Mass transfer in beef: effect of crossbreeding and ultrasound application

Marcio Augusto Ribeiro Sanches, Patrícia Maria Onofre Colombo Silva, Roger Darros Barbosa, Javier Telis Romero, Andrea Carla da Silva Barretto*

ABSTRACT: Cuts of muscle (biceps femoris) from three crossbreeds between the Nelore, Angus and Wagyu breeds of cattle (Crossbreed 1 (C1): ½ Angus and ½ Nelore; Crossbreed 2 (C2): ¾ Angus and ¼ Nelore; Crossbreed 3 (C3): ½ Wagyu, ¼ Nelore and ¼ Angus) were characterized by their chemical composition, pH, water activity and lipid oxidation. The cuts were submitted to wet brining in a 5% NaCl concentration at 5°C with either static brine (SB) or brine assisted with ultrasound (US). Samples of the treatments were taken after 30, 60, and 120 min of wet brining for later analysis. The experimental data were adjusted using the Peleg and Page models, and the Page template best described the experimental data. The crossbreeding did not affect the water and ash content; however, it significantly affected the levels of lipids and proteins. C3 presented higher fat content than C2, which, in turn, was higher than C1. C1 had higher protein content than C2, which had more protein than C3. When ultrasound was applied, crossbreeding influenced the water content and the water absorption rate during brining, which had the highest values for the highest protein content. The crossbreeding and ultrasound application also affected the NaCl content of the cuts. However, only ultrasound application increased the rate of NaCl absorption during wet brining. The results demonstrate that ultrasound accelerates the mass transfer in wet brining of the cuts of beef, regardless of the crossbreeds studied.

Keywords: NaCl, Wagyu, Angus, Nelore, water gain

Introduction

Salting is one of the oldest methods for processing and storing food thus one of the key steps in the preparation of a variety of salted meat products, many of which consumed in different countries.

In recent years, increased demand for animal proteins had driven beef production, due to a concomitant increase in population (Salami et al., 2019). Diet and breed (or crossbreeds) contribute to meat quality [Avilés et al., 2015]. In this context, beef cattle crossbreeding is an important option for improving meat quality [Lage et al., 2012]. This quality is related to the amount of intramuscular fat, also called marbling, a characteristic mainly associated to the genetic groups of certain breeds. Crosses between breeds, like Angus and Wagyu, lead to improvements in sensory characteristics of meat, such as juiciness and flavor [Frank et al., 2016; Motoyama et al., 2016], increasing acceptance by consumers. However, it is unclear whether crossbreeding can affect mass transfer kinetics during the salting process of meat products.

Among the salted beef products, jerked beef and sun-dried beef are products that can be transported and stored without refrigeration, due to the intermediate water activity. However, manufacture of these products is still a highly practical, experience-based process, involving long salting and drying stages [Vidal et al., 2019]. The time factor changes processing techniques, giving rise to so-called emerging technologies, which generally aim to optimize these production processes [Alarcon-Rojo et al., 2019; Chandrapala et al., 2012; Leonelli and Mason, 2010].

Different studies have used vacuum pulsing [Martins et al., 2019], ohmic heating [Coelho et al., 2019], electrodialysis [Xue et al., 2018], high pressure [Shinwari and Rao, 2018], microwave [Leonelli and Mason, 2010], and high-power ultrasound [Polachini et al., 2019; Goula et al., 2017], in order to accelerate mass transfer rates. Ultrasound is a promising emerging technology in the food processing industry, as it is innovative, achieving important results generally complementary to classical techniques, such as salting [Sharma and Dash, 2019]. There are no studies in the literature that assessed the influence of crossbreeding on mass transfer kinetics during the beef curing process in static brine and in brine with ultrasound. Thus, this study was conducted to evaluate the effect of crossbreeding of Nelore, Angus, and Wagyu cattle and the use of ultrasound on beef wet brining.

Materials and Methods

Raw materials

Cuts of muscle (biceps femoris) from three genetic crossbreeds of Nelore, Angus and Wagyu were used. The crossbreeding resulted in animals named here as Cross 1 - C1: ½ Angus and ½ Nelore; Cross 2 - C2: ¾ Angus and ¼ Nelore; Cross 3 - C3: ½ Wagyu, ¼ Nelore and ¼ Angus, all produced by a company, located in Nhandaera, São Paulo, Brazil (20° 41’ 53” S, 50° 2’ 33” W, altitude of 504 m). The animals were slaughtered at a slaughterhouse in Ipoá, São Paulo, Brazil (23° 31’ 4” S, 46° 20’ 38” W, altitude of 745 m). Samples from six different animals were collected for each crossbreed: muscle (biceps femoris) steaks 2.54 cm thick were cut...
from each animal for each crossbreed. The samples were frozen [−18 °C] until the wet curing process started.

Chemical composition of raw materials
The contents of water, protein, and ash were determined in accordance to the methods described by AOAC (2007). The lipid content was determined according to the method described by Bligh and Dyer (1959), with the removal of subcutaneous fat. The pH of the samples was determined in triplicate using a digital pH meter. The water activity of the samples was determined using a digital hygrometer (Aqualab, model 3).

Preparation of samples and brine
The samples were thawed for 24 h at 4 °C, cut into slices of similar weight and size, with approximately 50 grams each, size 30 × 100 × 20 mm, [width, length and height] respectively. The subcutaneous fat and connective tissue was removed before curing. The brine was prepared using distilled water and 50 g L−1 sodium chloride. The weight ratio of meat to brine was set at 1:10 to ensure constant solution concentration during the process.

Treatment by wet brining
The processes of static brining (SB) and salting with ultrasound application (US) were performed in a cylindrical stainless-steel vat [21 cm diameter, 42 cm height] immersed in a thermostatic bath with controlled temperature 5 ± 1 °C. After 30, 60, and 120 min of wet brining, the samples were taken for analytical determinations. All wet brining processes were performed separately for each sample and no significant increase in temperature was observed during the trials.

