03 in STU and 5.61 ± 0.01 in WST. Steaks were individually packaged on polystyrene trays with soaker pads, over-wrapped with oxygen-permeable polyvinyl chloride (PVC) film (0.014 mm thickness; Orleplast Ind. e Com. de Plásticos Ltda, Orleans, Santa Catarina, Brazil), and stored at 4°C for 9 days. Proximate composition was analyzed on day 0, whereas protein oxidation and texture profile were evaluated at days 0, 3, 6, and 9. All the analyses were performed in triplicate.

Proximate composition

For the determination of meat chemical composition, LL samples were homogenized in a food processor to a homogeneous mass, packaged, and identified. The contents of moisture, protein, and ash were determined according to Association of Official Analytical Chemists (AOAC, 2012). The content of total lipid was determined according to the methodology described by Bligh and Dyer (1959).

Protein oxidation

Protein oxidation was estimated by the method proposed by Oliver, Ahn, Moerman, Goldstein, and Stadtman (1987) with modifications (Armenteros, Heinonen, Ollilainen, Toldrá, & Estévez, 2009; Mercier, Gatellier, Viau, Remignon, & Renerre, 1998). Samples of 3 g were homogenized with 30 mL of 0.15 M KCl (pH 7.4) using an Ultra Turrax T18 basic (IKA, Wilmington, NC, USA) for 90 seconds. Homogenates were divided into two equal volumes (0.1 mL), individually homogenized with 1 mL of 10% TCA on a bench vortex and centrifuged at 5,000 × *g* for 5 minutes (4°C) for protein precipitation. Supernatants were discarded, and each pellet was homogenized with either 1 mL of 2 N HCl or 1 mL of 10 mM DNPH in 2 N HCl in order to estimate protein and carbonyl contents, respectively. Homogenates were incubated at 25°C for 1 hour with intermittent brief vortexing every 15 minutes following final precipitation with 1 mL of 10% TCA and centrifugation at 11,000 × g for 10 minutes (4°C).

The precipitate was washed three times with 1 mL of 1:1 ethanol/ethyl acetate (v/v) solution and centrifuged at 15,000 × g for 10 minutes (4ºC). Pellets were dissolved in 1.0 mL of a buffer consisting of 20 mM sodium phosphate (pH 6.5) and 6 M guanidine hydrochloride. Homogenates were centrifuged at 11,000 × *g* for 10 minutes to remove insoluble particles. The protein content was estimated by absorbance at 280 nm against a bovine serum albumin

standard curve, whereas carbonyl content was estimated by absorption at 370 nm and an absorptivity coefficient for the protein hydrazones at 21.0 mM^{-1} cm^{-1}. Results were expressed as nmol of carbonyl per mg of protein.

Texture profile analysis

The instrumental texture profile analysis (TPA) was performed according to Huidobro, Miguel, Blázquez, and Onega (2005) using a texture analyzer (TA.XT Plus; Stable Micro System, United Kingdom) equipped with a cylindrical metal probe (72 mm diameter). Cubes of 1.0 × 1.0 × 1.0 cm were obtained from STU and WST samples and compressed to 75% of their original height in the pre-test (3 mm s^{-1}), test (1 mm s^{-1}), and post-test (3 mm s^{-1}), with 2 seconds interval between compressions (Choe, Choi, Rhee, & Kim, 2016). The test was performed three times, and the average of readings was considered for statistical analyses. Data were processed with the Texture Exponent Software (Stable Micro System, United Kingdom) to obtain the results expressed as hardness, springiness, cohesiveness, and chewiness (Botinestean, Keenan, Kerry, & Hamill, 2016).

Statistical analysis

In this study, n=12 beef carcasses were divided into six (n=6) replicates of STU and WST treatments. A one-way ANOVA was applied to analyze proximate composition data. A two-way ANOVA was used to test data from protein oxidation and instrumental texture profile analyses. Pearson's correlation coefficients were estimated between parameters evaluated during the storage period.

All statistical analyses were run 5% significance level in the XLSTAT software, version 2014.5.03 (Addinsoft Inc., Brooklyn, USA).

