



## Effects of High Pressure on pH, Water-binding Capacity and Textural Properties of Pork Muscle Gels Containing Various Levels of Sodium Alginate

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**ABSTRACT :** The objective of this study was to investigate the effects of sodium alginate (SA) and pressurization levels on pH, water-binding and textural properties of pork muscle gels (PMG) containing salt. Ground lean pork with 1.0% NaCl and a given amount of SA (0.25, 0.5, 0.75 and 1.0%, respectively), was pressurized to 100, 200 or 300 MPa and subsequently gelled by heating. Results showed that addition of SA into pork muscle enhanced water-holding capacity (WHC) of PMG ( $p < 0.05$ ) as SA increased from 0.25% to 1.0%, with pH slightly increased ( $p > 0.05$ ). A decrease ( $p < 0.05$ ) was observed in all textural parameters (hardness, cohesiveness, springiness and chewiness). Pressurization had no effect on the tendency of WHC to increase or the decrease of the textural parameters. However, the effectiveness of pressurization to enhance textural properties of PMG was significant at some SA levels, especially  $\geq 200$  MPa and at  $\leq 0.75\%$  SA levels. Different combinations of pressure and SA levels could bring about variation in textural properties of PMG while SA enhanced WHC of pork muscle. The multiformity of the texture will open up a wide range of technological possibilities for the manufacture of pork-based restructured low-fat products. (**Key Words :** Sodium Alginate (SA), High Pressure, Water-holding Capacity (WHC), Textural Properties, Pork Muscle Gels (PMG))

### INTRODUCTION

Pork meat has been playing an important role in European and Chinese traditional food. However, consumers believe that pork meat contains a high amount of visible fat with a high content of saturated fatty acids and cholesterol (Toldra et al., 2004). Meanwhile, the role of fat as a binder in minced meat processing is widely known. A reduction in fat decreases many of the properties of comminuted meat products, such as texture and juiciness, (Cavestanya et al., 1994; Lurueña-Martínez et al., 2004). There were many reports suggesting various approaches to resolve this conflict, such as restructuring (Chin et al., 1999; Lurueña-Martínez et al., 2004).

Restructured muscle foods are important to the meat industry and investigations have been performed on restructuring methods of muscle including pressurization (Fernandez et al., 1998; Martin et al., 2002; Perez-Mateos et al., 2002) and the addition of various chemical ingredients and/or enzymes (Motzer et al., 1998; Chin et al., 1999; Perez-Mateos and Montero, 2000; Jarmoluk and Pietrasik, 2003; Pietrasik, 2003). Polysaccharides or hydrocolloids, derived from a variety of plants and microorganisms, are used extensively as ingredients in a number of ground meat products, to contribute to desirable binding characteristics, texture and appearance of the meat products (Chin et al., 1998; Perez-Mateos et al., 2000), and are considered as the most effective fat substitutes (Lurueña-Martínez et al., 2004).

Alginate is a polysaccharide and interaction has been found between it and meat proteins (Imeson et al., 1977; Bernal et al., 1987; Ensor et al., 1991). The algin/calcium gel restructured meat technology was patented in 1986 and approved by the Food Safety and Inspection Service of the United State Department of Agriculture for use in red meats (Ensor et al., 1989). Sodium alginate (SA) could improve the water holding capacity (WHC) and textural properties of

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**Table 1.** Composition of initial pork blend for the experiments

	GP (g)	Salt (g)	SA (g)	DW (ml)	Total (g)
0.00% SA (Control)	40.0	0.4	0.0	6.0	46.4
0.25% SA	40.0	0.4	0.1	5.9	46.4
0.50% SA	40.0	0.4	0.2	5.8	46.4
0.75% SA	40.0	0.4	0.3	5.7	46.4
1.00% SA	40.0	0.4	0.4	5.6	46.4

Deviation of weight of all additive ingredients were not in excess of  $\pm 0.8\%$ .

SA: Sodium alginate; GP: Ground pork; DW: Distilled water.

meat gels (Xiong and Blanchard, 1993; Perez-Mateos et al., 2000), and is also useful as a dietary fiber source for the prevention of obesity, hypercholesterolemia, and diabetes (Kimura et al., 1996). The supplementation of dietary complex containing SA had effects on meat qualities of pig and resulted in greatly higher carcass grade (Park et al., 2005). Thus, the application of SA in the production of pork products could provide a new approach towards increasing its value, such as supplying low-fat pork products (such as pork sausage trimmed off visible fat) for consumers.

