



Effects of NaHCO₃ on the colour, tenderness, and water distribution of raw and cooked marinated beef

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Abstract

Effects of NaHCO₃ addition (0.2-0.8%) on the colour, shear force, and water distribution of raw and cooked marinated beef were investigated. The pH and cooking yield were found to be significantly increased ($P < 0.05$), and the shear force, L* and b* values were significantly decreased with increasing NaHCO₃ amounts, except for samples with 0.6% and 0.8% NaHCO₃, for which the cooking yield and shear force remained unaffected. Furthermore, the initial relaxation times T₂₁ and T₂₂ of marinated beef were, and the mobility of water in the raw marinated beef was reduced ($P < 0.05$) with increasing NaHCO₃ content. This was due to the carbon dioxide production disrupting the beef structure during heating process. The mobility of water in the cooked marinated beef was found to be improved. In conclusion, NaHCO₃ addition improved the water holding capacity and tenderness of marinated beef.

Keywords: NaHCO₃; shear force; tenderness; colour; water mobility.

Practical Application: This study provides a method of using NaHCO₃ to improve the colour, tenderness, and water holding capacity of marinated beef, and should be expanded its use in meat products processing.

1 Introduction

Tenderness is an important quality measure for beef. Improving the tenderness can thus significantly improve beef quality and palatability. Tenderness is also regarded as one of the major factors affecting customer perception when purchasing meat products. A favourable profile of tenderness and juiciness is also needed for customer acceptance of marinated beef (Madhusankha & Thilakarathna, 2021). Phosphate is often used as water-retaining agent and tenderizer in processing of marinated beef. Phosphate improves the pH and ionic strength, chelates metal ions, and facilitates the dissociation of actomyosin. Phosphate is also cheap, effective, and easy-to-handle (Pinton et al., 2020; Leng et al., 2021). However, phosphate addition to meat products may also disrupt the optimal proportion of calcium and phosphorus in human body (Isiuku & Enyoh, 2020; Saito et al., 2021). Several studies found that NaHCO₃ might be used instead of phosphate to improve the water holding capacity and tenderness of meat and seafood products (Wachirasiri et al., 2016; Xiong et al., 2020; Kang et al., 2021).

NaHCO₃ is a commonly used and low-cost food material with a good buffering capacity due to the formation of bicarbonate ions (HCO₃⁻) between pH 6.4 and 10.3. NaHCO₃ is weakly alkaline in aqueous solution, and heat labile: it begins to decompose into carbon dioxide and sodium carbonate at 20 °C, and decomposes completely 100 °C (Skurray et al., 1986; Kang et al., 2021). Increasing the NaHCO₃ content of meat can facilitate the dissolving of actomyosin and myofibrillar protein, trigger the denaturation of the head domain of myosin, and promote the dissociation

of actomyosin from myofibrillar protein (Chantarasuwan et al., 2011; Mohan et al., 2016; Li et al., 2021). NaHCO₃ is also widely used in Chinese cooking technology to improve the tenderness and water retention of livestock and poultry meat and aquatic products, in addition to masking unpleasant odours (Hsieh et al., 1980; Åsli & Mørkøre, 2012). Several previous studies reported that NaHCO₃ can improve the palatability and processability of PSE (Pale, Soft, Exudative) meat, sow loins and PSE-like chicken breast by increasing the pH, enhancing electrostatic repulsion, increasing water retention, yield and tenderness of muscle tissue (Kauffman et al., 1998; Woelfel & Sams, 2001; Sindelar et al., 2003). NaHCO₃ has also been used in chicken, pork and beef meat batters to improve their techno-functional properties and gel structures (Mohan et al., 2016; Lu et al., 2021; Kang et al., 2021). However, as far as we know, the effect of NaHCO₃ addition on raw and cooked marinated beef has not been investigated thus far. Hence, we investigated here the effects of NaHCO₃ on the colour, water holding capacity, tenderness, and water distribution/mobility of raw and cooked marinated beef, and the relationship between tenderness and water mobility.

2 Materials and methods

2.1 Raw materials and ingredients

The 72 h postmortem beef (*ectogluteus*, 2 ± 2 °C, pH 5.68 ± 0.01) derived from Simmental bull (18 month old) was purchased four times in 4 d by Henan Yisai Beef Co., Ltd.