For brining with ultrasound, we used a VCX-1500 ultrasound processor that emits waves at a frequency of 20 kHz. The processor was equipped with a Ti-6Al-4V titanium probe with 2.5 cm in diameter that emits axial and radial ultrasound. The ultrasound power was adjusted to 600 W, as described in Barretto et al. (2018a). The probe was immersed in brine and positioned in the center of the container. The tip of the probe was immersed in the brine, with no direct contact between the probe and the beef cut. The muscle fibers were arranged parallel to the ultrasonic waves.

Analytical determination after wet brining
After wet brining in SB and US, the water content was determined by drying at 105 °C in a constant weight oven AOAC (2007) and the NaCl content by quantification of chlorides according to the Möhr method AOAC (2007) for samples wet brined for 30, 60, and 120 min.

Mathematical modeling
Peleg model
The equation proposed by Peleg (1988) is used to describe the kinetics of water sorption that approaches equilibrium. The model used in the present work was given by Eq. (1).

\[ C_x = C_{x0} \pm \frac{t}{k_1 + k_2 \cdot t} \]  

In the above equation, “±” becomes “+” if the process is hydration and “−” if the process is dehydration. \( C_x \) is the content water or NaCl [% bs] at time \( t \), \( C_{x0} \) is the initial water or NaCl content [% bs], \( t \) is the process time, \( k_1 \) and \( k_2 \) are constant in the Peleg model. The reciprocal value of \( k_1 \) refers respectively to the initial rate \( [t = 0] \) of the water or salt content [Eq. (2)].

\[ \frac{d[C_x]}{dt} \bigg|_{k_1} = \frac{1}{k_1} \]  

The reciprocal value of \( k_2 \) allows to determine the water content or the salt content in the ‘equilibrium’ \( (C_x \rightarrow \infty) \) [Eq. (3)].

\[ C_x \rightarrow \infty \pm \frac{1}{k_2} \]  

Page model
Page (1949) developed an equation to describe the drying of corn kernels. Currently, it is one of the most used equation to describe food hydration [Simpson et al., 2017], described by Eq. (4).

\[ \frac{X_t - X_e}{X_0 - X_e} = e \left( -k_p \cdot t^n \right) \]  

where \( X_t \) is the moisture at time \( t \) [% dry base - db]; \( X_e \) is the equilibrium moisture [% db]; \( X_0 \) the initial moisture [% db]; \( t \) is the time [h]; \( k_p \) is the absorption rate of the process [h−¹] and \( n \) is a dimensionless constant.

Statistical Analysis
Results were expressed as the mean and the standard error of mean. The treatment method [MT] (i.e. static brine - SB or brine with ultrasound - US), the cross between the breeds (CR) and the treatment time (TT) were defined as factors in the statistical analysis. A factor or its interaction was considered significant when \( p \) values were lower than 0.05. The data obtained were analyzed statistically using the generalized linear model (GLM) and the means compared using the Tukey test \( (p < 0.05) \).

The adjustment of the models to the experimental data of water content and NaCl content was performed using a non-linear estimation procedure of the Statistica software (Statistica, version 7.0). To assess the fit of the model to the experimental data, the percentage of explained variance [%VAR] was calculated using Eq. (3) [Kang et al., 2016; Sanjuán et al., 1999] and the calculated root mean square error (RMSE) by Eq. (4) [Schmidt et al., 2009].

\[ \%VAR = 1 - \frac{S^2_{Cal}}{S^2_{Exp}} \]  

where \( S^2_{Cal} \) and \( S^2_{Exp} \) are the calculated values and the experimental data values respectively.
Results and Discussion

Chemical Composition

Table 1 shows the effect of crossbreeding between three bovine breeds (Angus, Wagyu, and Nelore) on the chemical composition of the muscle cuts (biceps femoris) and their pH and water activity. Crossbreeding did not influence \( p > 0.05 \) the water and ash content, however, it significantly affected the contents of lipids and proteins \( p < 0.05 \). Rossato et al. (2010) also found no difference in water activity. Crossbreeding did not influence \( p > 0.05 \) the lipid content is the parameter with the greatest variation in the centesimal composition of the meat. When it increases, other parameters decrease, such as moisture, ash, or protein. Maggioni et al. (2010) also found a higher percentage of protein in Nelore cattle when compared to crossbred animals. Regarding the pH and water activity, crossbreeding showed no significant influence \( p > 0.05 \). The results obtained for pH and water activity are similar to those found by Bampi et al. (2016) in beef samples, with the pH at 5.6 and the water activity at 0.99.

Effect of crossbreeding and use of ultrasound on water transport in beef wet brining

Table 2 shows the effect of crossbreeding between three cattle breeds and the use of ultrasound on water transport of sample cuts after different wet brining times. Regarding the initial water content of the samples, there was no difference \( p > 0.05 \) between the crossbreeds. After 30 min of wet brining, the treatment method, the crossbreds and the interaction between these variables all had a significant effect \( p < 0.05 \), where C1-US and C2-US had the highest water content. In contrast, C3-US and C2-SB and C3-SB did not differ \( p > 0.05 \) from each other, showing the lowest water content.