Results and discussion

Proximate composition

The harvest method did not ($p > 0.05$) influence the proximate composition of STU and WST steaks (Table 1). STU and WST steaks exhibited similar ($p > 0.05$) moisture, protein, ash, and lipid values on zero-day of storage. In agreement, Kim et al. (2013) evaluated the effect of stunning (captive bolt stunner x CO2 gas) on the longissimus muscle of Korean native cattle (*Bos taurus coreanae*) and observed no differences in moisture, lipid, ash, and protein contents between treatments. In partial agreement, Bostami, Mun, and Yang (2018) evaluated the influence of halal and non-halal slaughter on the longissimus muscle of Korean native cattle and observed similar moisture, lipid, and protein contents in both treatments, whereas ash content was greater in longissimus from the halal group.

Table 1. Proximate composition of *longissimus lumborum* (LL) steaks (n=6) from Nellore (*Bos indicus*) cattle harvest with stunning (STU) or without stunning (WST) during storage at 4°C for 9 days.

	Harvest method		
Parameter	STU	WST	*P* value
Moisture (%)	75.08 ± 0.22[a]	75.11 ± 0.21[a]	0.7943
Protein (%)	16.60 ± 0.58[a]	18.39 ± 0.77[a]	0.1864
Ash (%)	0.98 ± 0.00[a]	0.98 ± 0.00[a]	0.1889
Lipid (%)	1.23 ± 0.04[a]	1.19 ± 0.07[a]	0.4211

Results are expressed as mean ± standard error of the mean (SEM). [a] Means with common superscripts are similar ($p > 0.05$).

Protein oxidation

There was a harvest × storage interaction ($p = 0.015$; Table 2) for protein oxidation. WST steaks demonstrated greater ($p < 0.05$) carbonyl content than STU counterparts on zero-day (Table 2), whereas both treatments exhibited similar ($p > 0.05$) carbonyl content on days 3, 6 and 9.

STU steaks showed an increase ($p < 0.05$) in carbonyl content on day 9 compared to zero day (Table 2), whereas, in WST steaks, the carbonyl content remained unchanged ($p > 0.05$) throughout the storage.

The greater protein oxidation initially observed in WST steaks may be attributed to differences in pre-slaughter stress (Estévez, Ventanas, Heinonen, & Puolanne, 2011), which is known to increase the susceptibility to oxidation of postmortem muscles. The harvest without stunning contributes to animal stress (D'Agata et al., 2009), enhancing oxidative changes of amino acids in muscles (Estévez et al., 2011). This, in turn, leads to generation of protein cross-linking and polymerization, contributing to the increase of carbonyl content (Xue, Huang, Huang, & Zhou, 2013).

Table 2. Protein oxidation, springiness and cohesiveness of *longissimus lumborum* steaks (n=6) from Nellore (*Bos indicus*) cattle harvest with stunning (STU) or without stunning (WST) during storage at 4°C for 9 days.

Parameter		Days of storage				
		0	3	6	9	*P* value
Protein oxidation *	STU	1.42 ± 0.0^{b}	1.73 ± 0.1^{ab}	1.89 ± 0.1^{ab}	2.14 ± 0.1^{a}	0.0248
	WST	2.19 ± 0.0^{a}	2.19 ± 0.0^{a}	1.99 ± 0.0^{a}	2.17 ± 0.1^{a}	0.4704
P value		< 0.0001	0.0606	0.0966	0.8833	
Springiness (ratio)	STU	0.68 ± 0.01^{ab}	0.64 ± 0.01^{bc}	0.70 ± 0.01^{a}	0.67 ± 0.02^{ab}	0.2183
	WST	0.71 ± 0.01^{a}	0.66 ± 0.01^{abc}	0.72 ± 0.01^{a}	0.60 ± 0.01^{c}	< 0.0001
P value		0.1015	0.3266	0.3508	0.0416	
Cohesiveness (ratio)	STU	0.22 ± 0.00^{b}	0.24 ± 0.00^{b}	0.25 ± 0.00^{a}	0.25 ± 0.00^{a}	0.0001
	WST	0.18 ± 0.00^{c}	0.26 ± 0.01^{a}	0.26 ± 0.00^{a}	0.28 ± 0.00^{a}	< 0.0001
P value		0.0021	0.0474	0.6308	0.1470	

Results expressed as mean ± standard error of mean (SEM). $^{a-c}$Mean values followed by different superscripts, in the same row, are significantly different ($p < 0.05$). * Result expressed as nmol carbonyl per mg protein.