However, there is no literature about the effects of various pressures on the characteristics of pork muscle gels (PMG) containing salt and different SA levels. The objective of this study was to investigate the effects of high pressure on pH, cooking loss (CL), WHC and textural properties of PMG containing various levels of SA.

## MATERIALS AND METHODS

### Preparation of initial materials

SA powder of food grade (viscosity of its 1.0% water solution is 100-200 cps at 20°C) was supplied by Dainippon Pharmaceutical Co. Ltd, Salt (containing NaCl  $\geq 99\%$ ) was provided by Salt Industry Center of Japan. Roller frozen pork 1320<sup>®</sup>, prepared from the loins of the LWD breed, was used in this experiment. This sample was obtained from Danish Crown Co., through Itoham Meat Processing Co., Japan. The frozen pork was drawn from a freezing compartment (-20°C) and thawed overnight in a cold room (3-4°C). Visible fat (reducing the fat content) and connective tissue were trimmed off, and then the pork was cut into small pieces (approximately 20×15×15 mm). The meat pieces were mixed well within 10 min and divided into several portions (about 150 g per portion). Each portion was transferred to a polyethylene bag, vacuum sealed, and stored again. These bags were randomly drawn for each experiment within two months.

Frozen pork portions were thawed for 6-8 h at 3-4°C, and ground once through a 2.5 mm plate of a meat grinder at room temperature. The proximate composition of the ground pork (GP) was: crude protein 21.5±0.3%, crude fat

3.4±0.1%, moisture 75.0±0.2%, and ash 1.1±0.0% (all n = 3). The crude protein and crude fat of the GP were determined according to standard AOAC methods 981.10 and 960.39(a) (1995), respectively. The ash content was determined by mineralizing for 5 h at 550°C. These determinations were made after the GP was dried in a VD-800F Freeze Dryer (Taitec Co., Japan) for 24 h. Moisture content was determined by the method of freeze-drying for 24 h and then being dehydrated at 104°C to a constant weight.

Salt, SA and distilled water (DW) were added sequentially to the ground pork as shown in Table 1 and manually mixed in a container suspended in an ice bath with a scraper. The minced pork was wrapped with a layer of food film (Saran Wrap Co. Ltd., Japan) and stored overnight (about 12 h) in the cold room. The chilled pork was mixed again, stuffed into small polyethylene bags (Ø1.5×10 cm) without entrapped air, sealed with a True Sealer (NL-201J, Japan) and subsequently stored in an ice bath (within 6 h) until pressurization or heating.

### Pressurization of the minced pork

The stuffed bag was pressurized using a cold isostatic press apparatus NBIP 45-120-70 (Nikkiso K. K., Tokyo) following the method described by Suzuki, Watanabe, Iwamura, Ikeuchi and Saito (1990). 100, 200 or 300 MPa pressure was applied to samples for 10 min at 13-15°C, respectively. The pressurized samples were then stored in an ice bath (within 5 h) prior to further treatment and analysis.

### Gelation of the minced pork

Both non-pressurized and pressurized bags were heated for 10 min in a water bath held at 73-75°C. Heated samples were then immediately chilled in iced water for about 10 min. These samples were removed from the ice bath and held for 20-24 h in a cold room until being analyzed.

### Measurement of pH

One gram of sample was homogenized in 5 ml (v/w) of deionized water, and pH value was measured according to the method described by Suzuki et al. (1990). All determinations were carried out in triplicate.

### Measurement of water-binding capacity (WBC)

*Determination of cooking loss* : Chilled pork gels from heated bags were weighted for determining their CL according to the method of Pietrasik (2003) with a slight modification. The gels were blotted dry two times, with a folio paper towel each time. CL was calculated as a percentage based on the raw stuffed weight. All measurements were carried out in quadruplicate.

*Measurement of water holding capacity* : Measurement

**Table 2.** Effect of 200 MPa pressure and various SA levels on mean pH values of the pork muscle blends and PMG containing 1% salt (mean±standard error, n = 3)

	Initial blend	12 h	12 h-HP	Heated	HP-heated
0.0% SA (C)	6.45±0.005	6.45±0.008	6.45±0.005	6.58±0.005	6.58±0.000
0.25% SA	6.47±0.005	6.47±0.000	6.48±0.008	6.63±0.005	6.62±0.005
0.5% SA	6.45±0.008	6.46±0.005	6.49±0.000	6.61±0.005	6.61±0.008
0.75% SA	6.50±0.005	6.50±0.000	6.52±0.000	6.65±0.005	6.64±0.005
1.0% SA	6.50±0.009	6.51±0.005	6.54±0.005	6.71±0.005	6.69±0.000

pH of raw pork and 1% SA solution were 6.51±0.008 (in sextuple) and 7.01±0.005 (about 21.5°C, in triplicate), respectively.