Table 1 – Chemical composition, pH, and water activity in beef.

| Composition (%) | C1     | C2     | C3     | SEM   | p value |
|-----------------|--------|--------|--------|-------|---------|
| Moisture        | 63.56a | 63.98b | 63.29a | 0.561 | 0.907   |
| Ash             | 0.84a  | 0.84a  | 0.86a  | 0.064 | 0.990   |
| Protein         | 20.59a | 19.66b | 17.24a | 0.355 | < 0.01  |
| Lipid           | 13.89a | 15.75b | 18.57a | 0.709 | < 0.01  |
| pH              | 5.68a  | 5.72a  | 5.71a  | 0.011 | 0.467   |
| Aw              | 0.984a | 0.985a | 0.986a | 0.004 | 0.100   |

For the fat content, C3 was greater than C2 \( p < 0.05 \) and C2 was greater than C1 \( p < 0.05 \). This difference is due to the crossbreeding with Wagyu cattle and its intense marbling (Motoyama et al., 2016; Yamada et al., 2014) mainly when compared to C1, which contains the largest contribution from the Nelore genetic group. Dias et al. (2016) also found higher fat content in Wagyu steaks when compared to meat from Nelore cattle. Duarte et al. (2013) studied the intramuscular fat content of Angus and Wagyu cattle managed under the same conditions and concluded that the intramuscular fat content was higher in Wagyu cattle.

For protein, C1 presented a significantly higher content \( p < 0.05 \) when compared to C2, and C2 showed a significantly higher content \( p < 0.05 \) compared to C3. The lower protein content of C3 is probably linked to its high lipid content, which, according to Lawrie (2005) the lipid content is the parameter with the greatest variation in the centesimal composition of the meat. When it increases, other parameters decrease, such as moisture, ash, or protein. Maggioni et al. (2010) also found a higher percentage of protein in Nelore cattle when compared to crossbred animals. Regarding the pH and water activity, crossbreeding showed no significant influence \( p > 0.05 \). The results obtained for pH and water activity are similar to those found by Bampi et al. (2016) in beef samples, with the pH at 5.6 and the water activity at 0.99.

Table 2 – Effect of crossbreeding and application of ultrasound in water transport (%) in beef.

| Time (min) | Static Brine (SB) | Ultrasound brine (US) | SEM 1 | MT | CR | MT × CR |
|------------|------------------|-----------------------|-------|----|----|---------|
|            | C1    | C2    | C3    | C1    | C2    | C3    |       |
| 0          | 62.89a | 63.18a | 62.72a | 62.89a | 63.18a | 62.72a | 0.226  |
| 30         | 63.09a | 63.04a | 62.66a | 63.84a | 64.21a | 62.40a | 0.130  |
| 60         | 64.52a | 63.71a | 63.23a | 65.03a | 64.73a | 63.23a | 0.186  |
| 120        | 65.89a | 65.16a | 63.17a | 68.42a | 67.68a | 64.64a | 0.334  |
| p value    | < 0.01 | 0.019 | 0.708 | < 0.01 | < 0.01 | < 0.01 |       |

For protein, C1 presented a significantly higher content \( p < 0.05 \) when compared to C2, and C2 showed a significantly higher content \( p < 0.05 \) compared to C3. The lower protein content of C3 is probably linked to its high lipid content, which, according to Lawrie (2005) the lipid content is the parameter with the greatest variation in the centesimal composition of the meat. When it increases, other parameters decrease, such as moisture, ash, or protein. Maggioni et al. (2010) also found a higher percentage of protein in Nelore cattle when compared to crossbred animals. Regarding the pH and water activity, crossbreeding showed no significant influence \( p > 0.05 \). The results obtained for pH and water activity are similar to those found by Bampi et al. (2016) in beef samples, with the pH at 5.6 and the water activity at 0.99.

Table 2 – Effect of crossbreeding and application of ultrasound in water transport (%) in beef.

| Time (min) | Static Brine (SB) | Ultrasound brine (US) | SEM 1 | MT | CR | MT × CR |
|------------|------------------|-----------------------|-------|----|----|---------|
|            | C1    | C2    | C3    | C1    | C2    | C3    |       |
| 0          | 62.89a | 63.18a | 62.72a | 62.89a | 63.18a | 62.72a | 0.226  |
| 30         | 63.09a | 63.04a | 62.66a | 63.84a | 64.21a | 62.40a | 0.130  |
| 60         | 64.52a | 63.71a | 63.23a | 65.03a | 64.73a | 63.23a | 0.186  |
| 120        | 65.89a | 65.16a | 63.17a | 68.42a | 67.68a | 64.64a | 0.334  |
| p value    | < 0.01 | 0.019 | 0.708 | < 0.01 | < 0.01 | < 0.01 |       |

For protein, C1 presented a significantly higher content \( p < 0.05 \) when compared to C2, and C2 showed a significantly higher content \( p < 0.05 \) compared to C3. The lower protein content of C3 is probably linked to its high lipid content, which, according to Lawrie (2005) the lipid content is the parameter with the greatest variation in the centesimal composition of the meat. When it increases, other parameters decrease, such as moisture, ash, or protein. Maggioni et al. (2010) also found a higher percentage of protein in Nelore cattle when compared to crossbred animals. Regarding the pH and water activity, crossbreeding showed no significant influence \( p > 0.05 \). The results obtained for pH and water activity are similar to those found by Bampi et al. (2016) in beef samples, with the pH at 5.6 and the water activity at 0.99.

Effect of crossbreeding and use of ultrasound on water transport in beef wet brining

Table 2 shows the effect of crossbreeding between three cattle breeds and the use of ultrasound on water transport of sample cuts after different wet brining times. Regarding the initial water content of the samples, there was no difference \( p > 0.05 \) between the crossbreeds. After 30 min of wet brining, the treatment method, the crossbreds and the interaction between these variables all had a significant effect \( p < 0.05 \), where C1-US and C2-US had the highest water content. In contrast, C3-US and C2-SB and C3-SB did not differ \( p > 0.05 \) from each other, showing the lowest water content.
After 60 min of treatment, there was a significant effect \( p < 0.05 \) from crossbreeding, where C1-US differed \( p < 0.05 \) from C3-US and C3-SB, but did not differ \( p > 0.05 \) from C2-US, C1-SB and C2-SB. After 120 min, only the treatment method and crossbreeding showed significant effect \( p < 0.05 \), where C1-US and C2-US had the highest water content \( p < 0.05 \); however, they did not differ from each other \( p > 0.05 \).