The increased protein oxidation observed in STU steaks may be attributed to postmortem biochemical changes, such as modification of cellular compartmentalization, release of iron, propagation of lipid oxidation and protein oxidation, which contribute to the increase of carbonyls during storage (Estévez et al., 2011).

In partial agreement, Sabow et al. (2016) verified a similar carbonyl content in *longissimus lumborum* muscle from goats harvested without stunning and their electrically stunned counterparts on days 1, 3, 7 and 14 of storage. Nevertheless, on zero day, these authors reported a lower carbonyl content in the *longissimus lumborum* muscle from non-stunned goats. The difference observed in carbonyl content on day zero may be attributed to differences in the stunning method (electrical stunning) used by the authors. Electrical stunning can result in inefficient bleeding and an increase of iron and myoglobin contents, known as highly oxidative agents in meat muscles (Sabow et al., 2016).

Regarding storage, Sabow et al. (2016) observed an increased carbonyl content in the *longissimus lumborum* muscle from goats harvested without stunning and their electrically stunned counterparts from day 3 to 14 of storage, which is in partial agreement with our results. Likewise, Nakyinsige et al. (2015) showed an increase in protein oxidation in the *longissimus lumborum* muscle from non-stunned rabbits during 14 days of storage.

The differences in carbonyl content observed in our study may be related to differences in the chemical composition of meat from different animal species, such as fatty acid composition and heme iron contents, which may have favored the oxidative process (Utrera & Estévez, 2013).

Texture profile

Hardness and chewiness

There was no effect of the harvest × storage interaction on hardness ($p = 0.878$; Table 3) and chewiness ($p = 0.518$; Table 3). However, there was an effect of storage ($p < 0.001$) on both parameters.

Huidobro et al. (2005) described hardness as the resistance at maximum compression during the first compression cycle; it is the force required to achieve sample deformation. Chewiness is the energy required to chew a solid sample to a steady state of swallowing and is influenced by hardness, cohesiveness and springiness (Huidobro et al., 2005).

STU and WST steaks demonstrated similar ($p > 0.05$) hardness and chewiness on days 0, 3, 6 and 9 (Table 3).

Table 3. Hardness and chewiness of *longissimus lumborum* steaks (n=6) from Nellore (*Bos indicus*) cattle harvest with stunning (STU) or without stunning (WST) during storage at 4°C for 9 days.

Parameter		Days of storage				
		0	3	6	9	*P* value
Hardness (N)	STU	222.78 ± 6.9^{abx}	195.31 ± 7.4^{bcdxyzw}	198.50 ± 6.14^{bcdx}	174.68 ± 4.3^{dx}	0.0002
	WST	226.14 ± 3.9^{axy}	198.91 ± 4.8^{abcdx}	201.20 ± 5.8^{abcx}	185.65 ± 6.9^{cdx}	< 0.0001
P value		0.6322	0.8582	0.8638	0.2443	
Chewiness (N × mm)	STU	32.23 ± 1.4^{ax}	27.76 ± 1.7^{abxy}	28.43 ± 1.7^{abxy}	24.87 ± 1.2^{by}	
	WST	32.71 ± 1.4^{ax}	30.64 ± 1.7^{abxy}	32.67 ± 0.8^{ax}	25.45 ± 1.4^{by}	
P value		0.9587	0.2409	0.0546	0.8551	

Results expressed as mean ± standard error of mean (SEM). $^{a-c}$ Mean values followed by different superscripts, in the same row, are significantly different ($p < 0.05$). $^{x-w}$ Mean values followed by different superscripts, in the same day of storage, are significantly different ($p < 0.05$).