Initial blend: minced pork containing 1% salt and various SA levels; 12 h: the blend after cold storing 12 h; 12 h-HP: the blend cold-stored for 12 h and then pressurized by 200 MPa; Heated: PMG non-pressurized and heated; HP-Heated: PMG pressurized by 200 MPa and then heated.

of WHC was performed following the procedure of Perez-Mateos et al. (2000). About 0.3 g (about 2×2×2 mm) of PMG was placed in a model W-MO Micro-Centricut with 0.45 µm filters (JuJi Field Inc., Japan). WHC was expressed as the percentage of the retained sample weight after centrifugation at 3,000 g for 30 min in relation to the corresponding initial gel weight (at least in triplicate).

### Textural profile analysis

Textural profile analysis (TPA) of the gel samples (Ø15 ×10 mm) was carried out at the room temperature by using a NRM-2002J Rheometer (Fudoh Co. Ltd., Tokyo) according to the method described by Okabe (1979) and Bourne (1978). All samples (in quadruplicate) were compressed twice to 40% of their original height with a cylinder adaptor (Ø10 mm) at a constant cross-head speed 60 mm/min, height up and down of 50 mm, full scale load range 2 kg, chart speed 12 cm/min and a voltage 10×0.1 mV. The measurement parameters for TPA were established on the basis of preliminary work. The textural values, namely hardness (HD) (peak force on first compression (N)), cohesiveness (CH) (the ratio of the area of the second compression to that of the first in force-displacement curve (dimensionless)), springiness (SPR) (distance the sample recovered after the first compression (mm)) and chewiness (CHW) (HD×CH×SPR (N×mm)), were computed.

### Statistical analysis

An F-test was used for statistical evaluation of the measurement parameters of PMG quality. The data were processed by analysis of variance. F-test's significant difference was used to determine differences between treatment means or levels. Statistical analysis of the experimental results was performed using an Excel program (Microsoft official Excel 2003 for Windows). All of the parameters were expressed as mean±standard error (SE).

## RESULTS AND DISCUSSION

### pH

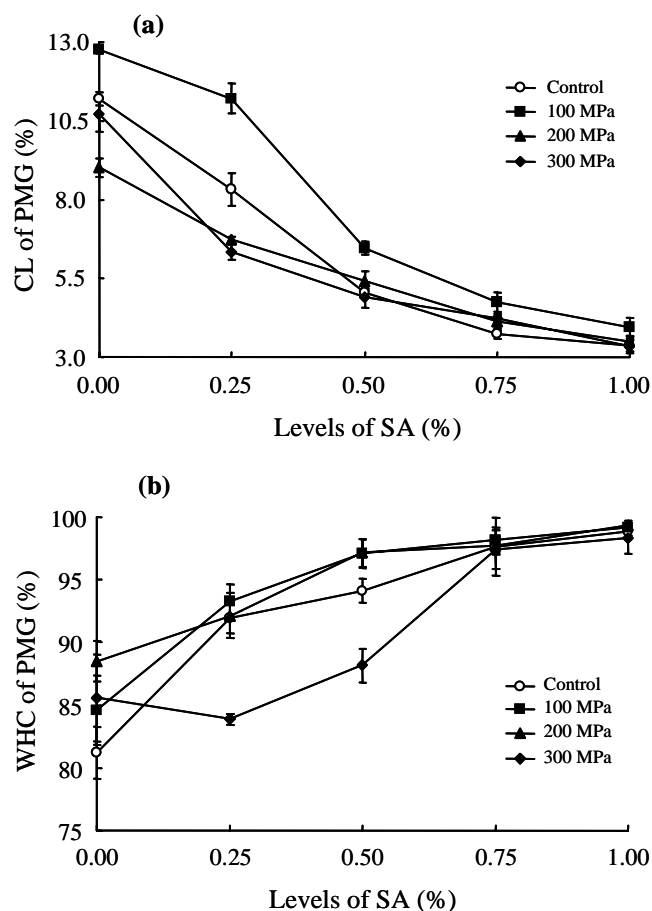
The effect of 200 MPa and various SA levels on pH of

the pork muscle blends and PMG containing 1.0% salt is shown in Table 2. pH of the blend increased slightly as the level of SA increased, and this change was retained after the formation of PMG caused by heating. Application of 200 MPa also caused a slight increase in pH ( $p>0.05$ ) (Table 2). The pressure-induced increase was similar to that observed from pressurized beef muscle (Han-Jun Ma and Ledward, 2004) and cod muscle (Angsupanich and Ledward, 1998). However, 200 MPa did not alter ultimate pH of PMG at all levels of SA ( $p>0.05$ ) (Table 2). Therefore, it is speculated that the increase in ultimate pH of PMG resulted chiefly from the level of SA used. Heating also increased pH as well as the addition of SA.