The results indicate that ultrasound use and crossbreeding significantly affected the water content in beef. Ultrasound increased the water content significantly with processing time and especially when associated with crossbreeds C1 and C2. This effect was possibly attributed to ultrasound causing asymmetric implosion cavitation bubbles near the solid surface of the cuts, generating microjets, which cause microinjections of brine to the meat surface affecting mass transfer [Alarcon-Rojo et al., 2019; Cárcel et al., 2007b].

The cuts composition may have also influenced these results, considering that C1 and C2 presented higher water content after brining and lower lipid content \( p < 0.05 \) when compared to C3. The higher intramuscular lipid content of C3 probably made water transport difficult. Water can bind strongly to food solid constituents and establish ionic bonds mainly with hydrophilic molecules, such as proteins, different from hydrophobic molecules, as is the case of fats, which are of a nonpolar nature [Damodaran et al., 2010].

Barretto et al. [2018b] studied cooked ham sorption isotherms and observed that ham with higher fat content had lower equilibrium moisture content when compared to low fat ham. As all samples from this experiment gained water at a 5 % NaCl concentration, this fact can be attributed to the salting-in phenomenon of proteins. In this phenomenon, the increase of protein solubility at low concentrations of salts in solution contributes to solubilization and hydration of muscle proteins, causing saline ions to interact with ionic charges of proteins increasing the available number of charges and the amount of water molecules attached to the protein ionsphere, causing greater water retention in beef and consequently increased water content [Schmidt et al., 2009].

### Effect of crossbreeding and use of ultrasound on NaCl transport in wet brining of beef

Table 3 shows the effect of crossbreeding and the use of ultrasound on NaCl transport after different wet brining times. Initially, there was no difference \( p > 0.05 \) in the contents of NaCl between the crossbreeds. After 30 min of treatment, there was a significant effect \( p < 0.05 \) from treatment method, where C1-US and C3-US had the highest NaCl content, differing from C3-SB.

After 60 min of wet brining, the use of ultrasound and crossbreeding both showed significant effects, where C1-US, C2-US and C3-US did not differ \( p < 0.05 \) from C3-SB while C3-SB differed \( p < 0.05 \) only from C1-SB. After 120 min of treatment, there was a significant effect of ultrasound treatment, crossbreeding, and the interaction between these two variables. C2-US and C3-US presented the highest \( p < 0.05 \) NaCl contents, different from C1-SB and C2-SB, which presented the lowest NaCl contents.

The results suggest that ultrasound use, crossbreeding, and interaction between these factors can significantly influence the NaCl content in beef. In brine with ultrasound application, there was also interaction between the treatment time (TT) and crossbreeding. The NaCl content in meat increased with treatment time, both in static brine and brine with ultrasound. Looking at the same treatment times, ultrasound samples presented higher NaCl content when compared to static brining. In this case, the effect of ultrasound can also be explained based on induced cavitation in the liquid medium.

| Time (min) | Static Brine (SB) | Ultrasound brine (US) | SEM 1 | p value |
|-----------|------------------|-----------------------|-------|---------|
|           | C1 | C2 | C3 | C1 | C2 | C3 | MT | CR | MT × CR |
| 0         | 0.031<sup>AD</sup> | 0.022<sup>BD</sup> | 0.022<sup>BD</sup> | 0.031<sup>AD</sup> | 0.022<sup>BD</sup> | 0.022<sup>BD</sup> | 0.002 | 0.461 | 0.422 | 0.164 |
| 30        | 0.398<sup>CD</sup> | 0.399<sup>C</sup> | 0.398<sup>CD</sup> | 0.451<sup>CD</sup> | 0.409<sup>B</sup> | 0.433<sup>C</sup> | 0.007 | <0.01 | 0.110 | 0.418 |
| 60        | 0.531<sup>BC</sup> | 0.541<sup>B</sup> | 0.591<sup>AB</sup> | 0.624<sup>B</sup> | 0.600<sup>BC</sup> | 0.642<sup>B</sup> | 0.008 | <0.01 | <0.01 | 0.301 |
| 120       | 0.832<sup>A</sup> | 0.821<sup>A</sup> | 0.860<sup>AB</sup> | 0.875<sup>A</sup> | 0.929<sup>A</sup> | 0.985<sup>A</sup> | 0.012 | <0.01 | <0.01 | 0.039 |
| p value   | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 | <0.01 |      |      |      |

Table 3 – Effect of crossbreeding and use of ultrasound in NaCl (%) transport in beef.

- <sup>a</sup>c Mean values in the same row not followed by the same letter differ significantly \( p < 0.05 \); AD Mean values in the same column not followed by the same letter differ significantly \( p < 0.05 \); C1 = (½ Angus and ½ Nelore); C2 = (¾ Angus and ¼ Nelore); C3 = (½ Wagyu, ¼ Nelore and ¼ Angus); MT = Method of treatment; CR = Crossbreeding; TT = Time of treatment; SEM 1 = Mean standard error for method of treatment (MT) and crossbreeding (CR) at each treatment time;SEM 2 = Mean standard error for treatment time and crossbreeding for static brine (SB) and ultrasound brine (US).
which leads to the formation of micro-jets that cause brine microinjection into the meat surface (Cárcel et al., 2007b; Kang et al., 2016).