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NaCl and NaHCO₃ (analytically pure) were purchased from JSC Chemical Technology Co., Ltd., China. The white pepper powder was purchased from a local market (Xinxiang, China).

2.2 Preparation of marinated beef

The visible fat and connective tissues of beef were removed, and cut into sections sized 4 cm × 4 cm × 2 cm. 20 samples were prepared by each treatment. Marinated beef samples were prepared using 1000 g beef, 15 g NaCl, 350 g ice water, 8.5 g white pepper powder. In addition, T1 added 0 g (0%) NaHCO₃, T2 added 2 g (0.2%) NaHCO₃, T3 added 4 g (0.4%) NaHCO₃, T4 added 6 g (0.6%) NaHCO₃, and T5 added 8 g (0.8%) NaHCO₃. The beef was marinated using the intermittent tumbling method at 2 ± 2 °C using the following procedure: 8 rpm, rolling was 20 min, stop was 10 min, a total of 60 min; vacuum degree was -0.08 MPa. After that, the beef was vacuum packaged (500 g/bag) and stored at 2 ± 2 °C for 12-16h.

2.3 pH determination

About 10 g of raw marinated beef and 40 mL of distilled water (2 ± 2 °C) were mixed uniformly by a T25 digital homogenizer (IKA Ltd., Germany; speed 15000 rpm, time 10 s) in an ice bath. The pH level of raw marinated beef was then determined using a digital pH meter (Hanna, Italy) at 4 °C. The method of the pH meter calibrated and was temperature compensation applied as follows: (1) The electrode was washed and dried using double distilled water. After that, immersed in pH 7.00 standard solution, and the instrument temperature compensation knob was placed at the solution temperature. When the indicating value was stable, adjusted the positioning knob to make the indicating value of the instrument be the pH of the standard solution. (2) The electrode was washed and dried using double distilled water. Following, immersed in pH 4.00 standard solution. After the indicating value was stable, adjusted the slope knob of the instrument, and the indicating value was the pH of the standard solution. (3) The electrode was washed and dried using double distilled water, and then immersed in pH 7.00 standard solution. If the error exceeded 0.02 units, repeated steps (1) and (2), until the correct pH was displayed in both standard solutions without the need for adjustment knobs. (4) The electrode was washed and dried using double distilled water. Following, adjusted the temperature compensation knob to the temperature of the sample solution, immersed the electrode in the sample solution, placed it still after shaking, and displayed the reading after stability.

2.4 Cooking yield

After stored overnight at 2 ± 2 °C, 12 samples of raw marinated beef were cooked using a water bath kettle (HH-42, Changzhou Guohua Electric Appliance Co., Ltd., China) at 85 ± 2 °C for 25 min until the core temperature was 72 °C. Following, the cooked beef was cooled using running water. After that, the lost water and fat from the marinated beef was wiped away. Then, the cooking yield was calculated according to the formula:

Cooking yield % = Weight of cooked marinated beef/Weight of raw marinated beef × 100%.

2.5 Colour measurement

A CR-400 Colorimeter (Minolta Camera Co, Japan) with a pulse xenon lamp (the aperture is a diameter of 11 mm) was used to determine the center colour of raw marinated beef at the 10° observer angle, and it was calibrated with a standard white colorimeter using the illuminant D65. The fresh slice from each raw marinated beef was measured at most 1 min.

2.6 Shear force measurement

The cooked marinated beef was left the laboratory at room temperature for 2 h after 2 ± 2 °C stored overnight. Then, the cooked marinated beef was cut into 4 cm × 1 cm × 1 cm slices along the direction of the muscle fibers. The shear force (N) was measured using a muscle tenderness meter (C-LM4, College of Engineering, Northeast Agricultural University, China).