Application of 300 MPa was similar to that of 200 MPa, but 100 MPa was ineffective at altered pH (not listed in Table 2). Heat-induced increases of pH were significant in both pressurized and non-pressurized samples ( $p<0.01$ ). These results demonstrate that the mechanism of pH increase caused by pressurization on the blend was similar to heating, and supported the view that pressure-induced increases in pH is associated with the denaturation of some protein fractions in meat (Angsupanich et al., 1998).

### Cooking loss

Figure 1a shows that CL of both pressurized and non-pressurized PMG decreased gradually as SA concentration increased ( $p<0.05$ ). But the CL was increased by 100 MPa at all SA levels ( $p<0.05$ ). A similar increase was observed in a pressurized lean beef meat containing the only salt (Macfarlane, Mckenzie, Turner and Jones, 1984). 200 and 300 MPa resulted in decreased CL at 0.25% SA ( $p<0.05$ ). These decreases were parallel to the results obtained from SA-free pork at 200 and 400 MPa (Colmenero, Cofrades, Carballo, Fernandez and Martin, 1998) and lean beef meat at 150 MPa (Macfarlane et al., 1984). Furthermore, the CL values produced by both 200 and 300 MPa at  $\geq 0.75\%$  SA levels were higher than that of corresponding control, but the differences between pressure treatments at a given level of SA were insignificant ( $p>0.05$ ). Therefore, these pressure-induced results implied that there was possibly the most proper level of pressurization in reducing CL of PMG, which might be near to 200 MPa.



**Figure 1.** Effects of pressurization on CL (a) and WHC (b) of PMG at various SA levels.

### WHC

As shown in Figure 1b, SA showed very strong capacity in enhancing WHC of PMG. WHC of non-pressurized PMG increased significantly with the elevation of SA level from 0.25% to 1.0% ( $p < 0.01$ ), and increased up to 98.8% from 81.2% though the initial pork contained about 15% additional water. Such an increase was similar to the results observed in SSP (salt-soluble protein) alginate composite gels (Xiong et al., 1993) and in blue whiting muscle system (Perez-Mateos et al., 2000), but also was consistent with the decrease in CL. This was also in support of the views, that relative differences of WHC of raw meat are retained after heating (Macfarlane et al., 1984), and that the higher the ultimate pH the less will be the diminution in WHC (Lawrie, 1991), because alginate is a hydrocolloid and effective gelling agent of muscular protein network structure (Perez-Mateos et al., 2002), and SA revealed the tendency of increasing pH of pork muscle.

SA-induced WHC was influenced by pressure level only at  $\leq 0.5\%$  SA (Figure 1b). The WHC at all SA levels increased under 100 and 200 MPa despite unobvious ( $p > 0.05$ ). But it declined evidently under 300 MPa at 0.25% and 0.5% SA ( $p < 0.01$ ), and became unchangeable at

