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ORIGINAL ARTICLE

Effect of sodium bicarbonate on gel properties and protein conformation of phosphorus-free chicken meat batters

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Texture

Abstract So as to investigate the effect of using sodium bicarbonate (SB) to replace sodium tripolyphosphate (ST) in the chicken meat batters with ST or SB (0.30% and 0.50%), the changes of gel properties and protein conformation were studied. The pH, salt-soluble protein solubility, cooking yield, b* value and texture properties were increased significantly ($p < 0.05$), the L* and a* values were decreased significantly ($p < 0.05$) when used the SB to replace ST. The β-sheet structure content was increased, accompanied by the random coil content was decreased ($p < 0.05$) when used the SB to replace ST. Meanwhile, more hydrophobic interactions were formed and more aliphatic residues were exposed to hydrophilic environment. The secondary and tertiary structures had little affect with the ST or SB were increased from 0.30% to 0.50%. Overall, it was obtained that the use of SB could produce the phosphorus-free chicken meat batters.

1. Introduction

In recent years, consumers pay more and more attention to healthy meat products. For their convenience and nutritional values, emulsified meat products are popular all over the word. But most of them added phosphates as phosphates are cheap, effective and easily handled, and supplied some advantages such as increased cooking yield, enhanced texture properties and prolonged the shelf-life, through increased pH, ionic strength, chelate metal ions, salt-soluble protein concentration and induced more protein structure changes (Sarjit and Dykes, 2015; Thangavelu et al., 2019; Gabriel et al., 2020). Some researchers found that phosphorus additives could cause hyperphosphatemia as they have highly bioavailable (almost 100%). It is well known that excessive dietary phosphorus intake can influence the optimal proportion of calcium and phosphorus in human body, which causes bone, cardiovas and cularchronic kidney disease (Yamada et al., 2018; Calvo

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et al., 2019). However, total or partial removed phosphates were lowered the water holding capacity, texture characteristics and shelf-life. Therefore, it is a challenge to remove them or reduce their amount without any negative effect in the emulsified meat products.

Sodium bicarbonate (SB) is a cheap, convenience and easy soluble in water, which has been widely used in meat products and sea foods to improve water holding capacity, juiciness, texture properties and flavor, as it has stronger buffering capacity and ionic strength (Petracci et al., 2013; Xiong et al., 2020). Zou et al. (2019) showed that the treatment of ultrasound combined with 0.2 M SB promoted the actomyosin of chicken breast meat degeneration, increased in α-helix content and decreased in fluorescence intensity of tyrosine and tryptophan, and increased the MFI, cooking yield and tenderness of cured chicken breast. Xiong et al. (2020) found that the use of SB or ultrasound combined with SB assisted curing could decrease significantly the cooking loss, shear force and surface hydrophobicity of chicken breast meat. Zhu et al. (2018) reported that the chicken meat batter with 0.5% SB had higher pH, cooking yield, textural properties, and β-sheet structure than the sodium chloride. Petracci et al. (2013) stated that as a superior marinating agent, the greater marination performances of SB was used the concentration less than 0.3% in chicken breast meat. For all we know, there is little information about the different changes of gel properties and protein conformation on the application of phosphates and SB in emulsified meat products. Due to sodium tripolyphosphate (ST) is the most popular form of phosphates used in meat industry (Thangavelu et al., 2019), therefore, the aim of this paper was to study the influences on gel properties and protein conformation of chicken batters was made with various amount of ST or SB alone, analyze the possibility of application in chicken meat batters.

2. Material and method

2.1. Materials

The Arbor Acres chickens breast meat was obtained after slaughter 10 min, and was chilled at 2 ± 2 °C for 12 h (pH, 5.90 ± 0.01) in the airtight plastic bags (PE). Then, the meat was ground by the MM-12 grinder (6 mm diameter holes plate, China). About 1.0 kg ground meat was vacuum packaged in nylon/PE bags and frozen storage (−20 °C) less than 14 d. SB and ST were the analytically pure.

2.2. Raw batter preparation

The raw batter was processed according to the method of Zhu et al. (2018). The formulas of raw batters were as follows: chicken breast meat 500 g, water 100 g, ST 1.8 g (T1) or 3 g (T2) alone, or SB 1.8 g (T3) or 3 g (T4) alone.