2.7 Low-field nuclear magnetic resonance (LF-NMR) measurements

The raw and cooked marinated beef were cut into about 2 g cubes and placed in Ziplock bags, and the marinated beef were left at a 32 °C thermostat (P series, Guangdong Hongzhan Technology Co., Ltd., China) for 30 min. LF-NMR relaxation measurements were performed by using a Niumag NMR analyzer (NMI20-040 V-1, China). During the process of measurement, a resonance frequency of 22.6 MHz at 32 °C was operated, the value of τ (the time between 90° and 180° pulse) was 200 μ s, the number of collected echoes was 5000, repeat the scan 32 times with a repetition interval of 6.5 s to get 12000 echoes. The data were analyzed using the equipped software.

2.8 Statistical analysis

The experiment was repeated four times at different occasions, using different samples (raw and cooked marinated beef). Data were analyzed by the linear mixed models (LMM) procedure (SPSS v.18.0), considering the treatments (marinated beef with 0%, 0.2%, 0.4%, 0.6%, and 0.8% NaHCO₃) as a fixed effect and the replicates as a random effect. The pH, cooking yield, colour, and shear force data were analysed in a similar fashion. LF-NMR data of raw and cooked marinated beef were included in the random term for the evaluation of the initial relaxation time and peak ratio, and the NMR data time as a fixed term along with the interaction with treatment. Approximate F-ratio tests for each fixed effect were conducted and critical value for a statistically important effect taken at $P < 0.05$. Non-significant terms were excluded from models using stepwise backward elimination.

3 Results and discussion

3.1 pH

The pH level is a major indicator of beef quality, as it directly affects palatability, tenderness, water retention, and shelf life. Figure 1 shows the effect of NaHCO₃ addition on pH of marinated beef. As expected, the pH of raw marinated beef was significantly increased ($P < 0.05$) from 5.68 to 6.57 with increasing NaHCO₃. This is because NaHCO₃ is a strong alkali

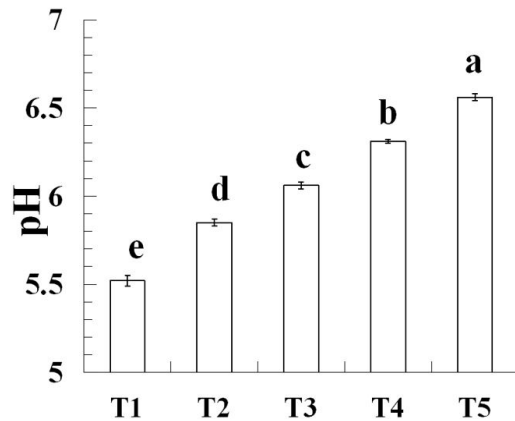


Figure 1. pH of raw marinated beef with various amounts of added NaHCO_3 . Each value represents the mean \pm SE, $n = 4$. ^{a-e} Different parameter superscripts indicate significant differences ($P < 0.05$).

weak acid salt, which produces hydroxide ions (OH^-) and carbonic acid (H_2CO_3) in aqueous solutions (Li et al., 2021). Thus, when used in marinated beef, NaHCO_3 increases the pH level. Similar findings were previously reported by Zhu et al. (2018), where the authors used NaHCO_3 to replace sodium chloride, resulting in significant increases in the pH of chicken breast meat batter. Petracci et al. (2014) also found that the pH of marinated chicken breast meat was increased with increasing the levels of NaHCO_3 .

3.2 Cooking yield

Figure 2 shows the effect of NaHCO_3 addition on cooking yield of marinated beef. The cooking yield of marinated beef with NaHCO_3 was significantly increased compared to that of T1. The pH of raw marinated beef was found to be very different ($P < 0.05$) from the isoelectric point of myofibrillar proteins upon NaHCO_3 addition. This could increase the net negative charge of proteins, and lead to swelling and dissolving of beef muscle fibers, prompting more water to be immobilized during marination (Offer & Knight, 1988). Kang et al. (2021) previously found that partial replacement of NaCl by NaHCO_3 significantly increased the salt-soluble protein concentration of raw pork batters. Mohan et al. (2016) also reported that ground beef with NaHCO_3 or KHCO_3 improved the water holding capacity during the heating process. As for the other samples, the cooking yield was significantly increased ($P < 0.05$) with increasing NaHCO_3 , except for T5. NaHCO_3 is known to cause generation of carbon dioxide (CO_2) during the heating process. If excessive amounts of NaHCO_3 are added, large amounts of produced carbon dioxide may disrupt the beef structure (Petracci et al., 2013; Kang et al., 2021). This may explain why similar cooking yields for T4 and T5 were obtained.