The results of the effect of crossbreeding for the three crossbreeds treated in static brine did not differ. However, when ultrasound is used, samples from the C3-US differ from all three crossbreeds treated with static brine and C1-US. One possibility is that crossbreeding can provide differences to the diameter of muscle fibers. Arrigoni et al. (2004) studied performance, muscle fibers, and meat from three crossbreeds of cattle (Angus × Nelore; Canchim × Nelore; and Simental × Nelore) and concluded that Angus × Nelore crossbreed cattle had smaller diameter muscle fiber when compared to the other two groups. Duarte et al. (2013) reported that muscle fiber diameter was larger in Wagyu cattle than in Angus cattle. According to Joo et al. (2013), muscle fiber characteristics depend on several factors, such as gender (Ozawa et al., 2000), diet (Ha et al., 2012), growth performance (Kim et al., 2013), muscle location (Hwang et al., 2010) and breed (Ryu et al., 2008).

In addition, treatment with ultrasound may be causing mechanical effects resulting from the cavitation process, which, in turn, can increase gaps between the muscle fibers of C2-US and C3-US samples, increasing the transfer of NaCl. Siró et al. (2009) reported a greater distance between muscle fibers of samples treated with sonicated brine when compared to samples treated with static brine and that this distance increased with increasing treatment time and ultrasound intensity. Therefore, there was possibly interaction between crossbreeds C2-US and C3-US, increasing the mass transfer rate.

**Mathematical modeling**

**Peleg model**

Table 4 presents the results of the Peleg model adjusted by non-linear regression for the water and NaCl contents during beef wet salting. Based on the Peleg model, the water and NaCl contents, respectively, presented a coefficient of determination ($R^2$) ranging from 0.967 to 0.999 and from 0.981 to 0.986 and RMSE from 0.0002 to 0.0124 and from 0.0208 to 0.0212, a low error rate RMSE (0.0008 – 0.0048) and low residues, as shown in Figure 1 for the water and NaCl contents.

| Water content | Static Brine (SB) | Ultrasound brine (US) | SEM | $p$ value |
|---------------|------------------|-----------------------|-----|-----------|
|               | C1               | C2                    | C3  | MT CR     |
| $k_1$         | 11.19b           | 30.91a                | 27.03a | 7.55c   |
| $k_2$         | -1.42            | -9.21                 | 14.05 | -1.67    | -1.13    | -15.02 |
| $R^2$         | 0.967            | 0.999                 | 0.986 | 0.999    |
| %VAR          | 0.9809           | 0.9994                | 0.9984 | 0.9994 |
| RMSE          | 0.0124           | 0.0002                | 0.0012 | 0.0003 |

| NaCl content | ($k \times 10^{-4}$) | ($k \times 10^{-3}$) | $R^2$ | %VAR | RMSE  |
|--------------|----------------------|----------------------|-------|------|-------|
|              | 9.32                 | 9.94                 | 9.44a | 9.24a| 1.17a |
|              | 9.66                 | 9.39                 | 9.13a | 8.89 | 8.66a |
| $R^2$        | 0.982               | 0.981                | 0.983 | 0.982|
| %VAR         | 0.8601              | 0.8600               | 0.8599 | 0.8603|
| RMSE         | 0.0211              | 0.0211               | 0.0212 | 0.0208|

Values in the same row not followed by the same letter differ significantly ($p < 0.05$); C1 = ($\frac{1}{2}$ Angus and $\frac{1}{2}$ Nelore); C2 = ($\frac{3}{4}$ Angus and $\frac{1}{4}$ Nelore); C3 = ($\frac{1}{2}$ Wagyu, $\frac{1}{4}$ Nelore and $\frac{1}{4}$ Angus); MT = Method of treatment; CR = Crossbreeding; SEM = Mean standard error for method of treatment (MT) and crossbreeding (CR) at each treatment time; %VAR = Percentage of explained variance; RMSE = root mean square error.

**Figure 1** – Residue between the predicted and experimental values of the Peleg model for the water (A) and NaCl (B) contents in samples treated with static brine (SB) and brine by ultrasound (US).
The constant $k_2$ is related to the equilibrium of water or NaCl contents ($1/k_2$). As a result, lower $k_1$ values indicate higher initial mass transfer rates and lower $k_2$ values indicate a higher equilibrium moisture content. The Peleg model showed higher initial rates ($p \leq 0.05$) of the water content in treatments C1-US and C2-US. Treatment C3-US showed the same ($p \geq 0.05$) initial mass transfer rate as treatments C2-SB and C3-SB. The capacity constant ($k_2$) for the water content showed negative values. The change in signal observed in this study is unexpected and it produces paradoxical results. Piergiovanni (2011) also reported negative values for parameter $k_2$ in their study on water adsorption kinetics in beans, which also reported no physical significance. Schmidt et al. (2009) reported in their study on wet chicken breast salting that the Peleg model tends to overestimate the equilibrium moisture content.

As for the NaCl content, the Peleg model showed higher NaCl absorption rates ($p \leq 0.05$) in the samples of treatments C2-US and C3-US. Treatment C1-US showed the same ($p \geq 0.05$) rates of NaCl absorption as treatments C1-SB, C2-SB and C3-SB. As for the equilibrium content ($1/k_2$) for the NaCl content, the Peleg model showed a higher equilibrium content ($p \leq 0.05$) for all treatments with the use of ultrasound when comparing to the samples treated in static brine.

### Page model

Table 5 presents the results of the Page model adjusted by non-linear regression for the water and NaCl contents during beef wet salting. Based on this model, the water and NaCl contents showed a determination coefficient ($R^2$) ranging from 0.974 to 0.999 and from 0.990 to 0.999 and RMSE from 0.0001 to 0.0100 and from 0.0011 to 0.0092 respectively, and low residues as shown in Figure 2 for the water and NaCl contents.