Although previous investigations described that the harvest method increases the levels of adrenaline (Sabow et al., 2016) and cortisol (Wernicki et al., 2006), which may negatively affect the meat quality traits (Salim et al.,

2019), Pighin et al. (2013) reported a negligible effect of adrenaline and cortisol on meat toughness and chewiness, which may explain our results. Moreover, similar results for hardness and chewiness found in STU and WST steaks could also be attributed to sarcomere length (King et al., 2006) and cross-section area and thickness of the LL muscle fiber type (Lachowicz, Zochowska, & Sobczak, 2004).

Sarcomere length plays a key role in the texture profile of fresh meat and is influenced by muscle type and animal species (Ertbjerg & Puolanne, 2017). King et al. (2006) argued that cattle with excitable temperament, such as Nellore (*Bos indicus*), have shorter sarcomeres than those with calm behavior. Moreover, Lachowicz et al. (2004) observed a relationship between hardness and cross-section area and thickness of muscle fiber, in which the muscles with larger-sized fibers (type IIB fibers) such as LL (Hwang, Kim, Jeon, Hur, & Joo, 2010) exhibit greater toughness than muscles with smaller fiber (type I fibers) (Hwang et al., 2010). Moreover, Kolczak, Palka, and Lacki (2005) reported a correlation between chewiness and the content of muscle fiber type IIB contained in LL. This study evaluated samples from the same muscle (*longissimus lumborum*; LL) and animal breed (Nellore), which may have contributed to the similarity in hardness and chewiness values.

Supporting our results, Önenç and Kaya (2004) verified similar hardness and chewiness in longissimus muscle from stunned and non-stunned *Bos taurus* cattle on days 4, 7, and 14 of storage. Furthermore, Sazili et al. (2013) reported no differences in tenderness in *longissimus lumborum* from stunned and non-stunned *Bos taurus* x *Bos indicus* crossbred cattle during 14 days of storage.

Both STU and WST steaks demonstrated a decrease ($p < 0.05$) in hardness and chewiness during storage (Table 3), which could be attributed to the enzymatic degradation and muscle proteolysis (Huff-Lonergan, Zhang, & Lonergan, 2010; Sabow et al., 2016). During storage, the calpain system is responsible for degrading the myofibrillar and cytoskeletal proteins, which in turn contribute to a decrease in hardness and chewiness (Huff-Lonergan et al., 2010).

Accordingly, Önenç and Kaya (2004) observed a reduction in hardness and chewiness in longissimus muscle from stunned and non-stunned *Bos taurus* cattle during 14 days of storage. Likewise, Hayes et al. (2015) documented a decrease in hardness in longissimus steaks from non-stunned *Bos taurus* cattle during 10 days of storage. Moreover, Lokman, Sabow, Abubakar, Adeyemi, and Sazili (2017) reported a decrease in shear force in muscle semitendinosus from stunned and non-stunned goats during 7 days of retail display. A positive correlation was detected in the present study between hardness and chewiness ($r=0.80$; $p < 0.05$), which further reiterates the relationship between these parameters (Table 3).

Springiness

There was an effect of the harvest × storage interaction ($P = 0.042$; Table 2) for springiness. Springiness is the force obtained at maximum compression during the second compression cycle. In other words, it represents the sample hardness at the moment of the second bite (Huidobro et al., 2005).

STU steaks demonstrated greater ($p < 0.05$) springiness than WST counterparts on day 9, whereas similar ($p > 0.05$) springiness was observed in both (STU and WST) steaks on days 0, 3, and 6 (Table 2). The observed difference in springiness could be related to protein oxidation (Xia et al., 2018). Changes in protein structure and amino acid side chains could alter protein-protein interactions and their gel properties, negatively affecting springiness (Xia et al., 2018).

In partial agreement with our results, Addeen (2014) presented similar springiness in chicken patties obtained from stunned and non-stunned broilers on days 0 and 12 of storage. Moreover, Zhang et al. (2017) reported similar springiness in silver carp fillets obtained from stunned (percussion and ice immersion) and non-stunned carps on days 0, 1, 2, and 3 of storage.