2.3. pH determined

10 g of raw chicken batter was homogenized at 15,000 rpm for 10 s with 40 mL of distilled water (4 °C) by a homogenizer (T25 digital polytron, IKA Ltd, Germany). After that, the pH was measured immediately (Hanna, Italy).

2.4. Salt-soluble protein (SSP)

The method of SSP of raw chicken batters was according to the procedure of Cofrades et al. (2008).

2.5. Cooking yield

After stored at 2 ± 2 °C overnight, the exudate separated of cooked batter was cleaned and weighed. Applied the following formula to calculate the cooking yield:

$$\text{Cooking yield\%} = \text{Weight of cooked batter/Weight of raw batter} \times 100\%.$$

2.6. Color

The internal color of cooked batter was determined through a CR-400 chromameter (Minolta Camera Co., Japan). The fresh samples with various amount of ST or SB were determined within 1 min.

2.7. Texture measured

The texture profile analysis of cooked batter was measured according to the method of Zhu et al. (2018). The values of hardness (N), springiness, adhesiveness and chewiness (N. mm) were obtained.

2.8. Raman spectroscopic

According to the method of Zhu et al. (2018), the changes of Raman spectroscopic in cooked batters with various amount of ST or SB were measured. The secondary structures were measured according to the amide I and III (Alix et al., 1988), the tertiary structures were measured from the Raman bands centered at 760 cm^{-1}, 830 cm^{-1} and 850 cm^{-1} (Herrero et al., 2008). All treatments were measured in four times.

2.9. Statistical analysis

The data was analyzed by an anlysis of variance (ANOVA) and the LSD procedure when significant differences ($p < 0.05$) were found (SPSS v.18.0).

3. Result and discussion

3.1. pH

Compared to the chicken batters with ST, the pH of the batters with SB were significantly increased ($p < 0.05$) (Table 1). At the same additive quantity, the pH of the batters with SB were increased about 0.13 units than that of ST. With the ST or SB increased, the pH was significantly increased ($p < 0.05$). It is well known that ST and SB are acid salt, and easily accept and donate protons, therefore, they have a good buffering capacity and can shift the pH of meat and meat products. In this study, the SB showed a stronger alkaline power in the

Table 1 pH and salt-soluble proteins of the raw chicken batters, and color (L*, a* and b* values) of the cooked chicken batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate.

Sample	pH	SSP	L* value	a* value	b* value
T1	6.12 ± 0.02^d	18.95 ± 0.95^d	83.26 ± 1.06^a	2.47 ± 0.14^a	11.62 ± 0.18^c
T2	6.19 ± 0.03^c	21.52 ± 0.87^c	82.75 ± 0.96^a	2.52 ± 0.12^a	11.37 ± 0.20^c
T3	6.26 ± 0.02^b	23.68 ± 0.82^b	80.42 ± 1.27^b	1.75 ± 0.18^b	12.07 ± 0.19^b
T4	6.32 ± 0.04^a	25.37 ± 0.76^a	80.19 ± 1.35^b	1.84 ± 0.15^b	13.39 ± 0.15^a

T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate. Each value represents the mean ± SD, n = 4.
$^{a\text{-}d}$Different parameter superscripts in the table indicate significant differences ($p < 0.05$).

chicken meat batters than ST, so the batters with SB had a higher pH. The result was agreement with Sheard and Tali (2004), who reported that the pH of pork meat were increased about 0.30 and 0.46 units when added the same concentration of ST or SB alone, respectively. Petracci et al. (2012) also found that the pH of marinaded broiler breast meat were increased 0.3 and 0.7 units when added the same concentration of ST or SB, respectively. Zhu et al. (2018) showed that the pH of chicken batter with 0.5% SB was higher approximately 0.4 units than that of adding sodium chloride. A similar finding manifesting that the pre- and post-chill broiler breast meat were marinated with 3% tetrasodium pyrophosphate or SB in 2% NaCl for 24 h at 4 °C, the pH of breasts with SB were higher than that of tetrasodium pyrophosphate (Sen et al., 2005).