3.3 Colour

The changes in colour of raw marinated beef with various amounts of added NaHCO_3 are shown in Table 1. The L^* and b^*

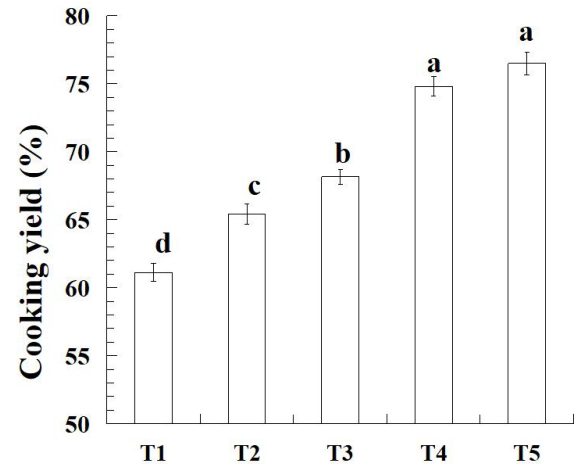


Figure 2. Cooking yield of marinated beef with various amounts of added NaHCO_3 . Each value represents the mean \pm SE, $n = 4$. ^{a-d} Different parameter superscripts indicate significant differences ($P < 0.05$).

Table 1. Color (L^* , a^* and b^* values) of raw marinated beef with various amounts of added NaHCO_3 .

Sample	L^*	a^*	b^*
T1	47.15 \pm 0.72 ^a	9.58 \pm 0.32 ^a	12.06 \pm 0.35 ^a
T2	46.16 \pm 0.41 ^b	9.36 \pm 0.16 ^a	10.73 \pm 0.31 ^b
T3	44.78 \pm 0.38 ^c	9.29 \pm 0.22 ^a	9.67 \pm 0.32 ^{bc}
T4	43.59 \pm 0.42 ^{cd}	9.21 \pm 0.19 ^a	9.41 \pm 0.22 ^c
T5	42.81 \pm 0.37 ^d	9.01 \pm 0.21 ^a	9.03 \pm 0.16 ^c

Each value represents the mean \pm SE, $n = 4$. ^{a-d} Different parameter superscripts indicate significant differences ($P < 0.05$).

values of raw marinated beef with NaHCO_3 were significantly decreased ($P < 0.05$) compared to that of T1. Specifically, the levels of these metrics significantly decreased with increasing NaHCO_3 addition from 0.2% to 0.4%, whereas the addition of NaHCO_3 at levels between 0.4% to 0.8% did not lead to significant differences. On the other hand, the a^* value showed an overall decreasing trend with NaHCO_3 addition, yet without any significant differences between the samples.

Several studies have reported effects of NaHCO_3 on colour of meat and sea food. Kaewthong et al. (2021) studied the effect of plant juices and NaHCO_3 on the quality of marinated goat meat, and showed that L^* , a^* and b^* values were decreased with increasing concentrations of NaHCO_3 , indicating that the marinating solution containing NaHCO_3 caused the marinated goat meat to obtain a dark-brown colour. Petracci et al. (2014) found that the L^* , a^* and b^* values of raw chicken breast meat following marination were not significantly affected by increasing levels of NaHCO_3 . Mohan et al. (2016) found that the L^* value of ground meat products were not significantly affected by NaHCO_3 addition due to added modified food or potato starch content of these products. However, the L^* values of salted Atlantic cod were decreased upon NaHCO_3 addition (Åsli & Mørkøre, 2012). The differences were due to the different materials and

processing method. Here, the beef samples included more myoglobin than goat meat, chicken meat, and Atlantic cod, and thus the stability of myoglobin was higher when the pH was increased (Lu et al., 2021). Hence, the addition of NaHCO₃ led to a darker coloured beef product.