Regarding the water content, parameter $k$, which represents the process rate, showed higher ($p \leq 0.05$) values for samples C1 - US and C2 - US, associated to ultrasound treatment. Considering only the treatments in static brine, C1-SB obtained the highest ($p \leq 0.05$) value of parameter $k$. The trend of $k$ values is consistent with the results of the water content (Table 2), where samples of treatments C1-US and C2-US showed the highest ($p < 0.05$) water content. Regarding parameter $n$ for water content, no evident relationship

| Water content | Static Brine (SB) | Ultrasound Brine (US) | SEM | p value |
|---------------|------------------|-----------------------|-----|---------|
|               | C1 | C2 | C3 | C1 | C2 | C3 | MT | CR | MT × CR |
| $k$           | 0.063a | 0.025b | 0.014c | 0.111a | 0.090a | 0.022b | 0.044 | < 0.01 | < 0.01 | 0.182 |
| $n$           | 1.320a | 1.949b | 0.644c | 1.598b | 1.535b | 2.028a | 0.021 | 0.024 | < 0.01 | 0.344 |
| $R^2$         | 0.974 | 0.999 | 0.976 | 0.998 | 0.998 | 0.999 |       |       |       |       |
| %VAR          | 0.9890 | 0.9989 | 0.9988 | 0.9956 | 0.9890 | 0.9998 |       |       |       |       |
| RMSE          | 0.0094 | 0.0011 | 0.0007 | 0.0040 | 0.0100 | 0.0001 |       |       |       |       |

| NaCl content  | SEM | p value |
|---------------|-----|---------|
|               | C1  | C2  | C3  | C1  | C2  | C3  | MT | CR | MT × CR |
| $k$           | 0.443a | 0.434b | 0.482c | 0.519b | 0.510c | 0.558a | 0.015 | < 0.01 | 0.321 | 0.282 |
| $n$           | 0.761a | 0.775b | 0.760c | 0.801b | 0.840b | 0.860a | 0.002 | < 0.01 | 0.140 | 0.311 |
| $R^2$         | 0.990 | 0.999 | 0.999 | 0.999 | 0.997 | 0.997 |       |       |       |       |
| %VAR          | 0.9840 | 0.9958 | 0.9980 | 0.9952 | 0.9898 | 0.9901 |       |       |       |       |
| RMSE          | 0.0092 | 0.0024 | 0.0011 | 0.0026 | 0.0058 | 0.0056 |       |       |       |       |

Values in the same row not followed by the same letter differ significantly ($p < 0.05$); C1 = (½ Angus and ½ Nelore); C2 = (¾ Angus and ¼ Nelore); C3 = (¼ Wagyu, ¼ Nelore and ¼ Angus); MT = Method of treatment; CR = Crossbreeding; SEM = Mean standard error for method of treatment (MT) and crossbreeding (CR) at each treatment time; %VAR = Percentage of explained variance; RMSE = root mean square error.

Figure 2 – Residue between the predicted and experimental values of the Page model for the water (A) and NaCl (B) contents in samples treated with static brine (SB) and brine by ultrasound (US).
was found between this parameter and the results of water content. In phenomenological terms, n depends on food microstructure (Simpson et al., 2017).

The increase of the process rates for water in brining with ultrasound is consistent with findings in other studies (Cárcel et al., 2007; Kang et al., 2016). The influence of crossbreeding may be related to differences in the chemical composition of the crossbreed samples, in which raw material with a higher lipid content like C3, which presented lower water absorption rates. The higher C3 lipid content probably hampered the water diffusion process. The use of ultrasound, when associated to raw material with higher protein content, seems to favor the water diffusion process. The effects of ultrasound on beef microstructure has already been reported by several authors (Cárcel et al., 2007a; Kang et al., 2016; Ozuna et al., 2013; Siró et al., 2009), showing increase in gaps between muscle fibers, which may have contributed to increasing muscle protein solubility and hydration.

For the NaCl content, parameter k presented the highest \( p < 0.05 \) values for samples C1-US, C2-US, and C3-US, which did not differ from each other \( p > 0.05 \), but differed from samples in static conditions SB. Treatments C1-SB, C2-SB, and C3-SB did not differ \( p > 0.05 \) from each other. In relation to parameter n for NaCl content, the highest \( p < 0.05 \) values of n were predicted for treatments C1-US, C2-US, and C3-US with ultrasound application, when compared to treatments in static brine SB.

The application of ultrasound may have increased the gap between muscle fibers allowing greater mass transfer of brine in beef. Some authors have proposed several mechanisms to explain these effects, such as implosions of cavitation bubbles near the product surface (Cárcel et al., 2007a; Knorr et al., 2004; Siró et al., 2009), the creation of microchannel, and the sponge effect, which can influence the mass transfer process (Siró et al., 2009).

For the NaCl absorption rate predicted by the Page model, only the treatment method had significant effect, in which all treatments with ultrasound showed the greatest NaCl absorption rate and differed \( p < 0.05 \) from all the treatments in static brine. The differences found in the NaCl content (Table 3) are a significant \( p < 0.05 \) consequence of crossbreeding; however, at chloride levels, they did not cause significant changes \( p > 0.05 \) to the NaCl absorption rate predicted by the model.

**Conclusion**

Crossbreeding between different breeds of cattle (Nelore, Angus, and Wagyu) provided different levels of lipids and proteins in the muscle cuts [biceps femoris]. The Page model was more suitable to describe the mass transfer kinetics. The use of ultrasound and crossbreeding affected the mass transfer rate. For the water content, the Page model showed higher water absorption rates for cuts with higher protein content. Crossbreeding and the use of ultrasound also influenced the NaCl content; however, only the use of ultrasound increased the NaCl absorption rate during brining. The results demonstrate that ultrasound accelerates the mass transfer brining process for cuts beef, regardless of the crossbreeds investigated here. This fact can be used to as an advantage in situations where cuts are processed, resulting from the genetic improvement.

**Acknowledgements**

This study was funded in part by the Higher Education Personnel Improvement Coordination - Brazil [CAPES] - Finance Code 001. The authors would like to thank Beef Passion for supplying the raw material.