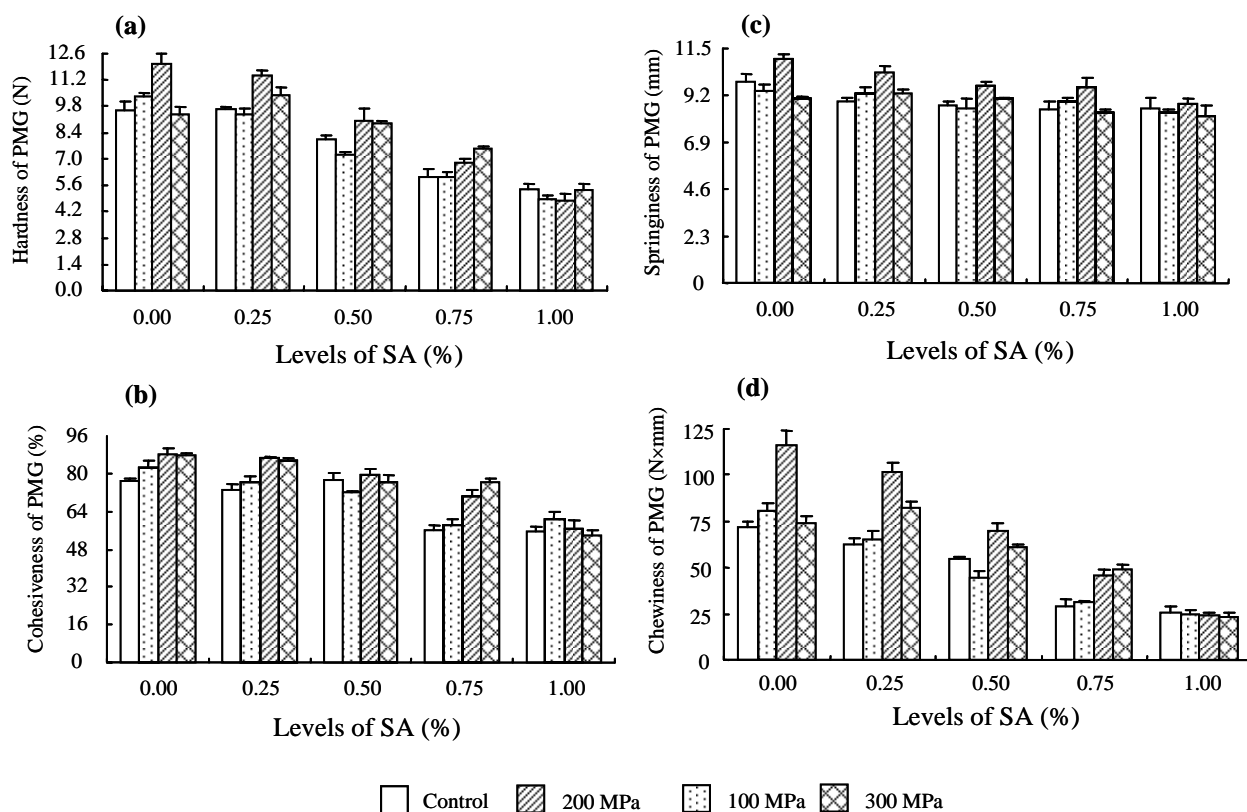
$\geq 0.75\%$  SA. Such pressure-induced pattern was similar to the result described in blue whiting gels with 0.5% SA at 200 and 375 MPa (Perez-Mateos et al., 2002). In addition, it showed that pressurization appeared to enhance the effectiveness of salt in increasing water-binding (Macfarlane et al., 1984; Martin et al., 2002), from the changes of CL and WHC of pressurized SA-free gels (at the level of 0.00% SA).

The mechanism of water-binding in muscle/SA/salt and pressure system has not yet been fully understood. But previous results gave some of information on this. It is well known that pressurization results in the denaturation of myofibrillar proteins, such as myosin, actin and actomyosin. The degree of the denaturation was closely related to the decreased WBC of meat (Stabursvik et al., 1984). So, it should not be surprising to find that the WHC under 300 MPa declined evidently at 0.25% and 0.5% SA levels. However, such decline was unobvious at other SA levels. The mechanism of electrostatic interaction within the PMG also appeared to be seized of virtual evidence, because the increase of pH at elevated SA levels (Table 2) should result in moving farther away from the isoelectric point of salt-soluble protein, and hence an enhancement of electrostatic interaction and an increased WHC. Besides, the possibility of binding water via capillary mechanism needs to be further confirmed. But present results further suggested that water might be held mainly via hydrogen bond (Bernal et al., 1987), since the number of hydroxyl and carboxyl groups, which forming hydrogen bond force, increased theoretically with the elevation of SA concentration within the PMG.

### Textural properties

HD of both pressurized and non-pressurized PMG decreased continuously to great extents ( $>40\%$ ) with the increase of SA level from 0.25% to 1.0% (Figure 2a). This decrease was consistent with the result of water-binding mentioned. Meanwhile, the effect of pressurization on the HD was significant. Both 200 and 300 MPa increased the HD at 0.25-0.75% SA levels ( $p < 0.05$ ). The increase under 200 MPa was the highest at  $\leq 0.5\%$  SA and so was under 300 MPa at 0.75% SA. The effect under 100 MPa was limited except a decrease at 0.5% SA, and all three pressures exerted weak effects at 1.0% SA ( $p > 0.05$ ). Similar pressure-induced increases/decreases of HD in other meat gels have been reported by Fernandez et al. (1998) and Han-Jun Ma et al. (2004). This information suggested that there was some synergic relation between SA concentration and pressure level, with respect to the effect of both on HD.