3.4 Shear force

Shear force is an important measure of beef tenderness. The smaller the shear force, the better the tenderness. The change in shear force of cooked marinated beef with different NaHCO₃ is shown in Figure 3. Accordingly, the shear force was significantly decreased ($P < 0.05$) with increasing NaHCO₃ amounts, except for T5. T4 and T5 yielded the highest shear force at similar levels. This is because NaHCO₃ can lead to the formation of disordered collagen tissue between muscle cells, and decrease in toughness of beef (Zou et al., 2019). In addition, NaHCO₃ added samples yielded high pH and ionic strength levels, which caused increased solubilisation of salt-soluble proteins, and decreased the hardness of muscle fibers (Saleem et al., 2015). Hence, myofibril fragmentation index was effectively increased upon NaHCO₃ addition, and reduced the shear force (Xiong et al., 2020; Zou et al., 2019). A similar study was shown by Xiong et al. (2020), who found that used the NaHCO₃ assisted curing of chicken breast meat has a lower shear force and larger myofibril fragmentation index (8.01 N and 56.82) than that of traditional wetting curing (6.99 N and 61.65). Lee et al. (2015) previously studied the effect of KHCO₃, NaHCO₃, potassium lactate, and salt injections on functional properties of whole chicken breast, and found that the shear force values were lower at 0.5% level for both KHCO₃ and NaHCO₃ compared with those of potassium lactate and salt.

3.5 LF-NMR measurements

Spin-spin relaxation time (T_2) measured by LF-NMR can reflect the distribution and mobility of water in marinated meat (Åsli et al., 2016). The T_2 of raw and cooked marinated beef meat with different NaHCO₃ are shown in Figure 4, Table 2 and 3. The peaks of T_{2b} , T_{21} , and T_{22} correspond to 0–10, 10–100, and 100–1000 ms, and represent bound water, immobile water and free water in the samples, respectively (Petracci et al., 2014; Kang et al., 2016).

LF-NMR measurements of raw marinated beef

Figure 4 and Table 2 show the changes in initial relaxation time and the peak ratio of raw marinated beef with various amounts of added NaHCO₃. The initial relaxation time of T_{2b} , T_{21} , and T_{22} , and the peak ratio of P_{22} were significantly decreased, whereas the peak ratio of P_{21} was significantly increased ($P < 0.05$) upon NaHCO₃ addition. On the other hand, the peak ratio of P_{2b} was not significantly affected. These results indicate that the water was tightly connected to the meat matrix, and the water mobility was reduced when NaHCO₃ was added (Kang et al., 2021). Several studies have previously reported that the addition of NaHCO₃ could shorten the relaxation time of cured pork and chicken meat compared to marination with NaCl (Bertram et al., 2008; Petracci et al., 2014; Xiong et al., 2020). With increasing

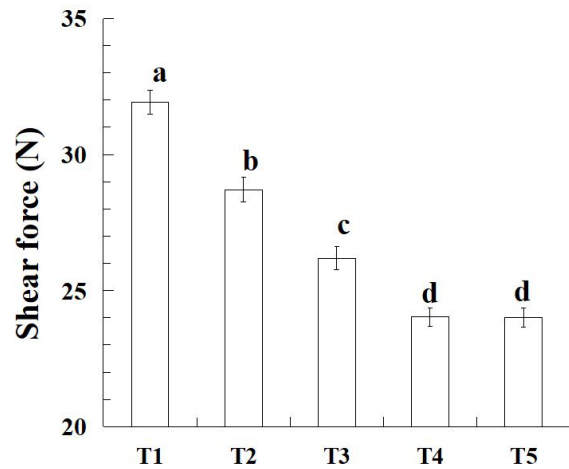


Figure 3. Shear force of cooked marinated beef with various amounts of added NaHCO₃. Each value represents the mean \pm SE, $n = 4$. ^{a-d} Different parameter superscripts indicate significant differences ($P < 0.05$).