**Authors’ Contributions**

- Conceptualization and Data acquisition: Sanches, M.A.R; Silva, P.M.O.C; Barbosa, R.D.; Romero, J.T.; Barretto, A.C.S.
- Data analysis and Writing and editing: Sanches, M.A.R; Silva, P.M.O.C; Barbosa, R.D.; Romero, J.T.; Barretto, A.C.S.

**References**

- Alarcon-Rojo, A.D.; Carrillo-Lopez, L.M.; Reyes-Villagranra, R.; Huerta-Jiménez, M.; García-Galicia, I.A. 2019. Ultrasound and meat quality: a review. Ultrasonics Sonochemistry 55: 369–382.
- Arrigoni, M.D.B.; Alves Júnior, A.; Dias, P.M.A.; Martins, C.L.; Costa Cervieri, R.; Silva, A. C.; Oliveira, H.N.; Chardulo, L.A.L. 2004. Performance, muscle fibers and meat of young cattle from three genetic groups = Desempenho, fibras musculares e carne de bovinos jovens de três grupos genéticos. Pesquisa Agropecuária Brasileira 39: 1033-1039.
- Association of Official Analytical Chemists - International [AOAC]. 2007. Official Methods of Analysis. 18ed. AOAC, Gaithersburg, MD, USA.
- Avilés, C.; Martínez, A.L.; Domenech, V.; Peña, F. 2015. Effect of feeding system and breed on growth performance, and carcass and meat quality traits in two continental beef breeds. Meat Science 107: 94–103.
- Bampi, M.; Domschke, N.N.; Schmidt, F.C.; Laurindo, J.B. 2016. Influence of vacuum application, acid addition and partial replacement of NaCl by KCl on the mass transfer during salting of beef cuts. LWT 74: 26–33.
- Barretto, T.L.; Polachini, T.C.; Barretto, A.C.S.; Telis-Romero, J. 2018a. Water sorption isotherms of cooked hams as affected by temperature and chemical composition. Food Science and Technology 33: 677–683.
- Barretto, T.L.; Pollonio, M.A.R.; Telis-Romero, J.; Barretto, A.C.S. 2018a. Improving sensory acceptance and physicochemical properties by ultrasound application to restructured cooked ham with salt [NaCl] reduction. Meat Science 145: 55–62.
Bligh, E.G.; Dyer, W.J. 1959. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiology 37: 911–917.

Cárcel, J.A.; Benedito, J.; Bon, J.; Mulet, A. 2007a. High intensity ultrasound effects on meat brining. Meat Science 76: 611–619.

Cárcel, J.A.; Benedito, J.; Roselló, C.; Mulet, A. 2007b. Influence of ultrasound intensity on mass transfer in apple immersed in a sucrose solution. Journal of Food Engineering 78: 472–479.

Chandrapala, J.; Oliver, C.; Kentish, S.; Ashokkumar, M. 2012. Ultrasونics in food processing. Ultrasonics Sonochemistry 19: 975–983.

Coelho, M.; Pereira, R.; Rodrigues, A.S.; Teixeira, J.A.; Pintado, M.E. 2019. Extraction of tomato by-products’ bioactive compounds using ohmic technology. Food and Bioproducts Processing 117: 329–339.

Damodaran, S.; Parkin, K.L.; Fennema, O.R. 2010. Fennema’s Food Chemistry = Química de Alimentos de Fennema. 4ed. Artmed, Porto Alegre, RS, Brazil.

Dias, L.S.; Hadlich, J.C.; Luzia, D.M.M.; Jorge, N. 2016. Influence of breed on beef and intramuscular fat quality from nellore (Bos indicus) and wagyu (Bos taurus) crossbreed cattle. International Food Research Journal 23: 1523–1530.

Duarte, M.S.; Paulino, P.V.R.; Das, A.K.; Wei, S.; Serao, N.V.L.; Fu, X.; Harris, S.M.; Dodson, M. 2013. Enhancement of adipogenesis and fibrogenesis in skeletal muscle of Wagyu compared with Angus cattle. Journal of Animal Science 91: 2938–2946.

Frank, D.; Ball, A.; Hughes, J.; Krishnamurthy, R.; Piyasiri, U.; Stark, J.; Watkins, P.; Warner, R. 2016. Sensory and flavor characteristics of ausralian beef: influence of intramuscular fat, feed, and breed. Journal of Agricultural and Food Chemistry 64: 4299–4311.

Goulia, A.M.; Kokolaki, M.; Dafsiou, E. 2017. Use of ultrasound for osmotic dehydration: the case of potatoes. Food and Bioproducts Processing 105: 157–170.

Hwang, Y.H.; Kim, G.D.; Jeong, 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: 456–461.

Joo, S.T.; Kim, G.D.; Hwang, Y.H.; Ryu, Y.C. 2013. Control of fresh meat quality through manipulation of muscle fiber characteristics. Meat Science 95: 828–836.

Kang, D.; Wang, A.; Zhou, G.; Zhang, W.; Xu, S.; Guo, G. 2016. Power ultrasonic on mass transport of beef: effects of ultrasound intensity and NaCl concentration. Innovative Food Science and Emerging Technologies 35: 36–44.

Kim, G.D.; Kim, B.W.; Jeong, J.Y.; Hur, S.J.; Cho, I.C.; Lim, H.T.; Joo, S.T. 2013. Relationship of carcass weight to muscle fiber characteristics and pork quality of crossbred [Korean native black pig × Landrace] F2 Pigs. Food and Bioprocess Technology 6: 522–529.

Knorr, D.; Zenker, M.; Heinz, V.; Lee, D.U. 2004. Applications and potential of ultrasomics in food processing. Trends in Food Science and Technology 15: 261–266.