3.2. Protein solubility

The changes of SSP concentrations for raw batters with various amounts of ST and SB were effected significantly (Table 1). The SSP concentrations of batters with SB were higher ($p < 0.05$) than the ST. Increased the ST or SB, the SSP concentrations were significantly increased ($p < 0.05$). It is well known that pH plays a key role for swelling and dissolving the myofibrillar proteins, because the myofibrillar proteins is improving when the pH is keep away from the isoelectric point (Lee et al., 2015). Kaewjumpol et al. (2013) found that both SB and phosphate could shift the isoelectric point and improve pH, that results in enhancing the SSP extraction. Besides the pH, the ionic strength of meat batter was another important factor to SSP concentrations. Generally, SB can completely dissociate into ions, therefore the batters with SB contained many ions. The use of pyrophosphate alone in chicken red and white muscles has a relatively small effect on ionic strength, and a strong synergistic effect was generated after the addition of sodium chloride. Some researchers reported that because bicarbonates have a higher ionic strength and buffering capacity, they could product a greater effect than the phosphates (Sen et al., 2005).

3.3. Cooking yield

Currently, the cooking yield is used to represent the water holding capacity and quality of raw batter. The cooking yield of the batters with various amount of ST or SB is shown in Fig. 1. Compared to the chicken batters with ST, the cooking yield of the batters with SB were higher ($p < 0.05$). The main reason is that the batters with SB had a higher pH and SSP concentrations than the ST. SSP, myosin and actin as the main component, which could form gel during heating. More SSP were dissolved and hydration of SSP were generated at higher pH, then more stable gel was formed and lowered the cooking loss (Xiong et al., 2012; Kang et al., 2014; Zhao et al., 2020). Some researchers have reported that the ability to bind and retain water of myofibrillar protein was increased when the pH was increased (Ni et al., 2014; Shen et al., 2019). Kaewthong and Wattanachant (2018) found that compared to the sodium chloride solution, ST and SB had greater capacities to increase the total yield of marinated broiler breast meat at the same electrical conductivity. In the present study, the cooking yield was significantly increased ($p < 0.05$) with the ST was increased. The reason is possible that the pH and SSP concentrations were increased with increasing ST (Table 1). Meanwhile, with increasing SB, the cooking yield was not significant differences ($p > 0.05$). The reason is that too much SB added, more carbon dioxide was generated, the gas caused some air holes was formed in the batter and declined the cooking yield (Zhu et al., 2018).

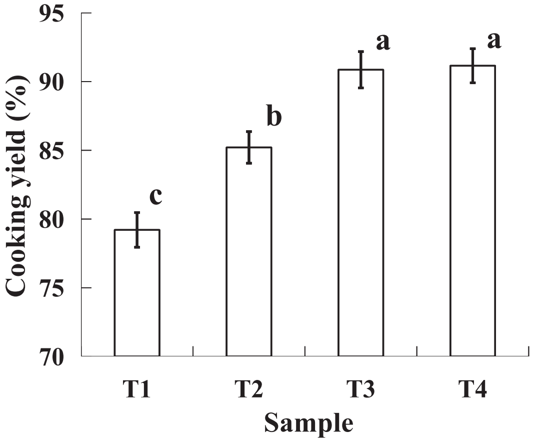

Fig. 1 The cooking yield (%) of the chicken batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate. T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate. Each value represents the mean ± SD, n = 4. $^{a\text{-}c}$Different parameter superscripts in the figure indicate significant differences ($p < 0.05$).

3.4. Color

The color of the cooked batters with various amount of ST or SB is presented in Table 1. Compared to the chicken batters with ST, the L* and a* values of the batters with SB were decreased significantly ($p < 0.05$), to the contrary, the b* value was improved significantly ($p < 0.05$). The L*, a* and b* values were not affected ($p > 0.05$) with ST increased. Meanwhile, the L* and a* values were not significant differences ($p > 0.05$) with ST increased, and b* value was increased significantly ($p < 0.05$) with increasing SB. The differences in color was possible caused by the differences of function between ST and SB, and the pH and SSP concentration in the batters (Table 1). Increased the pH of meat could enhance the thermal stability of myoglobin, and decrease the denaturation during cooking, therefore increased pinkness (Trout, 1989; Sen et al., 2005). Sen et al. (2005) found that compared to the cooked pre- and post-chill broiler breast meat were marinated with 3% tetrasodium pyrophosphate, the L* values of the SB were increased, b* values were decreased, but the a* values were increased in the pre-chill meat, and not affected in the post-chill meat. Asli and Mørkøre (2012) showed that due to the higher muscle pH, the L* values of salted Atlantic cod with SB had a tendency of decline. In the ground meat products, the L* value was not significant differences ($p > 0.05$) when the SB or sodium chloride was added (Mohan et al., 2016; Zhu et al., 2018). The differences were produced by the different types of meat products (emulsion and cured meat products). The other, the addition of ST or SB improved the susceptibility of myoglobin during heat denaturation. Thus, the SB had a stronger effect on enhancing the susceptibility of myoglobin in the chicken meat batters than that of ST.