CH of the gels also showed the tendency of decrease with the elevated SA levels. CH of non-pressurized gels did not change at  $\leq 0.5\%$  SA ( $p > 0.05$ ), but decreased evidently



**Figure 2.** Effects of pressurization on hardness (a), cohesiveness (b), springiness (c) and chewiness (d) of PMG at various SA levels.

at  $\geq 0.75\%$  ( $p < 0.01$ ). The effects of pressurization were complex (Figure 2b). Comparing with corresponding controls, 200 and 300 MPa resulted in obvious increases of CH at  $\leq 0.25\%$  and  $0.75\%$  SA, and were ineffective at  $0.5\%$  and  $1.0\%$  SA. A similar increase has also been observed in beef muscle gels (Han-Jun Ma et al., 2004). 100 MPa caused evident decrease of CH at  $0.5\%$  SA and obvious increase at  $1.0\%$  SA ( $p < 0.05$ ). However, in comparison with the non-pressurized sample free of SA, the increases under 200 and 300 MPa only occurred at  $\leq 0.25\%$  SA, the decrease appeared at  $1.0\%$  ( $p < 0.01$ ), and CH did not change at  $0.5$  and  $0.75\%$  ( $p > 0.05$ ). The difference of CH caused by both pressures was not significant except for sample of  $0.75\%$  SA. The obvious decrease of CH under 100 MPa appeared at  $\geq 0.5\%$  SA ( $p < 0.05$ ).

SPR of non-pressurized PMG was not influenced by added SA levels in the range of  $0.25$ - $1.0\%$  ( $p > 0.05$ ), but pressurization changed this pattern (Figure 2c). The pressurization of 200 MPa increased SPR of PMG at SA levels. However, the effects at both 100 and 300 MPa were limited. There was no evident variation of SPR between pressurized and non-pressurized samples at  $1.0\%$  SA. These phenomena were different from the results described in beef muscle by Han-Jun Ma et al. (2004), and in blue whiting muscle by Perez-Mateos et al. (2000), however, the result of SA-free sample was similar to the one observed in chicken

meat by Fernandez et al. (1998). The conflict was probably caused by different raw material, gelation and TPA determination conditions.

The result of CHW was similar to that of the hardness (Figure 2d). The effectiveness of pressurization decreased with the increase of SA level. There was no difference among non-pressurization and various pressure levels when SA concentration was up to  $1.0\%$ . Based on the relation between HD, CH, SPR and CHW, the TPA results appeared to indicate that the CHW of PMG was primarily dependent on the HD in both pressurized and non-pressurized samples because of the consistency between HD and CHW.

In addition, there were steep initial slopes and sharp peaks for the first bite in all force-distance curves of low-level SA samples ( $\leq 0.5\%$ ), and were no negative force areas but  $0.75\%$  SA levels. These implied that high rigidity and little adhesiveness of SA-free pork muscle gels were retained in the PMG containing  $\leq 0.5\%$  SA. Beyond  $0.75\%$  SA levels, however, the structure of crumbly gels was easily formed, regardless of pressurized or non-pressurized samples.

## CONCLUSIONS

Addition of SA into pork muscle could evidently enhance WHC of PMG ( $p < 0.05$ ), decrease CL ( $p < 0.05$ ) and

result in a slight increase of its pH ( $p > 0.05$ ). The degree of this enhancement depended primarily on the SA levels used, and the effectiveness of pressurization was minor, even reverse. However, the effect of pressurization on enhancing textural properties of PMG was significant; especially for those at  $\geq 200$  MPa and  $\leq 0.75\%$  SA levels, although the decreasing tendency of all TAP parameters with the elevated SA did not vary with pressurization. And it was important that different combinations of pressure and SA level could bring about variation of textural properties of PMG while SA enhanced WHC of pork muscle. Moreover, the multiformity of PMG texture will open up a wide range of technological possibilities for the manufacture of pork-based restructured low-fat products (such as pork sausage trimmed off visible fat), for example, a soft, juicy and low-chewy pork sausage (at 100 MPa and 1.0% SA) for older persons who have difficulty in mastication, or a firm (high rigidity and HD) and elastic pork food (at 200 MPa and 0.25% SA) for young people.

Besides, further research on the mechanism of the interaction between protein-SA-salt and pressurization should also be expected.

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