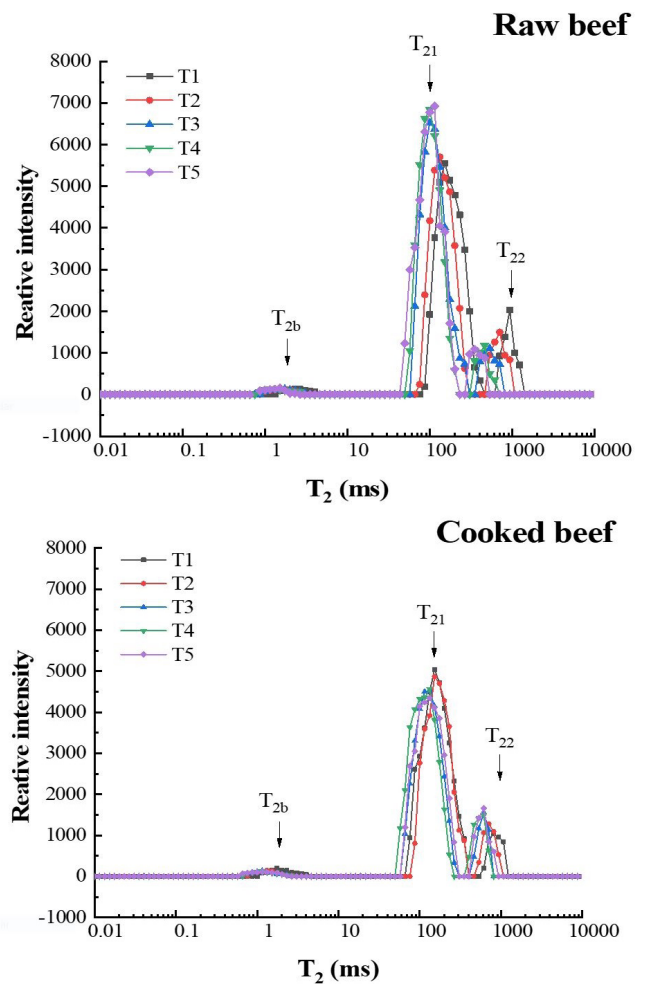


Figure 4. The changes in LF-NMR relaxation times and peak ratio of raw and cooked marinated beef with various amounts of added NaHCO₃.

Table 2. The initial relaxation time (ms) and the peak ratio (%) of raw marinated beef with various amounts of added NaHCO₃.

Sample	Initial relaxation time (ms)			Peak ration (%)		
	T _{2b}	T ₂₁	T ₂₂	P _{2b}	P ₂₁	P ₂₂
T1	1.41 ± 0.07 ^a	85.52 ± 1.82 ^a	679.38 ± 15.14 ^a	2.04 ± 0.06 ^a	86.18 ± 0.44 ^c	11.87 ± 0.18 ^a
T2	1.02 ± 0.06 ^b	78.91 ± 2.02 ^b	551.78 ± 16.86 ^b	2.07 ± 0.05 ^a	89.26 ± 0.48 ^b	9.05 ± 0.16 ^b
T3	0.96 ± 0.06 ^b	70.23 ± 1.76 ^c	412.81 ± 14.72 ^c	1.96 ± 0.06 ^a	92.65 ± 0.51 ^a	5.58 ± 0.18 ^c
T4	0.87 ± 0.09 ^b	61.74 ± 1.56 ^d	348.17 ± 15.79 ^d	1.94 ± 0.07 ^a	92.25 ± 0.47 ^a	5.23 ± 0.15 ^c
T5	0.85 ± 0.095 ^b	53.49 ± 1.55 ^e	280.17 ± 15.14 ^e	1.97 ± 0.05 ^a	93.57 ± 0.45 ^a	4.72 ± 0.17 ^c

Each value represents the mean ± SE, n = 4. ^{a-c}Different parameter superscripts indicate significant differences ($P < 0.05$).

Table 3. The initial relaxation time (ms) and the peak ratio (%) of cooked marinated beef with various amounts of added NaHCO₃.