3.5. Texture properties

The texture parameters of cooked chicken batters were effected by the various amount of ST or SB is shown in Table 2. Compared to the chicken batters with ST, the hardness, springiness, adhesiveness, and chewiness of the batters with SB were increased significantly ($p < 0.05$), and the hardness, springiness, adhesiveness, and chewiness were increased significantly ($p < 0.05$) with the ST or SB increased. Replaced the ST by SB, and increased the ST or SB, they all improved the ionic strength and pH, increased SSP concentration (Table 1). SSP determines the textural properties of cooked batters, increased SSP concentration could enhance the protein-protein, protein-water and protein-fat interactions, and induce more myosin and actin unfolding, which promoted to form good three-dimensional gels after heating (Zhang et al., 2018; Zheng et al., 2019). The other, higher SSP concentrations promoted more larger size protein aggregations were formed, which induced the gel matrix was more stable and elastic (Kang et al., 2018). Moreover, some researchers reported that the SB increased protein solubility, turbidity of soluble fraction and reactive SH groups, and weaken actomyosin interaction and hydrogen bonds (Chantarasuwan et al., 2011; Saleem et al., 2015). But a relatively small effect on ionic strength was produced in chicken muscles when used pyrophosphate alone, which in combination with sodium chloride can increase ionic strength and as a consequence the solubility of muscular proteins improved (Petracci et al., 2013). Thus, the cooked batters with SB had a better texture than that of ST.

3.6. Raman spectroscopic analysis

ST and SB are alkaline, which have an influence on protein structure and break the bonds between proteins. The protein conformation is a key role to research the properties of meat proteins, such as β-sheet structure, Tryptophan and Tyrosine residues (Alix et al., 1988; Li-Chan, 1996). A typical Raman spectroscopic of cooked batters with various amount of ST or SB in the 700–1800 cm^{-1} is presented in Fig. 2. Table 3 and 4 show the quantitative analysis of the selected peaks from Raman bands, respectively, which indicated with numbers in the Fig. 2.

3.6.1. Secondary structure

The amide I vibrational mode of Raman spectra of cooked batters locates at the intense band about 1660 cm^{-1}, which can supply the information about protein secondary structural, such as C—N stretching, C=O stretching and to lesser degrees of peptide groups (Li-Chan, 1996). The overlapping band of 1650–1660 cm^{-1}, 1665–1680 cm^{-1}, 1680 cm^{-1} and 1660–1665 cm^{-1} ranges are on behalf of α-helix, β-sheet, β-turn and random coil structures, respectively (Kang et al., 2017). The protein secondary structural were instability when the changes in the hydrogen bonding scheme referring the peptide linkages (Nunes et al., 2019). According to the Fig. 2, the amide I bands of cooked batters with 0.30% and 0.50% ST centered at 1660 ± 0.35 cm^{-1} and 1660 ± 0.46 cm^{-1}, respectively. When the 0.30% and 0.50% SB were added, the intensity maximum of amide I bands were slightly shift to 1661 ± 0.58 cm^{-1} and 1661 ± 0.58 cm^{-1}, respectively. The results

Table 2 Texture properties of the chicken batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate.

Sample	Hardness (N)	Springiness	Adhesiveness	Chewiness (N.mm)
T1	37.82 ± 1.22^{d}	0.702 ± 0.012^{d}	0.523 ± 0.011^{d}	18.09 ± 0.52^{d}
T2	42.35 ± 0.96^{c}	0.835 ± 0.015^{c}	0.618 ± 0.009^{c}	21.86 ± 0.48^{c}
T3	47.60 ± 1.05^{b}	0.856 ± 0.013^{b}	0.651 ± 0.014^{b}	27.03 ± 0.56^{b}
T4	51.26 ± 0.81^{a}	0.873 ± 0.016^{a}	0.681 ± 0.012^{a}	31.28 ± 0.61^{a}

T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate. Each value represents the mean ± SD, n = 4.