Lage, J.F.; Paulino, P.V.R.; Valadares Filho, S.C.; Souza, E.J.O.; Duarte, M.S.; Benedeti, P.D.B.; Souza, N.K.P.; Cox, R.B. 2012. Influence of genetic type and level of concentrate in the finishing diet on carcass and meat quality traits in beef heifers. Meat Science 90: 770–774.

Lawrie, R. A. 2005. Meat Science = Ciência da Carne. 6ed. Artmed, Porto Alegre, RS, Brazil.

Leonelli, C.; Mason, T.J. 2010. Microwave and ultrasonic processing: now a realistic option for industry. Chemical Engineering and Processing: Process Intensification 49: 885–900.

Maggioni, D.; Marques, J.A.; Rotta, P.P.; Perotto, D.; Ducatti, T.; Visentainer, J.V.; Prado, I. N. 2010. Animal performance and meat quality of crossbred young bulls. Livestock Science 127: 176–182.

Martins, M.G.; Chada, P.S.N.; Silva, R.P. 2019. Application of pulsed-vacuum on the salt impregnation process of pirarucu fillet. Food Research International 120: 407–414.

Motoyama, M.; Sasaki, K.; Watanabe, A. 2016. Wagyu and the factors contributing to its beef quality: a Japanese industry overview. Meat Science 120: 10–18.

Ozawa, S.; Mitsuhashi, T.; Mitsumoto, M.; Matsumoto, S.; Itoh, N.; Itagaki, K.; Kohno, Y.; Dooho, T. 2000. The characteristics of muscle fiber types of longissimus thoracis muscle and their influences on the quantity and quality of meat from Japanese Black steers. Meat Science 54: 65–70.

Ozuna, C.; Puig, A.; García-Pérez, J.V.; Mulet, A.; Cárcel, J.A. 2013. Influence of high intensity ultrasound application on mass transport, microstructure and textural properties of pork meat [Longissimus dorsi] brined at different NaCl concentrations. Journal of Food Engineering 119: 84–93.

Page, G.E. 1949. Factors influencing the maximum rates of air-drying shelled corn in thin layer. MSc. Thesis. Department of Mechanical Engineering, Purdue University, West Lafayette, IN, USA.

Peleg, M. 1988. An empirical model for the description of moisture sorption curves. Journal of Food Science 53: 1216–1217.

Piergiovanni, A.R. 2011. Kinetic of water adsorption in common bean: considerations on the suitability of Peleg’s model for describing bean hydration. Journal of Food Processing and Preservation 35: 447–452.

Polachini, T.C.; Mulet, A.; Telis-Romero, J.; Cárcel, J.A. 2019. Influence of high intensity ultrasound application on the kinetics of sugar release from acid suspensions of artichoke [Cynara scolymus] biomass. Chemical Engineering and Processing - Process Intensification 145: 107681.

Rossato, L.V.; Bressan, M.C.; Rodrigues, É.C.; Gama, L.T.; Bessa, R.J.B.; Alves, S.P.A. 2010. Physicochemical parameters and fatty acid profiles in Angus and Nellore cattle finished on pasture. Revista Brasileira de Zootecnia 39: 1127–1134 (in Portuguese, with abstract in English).

Ryu, Y.C.; Choi, Y.M.; Lee, S.H.; Shin, H.G.; Choe, J.H.; Kim, J.M.; Hong, K.C.; Kim, B.C. 2008. Comparing the histochemical characteristics and meat quality traits of different pig breeds. Meat Science 80: 363–369.

Sanjuán, N.; Simal, S.; Bon, J.; Mulet, A. 1999. Modelling of broccoli stems rehydration process. Journal of Food Engineering 42: 27–31.

Schmidt, F.C.; Carciofi, B.A.M.; Laurindo, J.B. 2009. Application of diffusive and empirical models to hydration, dehydration and salt gain during osmotic treatment of chicken breast cuts. Journal of Food Engineering 91: 553–559.
Sharma, M.; Dash, K.K. 2019. Effect of ultrasonic vacuum pretreatment on mass transfer kinetics during osmotic dehydration of black jamun fruit. Ultrasonics Sonochemistry 58: 104693.
Shinwari, K.J.; Rao, P.S. 2018. Thermal-assisted high hydrostatic pressure extraction of nutraceuticals from saffron [Crocus sativus]: process optimization and cytotoxicity evaluation against cancer cells. Innovative Food Science and Emerging Technologies 48: 296–303.
Simpson, R.; Ramírez, C.; Nuñez, H.; Jaques, A.; Almonacid, S. 2017. Understanding the success of Page’s model and related empirical equations in fitting experimental data of diffusion phenomena in food matrices. Trends in Food Science and Technology 62: 194-201.
Siró, I.; Vén, C.; Balla, C.; Jónás, G.; Zeke, I.; Friedrich, L. 2009. Application of an ultrasonic assisted curing technique for improving the diffusion of sodium chloride in porcine meat. Journal of Food Engineering 91: 353–362.
Vidal, V.A.S.; Biachi, J.P.; Paglarini, C.S.; Pinton, M.B.; Campagnol, P.C.B.; Esmerino, E.A.; Cruz, A.G.; Morgano, M.A.; Pollonio, M.A.R. 2019. Reducing 50 % sodium chloride in healthier jerked beef: an efficient design to ensure suitable stability, technological and sensory properties. Meat Science 152: 49–57.
Xue, S.; Wu, C.; Wu, Y.; Zhang, C. 2018. An optimized process for treating sodium acetate waste residue: coupling of diffusion dialysis or electrodialysis with bipolar membrane electrodialysis. Chemical Engineering Research and Design 129: 237–247.
Yamada, T.; Higuchi, M.; Nakanishi, N. 2014. Fat depot-specific differences in pref-1 gene expression and adipocyte cellularity between Wagyu and Holstein cattle. Biochemical and Biophysical Research Communications 445: 310–313.