Regarding storage, WST steaks demonstrated a decrease ($p < 0.05$) in springiness on day 9, compared to day 0 of storage (Table 2), whereas in STU steaks, springiness values remained overall unchanged ($p > 0.05$). The decline in springiness observed in WST steaks may be associated with muscle proteolysis and protein degradation, which contribute to the decrease in springiness during storage (Zhang et al., 2017). Moreover, we hypothesized that the non-stunned slaughter may have increased the release of stress hormones, which was not enough to influence the hardness and chewiness but may have contributed to weakening the muscle connections in the second compression cycle, in association with postmortem changes during storage, influencing springiness.

In partial agreement, Addeen (2014) reported a decrease in springiness in chicken patties obtained from both stunned and non-stunned broilers during 12 days of storage. In addition, Zhang et al. (2017) documented a decrease in springiness in silver carp fillets obtained from stunned and non-stunned animals during 3 days of storage.

Cohesiveness

There was a harvest × storage interaction ($p < 0.001$; Table 2) for cohesiveness. Cohesiveness is the strength of internal bonds making up the body of the sample (Huidobro et al., 2005). STU steaks exhibited greater ($p < 0.05$) cohesiveness than their WST counterparts on zero-day, whereas both treatments exhibited similar ($p > 0.05$) cohesiveness on days 3, 6, and 9.

Both STU and WST steaks demonstrated an increase ($p < 0.05$) in cohesiveness during storage. However, the increase was more pronounced in WST than in STU steaks (Table 2). The observed differences in cohesiveness between STU and WST steaks can be attributed to protein oxidation (Nakyinsige et al., 2015; Huff-Lonergan et al., 2010). Protein oxidation induces changes in conformational and functional properties of proteins (Xiong, 2000), which may increase cohesiveness (Cichoski et al., 2019).

In partial agreement, Addeen (2014) documented similar cohesiveness in chicken patties obtained from stunned and non-stunned broilers on day zero and day 12 of storage. These authors also reported an increase of cohesiveness in chicken patties obtained from non-stunned broilers from day zero to day 12, whereas their counterparts from stunned animals exhibited steady cohesiveness during storage.

Conclusion

The harvest method influenced some quality traits of LL steaks from *Bos indicus* (Nellore) cattle. STU and WST steaks exhibited similar proximate composition, hardness, and chewiness. WST steaks exhibited greater initial protein oxidation and lower cohesiveness than their STU counterparts. Furthermore, WST steaks demonstrated a decrease in springiness and a more pronounced increase in cohesiveness than their STU counterparts throughout the refrigerated storage period. These results suggest the necessity of further studies to evaluate the molecular influence of harvest on oxidative stability and texture profile of beef from *Bos indicus* cattle.

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References

Addeen, A. (2014). *Impact of halal slaughtering on quality and shelf-life of broiler chicken meat* (154f) (Doctoral Thesis). Food Science and Technology, Songkla University, Songkla.

Association of Official Analytical Chemists [AOAC]. (2012). *Official methods of analysis* (19th ed.). Washington, DC: AOAC International.

Armenteros, M., Heinonen, M., Ollilainen, V., Toldrá, F., & Estévez, M. (2009) Analysis of protein carbonyls in meat products by using the DNPH-method, fluorescence spectroscopy and liquid chromatography–electrospray ionisation–mass spectrometry (LC–ESI–MS). *Meat Science*, *83*(1), 104-112. DOI: https://doi.org/10.1016/j.meatsci.2009.04.007

Bligh, E. G., & Dyer, W. J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, *37*(8), 911-917. DOI: https://doi.org/10.1139/o59-099

Bostami, A. B. M. R., Mun, H. S., & Yang, C. J. (2018). Loin eye muscle physico-chemical attributes sensory evaluation and proximate composition in Korean Hanwoo cattle subjected to slaughtering along with stunning with or without pithing. *Meat Science*, *145*, 220-229. DOI: https://doi.org/10.1016/j.meatsci.2018.06.032

Botinestean, C., Keenan, D. F., Kerry, J. P., & Hamill, R. M. (2016). The effect of thermal treatments including sous-vide, blast freezing and their combinations on beef tenderness of M. semitendinosus steaks targeted at elderly consumers. *LWT - Food Science and Technology*, *74*, 154-159. DOI: https://doi.org/10.1016/j.lwt.2016.07.026

Brasil, Ministério da Agricultura e Pecuária. (2017). Decreto n. 9.013, de 29 de Março de 2017. Regulamenta a Lei n. 1.283, de 18 de Dezembro de 1950, e a Lei n. 7.889, de 23 de Novembro de 1989, que dispõem sobre a inspeção industrial e sanitária de produtos de origem animal. *Diário Oficial da União* de 30/03/2017 Seção 1, Página 3.