$^{a\text{-}d}$Different parameter superscripts in the figure indicate significant differences ($p < 0.05$).

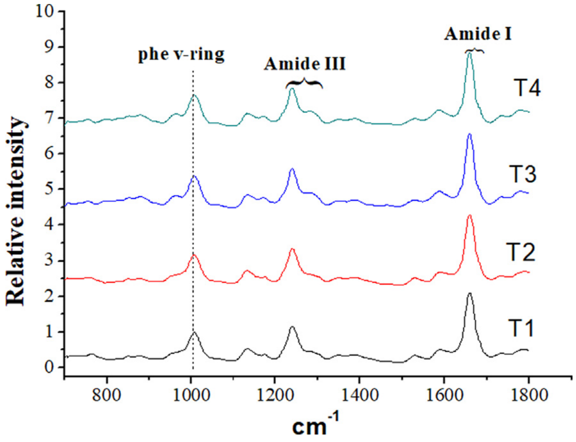

Fig. 2 Raman spectra of cooked chicken meat batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate in the region 700–1800 cm^{-1}. T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate.

meant that the improve of β-sheet content together with the decline of α-helix content (Ngarize et al., 2004).

The band of amide III is range from 1225 to 1350 cm^{-1}, which also supplies the information of secondary structural (Li-Chan, 1996; Herrero et al., 2008). A weak band locates range from 1260 to 1300 cm^{-1} indicates the proteins with higher α-helix structure, a more intense band locates range from 1230 to 1245 cm^{-1} indicates the β-sheet structure produced, and the band centers near 1245 cm^{-1} on behalf of the random coil structure form (Herrero et al., 2008). According to the Fig. 2, the amide III bands of cooked batters with 0.30% and 0.50% ST centered at 1268 ± 0.58 cm^{-1} and 1269 ± 0.83 cm^{-1}, and a weaker peak at 1242 ± 0.58 cm^{-1} and 1242 ± 0.58 cm^{-1}, respectively. Meanwhile, the peak of cooked batters with 0.30% and 0.50% SB was centered at 1274 ± 0.66 cm^{-1} and 1275 ± 0.58 cm^{-1}, and a weaker peak at 1237 ± 0.58 cm^{-1} and 1238 ± 1 cm^{-1}, respectively. These implied that added SB could increase the β-sheet structure content and lower the random coil structure content.

As we can see from the Table 3, compared to the batters with ST, there were significant effected the β-sheet and random coil structure contents in cooked batters with SB. A significant decrease in random coil structure and accompanied by a significant increase ($p < 0.05$) in β-sheet structure when the addition of SB. Added various amount of ST or SB have not effect ($p > 0.05$) on secondary structural, respectively. The possible reason is that the pH and ionic strength of raw batters with SB were higher than the ST, which induced more buried residues in protein molecules were exposed, more hydrogen bonds were formed, and protein molecules become more order, then more β-sheet structures were produced during the processing. Some researchers reported that the hardness of gel is enhancing with the β-sheet structure increasing, the result was agreement with the changes of hardness (Table 2) (Zhu et al., 2018). Thus, the use of SB could produce a greater effect on secondary structural than that of ST.

3.6.2. Tertiary structure

The information on the tertiary structure could been supplied by the bands of Raman spectra, such as 760 cm^{-1}, 830 cm^{-1} and 850 cm^{-1}, which involves mainly about hydrophobic interactions of tryptophan residues and tyrosine residues (Zhu et al., 2018).

The Raman band is centered near 760 cm^{-1} could provide the information about the stretching vibration of the tryptophan residues ring (Herrero et al., 2008). It was a significant difference ($p < 0.05$) in the normalized intensities of the 760 cm^{-1} among the cooked chicken meat batters were made with various amount of ST or SB (Table 4). Compared to the batters with ST, the Raman band locates at 760 cm^{-1} of the cooked batters with 0.30% and 0.50% SB was significantly decreased ($p < 0.05$) to 0.30 ± 0.03 and 0.28 ± 0.02, respectively. In addition, there was not significant differences ($p > 0.05$) in the 760 cm^{-1} with the ST or SB increased. The results manifested that the SB caused more hydrophobic microenvironment exposure to the polar aqueous solvent than the ST (Herrero et al., 2008; Kang et al., 2017).