Sample	Initial relaxation time (ms)			Peak ration (%)		
	T _{2b}	T ₂₁	T ₂₂	P _{2b}	P ₂₁	P ₂₂
T1	1.18 ± 0.04 ^a	77.53 ± 1.65 ^a	607.58 ± 14.36 ^a	3.27 ± 0.08 ^a	91.53 ± 0.48 ^a	6.26 ± 0.13 ^c
T2	0.85 ± 0.05 ^b	71.09 ± 1.64 ^b	512.87 ± 13.17 ^b	2.80 ± 0.06 ^b	88.07 ± 0.45 ^b	9.37 ± 0.12 ^d
T3	0.81 ± 0.05 ^b	62.87 ± 1.64 ^c	433.69 ± 13.74 ^c	2.58 ± 0.07 ^c	86.15 ± 0.49 ^c	11.26 ± 0.14 ^c
T4	0.76 ± 0.04 ^b	59.07 ± 1.54 ^c	427.05 ± 12.84 ^c	2.21 ± 0.06 ^d	85.42 ± 0.51 ^c	12.84 ± 0.11 ^b
T5	0.70 ± 0.05 ^b	63.49 ± 1.71 ^c	446.74 ± 14.26 ^c	1.76 ± 0.07 ^e	83.90 ± 0.48 ^{cd}	14.27 ± 0.13 ^a

Each value represents the mean ± SE, n = 4. ^{a-e}Different parameter superscripts indicate significant differences ($P < 0.05$).

NaHCO₃, the initial relaxation time of T_{2b} was not significant different ($P > 0.05$), T₂₁ and T₂₂ were significantly decreased ($P < 0.05$); the peak ratio of P₂₁ and P₂₂ were significantly increased, but T3, T4 and T5 were not significant different ($P > 0.05$). The results indicated that the immobile water and free water were tied closely in the beef meat, and water mobility was reduced with increasing NaHCO₃. The reason is possible that the pH, ionic strength and electrostatic repulsion between the proteins were increased with increasing NaHCO₃, leading to the beef myofibrillar protein shifted away from the isoelectric point and promoted it dissolution, thereby increased spacing between filaments (Alvarado & Sams, 2003; Li et al., 2021). On the other hand, more collagen tissue was destroyed with increasing NaHCO₃, and more water can be held in the raw marinated beef (Sultana et al., 2009).

NMR measurements of cooked marinated beef

Figure 4 and Table 3 show the changes in initial relaxation time and the peak ratio of cooked marinated beef with various amounts of added NaHCO₃. The initial relaxation time of T_{2b}, T₂₁, and T₂₂, and the peak ratio of P_{2b} and P₂₁ were significantly decreased ($P < 0.05$), whereas the peak ratio of P₂₂ was significantly increased upon NaHCO₃ addition. This also indicates that water was tightly connected to the meat matrix, and the water mobility was increased when NaHCO₃ was added (Bertram et al., 2008). This result contrasts with that obtained for raw marinated beef (Table 2), yet consistent with cooking yield results of raw beef (Figure 2). Since the marinated beef with NaHCO₃ returned a higher cooking yield than that of T1, the peak ratio of P_{2b} of marinated beef with NaHCO₃ was also lower. Carbon dioxide production due to the addition of NaHCO₃ may cause formation of air bubbles in the marinated beef during heating process, and result in disruption of the structure of cooked marinated beef (Zhu et al., 2018). For this reason, the initial relaxation

times of T₂₁ and T₂₂, and the peak ratio of P₂₁ were significantly decreased ($P < 0.05$) upon NaHCO₃ addition as well, yet T3, T4, and T5 were not significantly affected. The peak ratio of P₂₂ was also significantly increased, implying the water mobility was increased when excessive NaHCO₃ was added.

4 Conclusion

In the study, we showed that NaHCO₃ addition significantly affected the colour, tenderness, and water holding capacity of marinated beef. The addition of NaHCO₃ significantly increased the pH and cooking yield, and significantly decreased the shear force, L* and b* values, and the initial relaxation time of T_{2b}, T₂₁ and T₂₂ of marinated beef. NaHCO₃ also caused significant increases in the pH and cooking yield, whereas shear force and initial relaxation times of T_{2b}, T₂₁, and T₂₂ were significantly decreased, except for the cooking yield and shear force of sample with 0.8% NaHCO₃ content. The water mobility of raw marinated beef was reduced with increasing NaHCO₃ content, whereas that of cooked marinated beef was increased. Overall, NaHCO₃ addition could lead the water to form a tight connection to the beef matrix, and thereby improve the tenderness and cooking yield of marinated beef.

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