Choe, J. H., Choi, M. H., Rhee, M. S., & Kim, B. C. (2016). Estimation of sensory pork loin tenderness using Warner-Bratzler shear force and texture profile analysis measurements. *Asian-Australasian Journal of Animal Sciences*, *29*(7), 1029-1036. DOI: https://doi.org/10.5713/ajas.15.0482

Cichoski, A. J., Silva, M. S., Leães, Y. S. V., Brasil, C. C. B., Menezes, C. R., Barin, J. S., ... Campagnol, P. C. B. (2019). Ultrasound: A promising technology to improve the technological quality of meat emulsions. *Meat Science*, *148*, 150-155. DOI: https://doi.org/10.1016/j.meatsci.2018.10.009

D'Agata, M., Russo, C., & Preziuso, G. (2009). Effect of Islamic ritual slaughter on beef quality. *Italian Journal of Animal Science*, *8*(2), 489-491. DOI: https://doi.org/10.4081/ijas.2009.s2.489

Ertbjerg, P., & Puolanne, E. (2017). Muscle structure sarcomere length and influences on meat quality: A review. *Meat Science*, *132*, 139-152. DOI: https://doi.org/10.1016/j.meatsci.2017.04.261

Estévez, M., Ventanas, S., Heinonen, M., & Puolanne, E. (2011). Protein carbonylation and water-holding capacity of pork subjected to frozen storage: effect of muscle type premincing and packaging. *Journal of agricultural and food chemistry*, *59*(10), 5435-5443. DOI: https://doi.org/10.1021/jf104995j

Farouk, M. M. (2013). Advances in the industrial production of halal and kosher red meat. *Meat science*, *95*(4), 805-820. DOI: https://doi.org/10.1016/j.meatsci.2013.04.028

Farouk, M. M., Al-Mazeedi, H. M., Sabow, A. B., Bekhit, A. E. D., Adeyemi, K. D., Sazili, A. Q., & Ghani, A. (2014). Halal and kosher slaughter methods and meat quality: A review. *Meat Science*, *98*(3), 505-519. DOI: https://doi.org/10.1016/j.meatsci.2014.05.021

Francisco, C. L., Resende, F. D., Benatti, J. M. B., Castilhos, A. M., Cooke, R. F., & Jorge, A. M. (2015). Impacts of temperament on Nellore cattle: Physiological responses feedlot performance and carcass characteristics. *Journal of Animal Science*, *93*(11), 5419-5429. DOI: https://doi.org/10.2527/jas.2015-9411

González-Fuentes, H., Hamedy, A., von Borell, E., Luecker, E., & Riehn, K. (2014). Tenacity of *Alaria alata mesocercariae* in homemade German meat products. *International Journal of Food Microbiology*, *176*, 9-14. DOI: https://doi.org/10.1016/j.ijfoodmicro.2014.01.020

Hayes, N. S., Schwartz, C. A., Phelps, K. J., Borowicz, P., Maddock-Carlin, K. R., & Maddock, R. J. (2015). The relationship between pre-harvest stress and the carcass characteristics of beef heifers that qualified for kosher designation. *Meat Science, 100*, 134-138. DOI: https://doi.org/10.1016/j.meatsci.2014.09.145

Huff-Lonergan, E., Zhang, W., & Lonergan, S. M. (2010). Biochemistry of postmortem muscle — Lessons on mechanisms of meat tenderization. *Meat Science*, *86*(1), 184-195. DOI: https://doi.org/10.1016/j.meatsci.2010.05.004

Huidobro, F. R., Miguel, E., Blázquez, B., & Onega, E. (2005). A comparison between two methods (warner–bratzler and texture profile analysis) for testing either raw meat or cooked meat. *Meat Science*, *69*(3), 527-536. DOI: https://doi.org/10.1016/j.meatsci.2004.09.008