The double Raman bands are appointed to vibrations of the *para*-substituted benzene ring about tyrosine residues, they centered at between 830 cm^{-1} and 850 cm^{-1}, respectively (Herrero et al., 2008). The I_{850}/I_{830} could provide the information about exposed or buried in the solvent of the microenvironment around tyrosyl residues. When I_{850}/I_{830} is the range 0.7 to 1.0, that indicated that the tyrosine residues are buried. On the contrary, when I_{850}/I_{830} is the range 0.90 to 2.5, that meant the tyrosine residues are exposed to the hydrophilic environment (Li-Chan, 1996). It was a significant difference ($p < 0.05$) in the I_{850}/I_{830} among the cooked chicken batters were made with various amount of ST or SB (Table 4). Compared to the batters with ST, the I_{850}/I_{830} of the cooked batters with 0.30% and 0.50% SB was significantly increased ($p < 0.05$) to 1.32 ± 0.02 and 1.37 ± 0.03, respectively. In

Table 3 Percentages of protein secondary structure (α-helice, β-sheet, β-turns, and random coil) in the cooked chicken batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate.

Sample	α-helice	β-sheet	β-turn	Random coil
T1	50.51 ± 2.23^{a}	21.98 ± 1.45^{b}	16.53 ± 0.53^{a}	11.43 ± 0.25^{a}
T2	49.35 ± 2.12^{a}	22.03 ± 1.37^{b}	16.64 ± 0.48^{a}	11.62 ± 0.17^{a}
T3	47.83 ± 1.96^{a}	25.82 ± 1.42^{a}	16.23 ± 0.42^{a}	10.58 ± 0.21^{b}
T4	47.33 ± 2.26^{a}	25.50 ± 1.50^{a}	16.43 ± 0.51^{a}	10.75 ± 0.19^{b}

T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate. Each value represents the mean ± SD, n = 4.

$^{a-b}$Different parameter superscripts in the figure indicate significant differences ($p < 0.05$).

Table 4 Normalized intensities of the 760 cm^{-1} (tryptophan) band and tyrosyl doublet at 850/830 cm^{-1} in the cooked chicken batters were made with various amount of sodium tripolyphosphate or sodium bicarbonate.

Sample	$I_{760/I1003}$	$I_{850/I830}$
T1	0.38 ± 0.02^a	1.23 ± 0.03^b
T2	0.35 ± 0.01^a	1.26 ± 0.01^b
T3	0.30 ± 0.03^b	1.32 ± 0.02^a
T4	0.28 ± 0.02^b	1.37 ± 0.03^a

T1: 0.3% sodium tripolyphosphate; T2: 0.5% sodium tripolyphosphate; T3: 0.3% sodium bicarbonate; T4: 0.5% sodium bicarbonate. Each value represents the mean ± SD, n = 4.
$^{a-b}$Different parameter superscripts in the figure indicate significant differences ($p < 0.05$).

addition, there was not significant differences ($p > 0.05$) in the I_{850}/I_{830} with the ST or SB increased. All the I_{850}/I_{830} of cooked chicken batters were larger than 1.0, which manifested that the ST and SB could induce more tyrosine residues became exposed to hydrophilic environment. In addition, there was not significant differences ($p > 0.05$) in the I_{850}/I_{830} with the ST or SB was increased. Overall, due to SB induced more protein denaturation than that of ST, it could promote the tyrosine residues to expose the hydrophilic environment.

4. Conclusion

In the study, the addition of 0.30% and 0.50% ST or SB were significantly effected the gel properties and protein conformation of chicken breast meat batters. The batters with SB have a higher pH, SSP concentrations, cooking yield, b* value and texture properties. Meanwhile, more β-sheet structure and hydrophobic interactions were formed than that of SB. On the contrary, the L* and a* values, random coil structure content and $I_{760/I1003}$ were decreased significantly. Increased the ST or SB, pH, SSP concentrations, hardness, springiness, adhesiveness, and chewiness were significantly increased, and the color and protein conformation had little affect. From the above, the use of SB could produce the chicken meat batter with higher cooking yield and a stronger gel network.

Declaration of Competing Interest

The authors (Fei Lu, Zhuang-Li Kang, Li-Peng Wei, Yan-ping Li) declare that there are no financial interests or personal relationships that will influence the work reported in this paper.

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