Hwang, Y. H., Kim, G. D., Jeon, J. Y., Hur, S. J., & Joo, S. T. (2010). The relationship between muscle fiber characteristics and meat quality traits of highly marbled *Hanwoo* (Korean native cattle) steers. *Meat Science*, *86*(2), 456-461. DOI: https://doi.org/10.1016/j.meatsci.2010.05.034

Kim, G. D., Lee, H. S., Jung, E. Y., Lim, H. J., Seo, H. W., Lee, Y. H., ... Yang, H. (2013). The effects of CO_2 gas stunning on meat quality of cattle compared with captive bolt stunning. *Livestock Science*, *157*(1), 312-316. DOI: https://doi.org/10.1016/j.livsci.2013.05.025

King, D. A., Pfeiffer, S. C. E., Randel, R. D., Welsh Jr, T. H., Oliphint, R. A., Baird, B. E., ... Savell, J. W. (2006). Influence of animal temperament and stress responsiveness on the carcass quality and beef tenderness of feedlot. cattle. *Meat science*, *74*(3), 546-556. DOI: https://doi.org/10.1016/j.meatsci.2006.05.004

Kolczak, T., Palka, K., & Lacki, J. (2005). Water retention shear force and texture parameters of cattle psoas and semitendinosus muscles unfrozen and frozen during post-mortem ageing. *Polish Journal of Food and Nutrition Sciences*, *55*(1), 17-26.

Lachowicz, K., Zochowska, J., & Sobczak, M. (2004). Comparison of the texture and structure of selected muscles of piglets and wild boar juveniles. *Polish Journal of Food and Nutrition Sciences*, *54*(1), 75-79.

Linares, M. B., Berruga, M. I., Bornez, R., & Vergara, H. (2007). Lipid oxidation in lamb meat: Effect of the weight handling previous slaughter and modified atmospheres. *Meat Science*, *76*(4), 715-720. DOI: https://doi.org/10.1016/j.meatsci.2007.02.012

Lokman, N. S., Sabow, A. B., Abubakar, A. A., Adeyemi, K. D., & Sazili, A. Q. (2017). Comparison of carcass and meat quality in goats subjected to preslaughter head-only electrical stunning or slaughtered without stunning. *CyTA - Journal of Food*, *15*(1), 99-104. DOI: https://doi.org/10.1080/19476337.2016.1217049

Mancini, R. A., & Hunt, M. C. (2005). Current research in meat color. *Meat Science*, *71*(1), 100-121. DOI: https://doi.org/10.1016/j.meatsci.2005.03.003

Mercier, Y., Gatellier, P., Viau, M., Remignon, H., & Renerre, M. (1998). Effect of dietary fat and vitamin E on colour stability and on lipid and protein oxidation in turkey meat during storage. *Meat Science*, *48*(3-4), 301-318. DOI: https://doi.org/10.1016/S0309-1740(97)00113-7

Nakyinsige, K., Sazili, A. Q., Aghwan, Z. A., Zulkifli, I., Goh, Y. M., Abu Bakar, F., & Sarah, S. A. (2015). Development of microbial spoilage and lipid and protein oxidation in rabbit meat. *Meat Science*, *108*, 125-131. DOI: https://doi.org/10.1016/j.meatsci.2015.05.029

Neethling, N. E., Sigge, G. O., Hoffman, L. C., Suman, S. P., & Hunt, M. C. (2017). Exogenous and endogenous factors influencing color of fresh meat from ungulates. *Meat and Muscle Biology*, *1*(1), 253-275. DOI: https://doi.org/10.22175/mmb2017.06.0032

Oliver, C. N., Ahn, B. W., Moerman, E. J., Goldstein, S., & Stadtman, E. R. (1987). Age-related changes in oxidized proteins. *Journal of Biological Chemistry*, *262*(12), 5488-5491.

Önenç, A., & Kaya, A. (2004). The effects of electrical stunning and percussive captive bolt stunning on meat quality of cattle processed by Turkish slaughter procedures. *Meat Science*, *66*(4), 809-815.

Pighin, D. G., Cunzolo, S. A., Zimerman, M., Pazos, A. A., Domingo, E., Pordomingo, A. J., & Grigioni, G. (2013). Impact of adrenaline or cortisol injection on meat quality development of Merino hoggets. *Journal of Integrative Agriculture*, *12*(11), 1931-1936. DOI: https://doi.org/10.1016/S2095-3119(13)60630-9

Sabow, A. B., Zulkifli, I., Goh, Y. M., Ab Kadir, M. Z., Kaka, U., Imlan, J. C., ... Sazili, A. Q. (2016). Bleeding efficiency microbiological quality and oxidative stability of meat from goats subjected to slaughter without stunning in comparison with different methods of pre-slaughter electrical stunning. *PLoS One*, *12*(5), 1-18. DOI: https://doi.org/10.1371/journal.pone.0152661

Salim, A. P. A. A., Suman, S. P., Viana, F. M., Monteiro, M. G., Panzenhagen, P. H., Canto, A. C. V. C. S., & Conte-Junior, C. A. (2019). Harvest method influences color stability of *longissimus lumborum* steaks from *Bos Indicus* cattle. *Meat and Muscle Biology*, *3*(1), 33-41. DOI: https://doi.org/10.22175/mmb2018.07.0021

Sazili, A. Q., Norbaiyah, B., Zulkifli, I., Goh, Y. M., Lotfi, M., & Small, A. H. (2013). Quality assessment of longissimus and semitendinosus muscles from beef cattle subjected to non-penetrative and penetrative percussive stunning methods. *Asian-australasian journal of animal sciences*, *26*(5), 723-731. DOI: https://doi.org/10.5713/ajas.2012.12563

Suman, S. P., Hunt, M. C., Nair, M. N., & Rentfrow, G. (2014). Improving beef color stability: Practical strategies and underlying mechanisms. *Meat Science*, *98*(3), 490-504. DOI: https://doi.org/10.1016/j.meatsci.2014.06.032

United States Department of Agriculture [USDA]. (2022). *Brazil: Livestock and Products Semi-annual 2020 Semi-Annual Livestock Report.* Retrieved from https://gainfasusdagov/Recent%20GAIN%20Publications/Livestock%20and%20Products%20Semi-annual_Brasilia_Brazil_2-28-2018pdf

Utrera, M., & Estévez, M. (2013). Oxidative damage to poultry pork and beef during frozen storage through the analysis of novel protein oxidation markers. *Journal of Agriculture and Food Chemistry*, 61(33), 7987-7993. DOI: https://doi.org/10.1021/jf402220q

Wernicki, A., Urban-chmiel, R., Kankofer, M., Mikucki, P., Puchalski, A., & Tokarzewski, S. (2006). Evaluation of plasma cortisol and TBARS levels in calves after short-term transportation. Retrieved from https://wwwrevmedvetcom/2006/RMV157_30_34pdf

Xia, M., Chen, Y., Guo, J., Feng, X., Yin, X., Wang, L., ... Ma., J. (2018). Effects of oxidative modification on textural properties and gel structure of pork myofibrillar proteins. *Food Research International*, *121*, 678-683. DOI: https://doi.org/10.1016/j.foodres.2018.12.037

Xiong, Y. L. (2000). Protein oxidation and implications for muscle food quality. In E. Decker, C. Faustman, & C. J. Lopez-Bote (Orgs.), *Antioxidants in muscle foods* (p. 85-111). New York, USA: Wiley-Interscience.

Xue, M., Huang, F., Huang, M., & Zhou, G. (2013). Influence of oxidation on myofibrillar proteins degradation from bovine via μ-calpain. *Food Chemistry*, *134*(1), 106-112. DOI: https://doi.org/10.1016/j.foodchem.2012.02.072

Zhang, L., Li, Q., Lyu, J., Kong, C., Song, S., & Luo, Y. (2017). The impact of stunning methods on stress conditions and quality of silver carp (*Hypophthalmichthys molitrix*) fillets stored at 4°C during 72h postmortem. *Food chemistry*, *216*(1), 130-137. DOI: https://doi.org/10.1016/j.foodchem.2016.08.004