Contribution of Milkfish Sarcoplasmic Protein to the Thermal Gelation of Myofibrillar Protein

Wen-Ching Ko and Meng-Shan Hwang

Department of Food Science, National Chung Hsing University, Kuokuang Road, Taichung, Taiwan, ROC

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This research investigated sarcoplasmic protein (Sp-P) contribution to the thermal gelation of meat paste and myofibrillar protein. The Sp-P was recovered by an ultrafiltration unit from the wash water of milkfish meat paste preparations. Washed meat paste showed gel strength 1.4–2.0 times that of unwashed paste. This may be caused by the concentration effect of myofibrillar protein. Adding Sp-P increased gel strength in both washed and unwashed meat pastes. The larger the amount of Sp-P added, the stronger the effect. *Suwari* occurred at 40–50°C and *modori* occurred at 60–70°C for milkfish meat pastes irrespective of the addition of Sp-P. Sp-P addition (10 mg/g) caused *suwari* indices to change from 41 to 51, and *modori* indices from 37 to 33. These results show that the addition of Sp-P improves thermal gelation, has a promotive effect on *suwari* and a restrictive effect on *modori*.

Key words: thermal gelation, sarcoplasmic protein, myofibrillar protein, milkfish

Since water-soluble proteins of fish meat are regarded as a hindrance in gelation, they are generally removed by washing during surimi processing.^{1–4)} The operation increases costs and may also cause water pollution.⁵⁾ Some research has indicated that fish water-soluble proteins not only do not interfere with gel-formation of myofibrillar proteins, but also form a gel, if sufficiently concentrated.⁷⁾ Many methods such as heating, adding chemicals containing acid, alkali or coagulants, and bubble separation have been attempted to recover the proteins from the wastewater.^{8–13)} However, the low level of proteins in large amounts of wastewater makes the above processes uneconomical. These processes may also cause protein denaturing and even secondary contamination. In contrast, an ultrafiltration unit with MWCO 30,000 can be used to recover 70% of milkfish sarcoplasmic proteins (Sp-P) without denaturation.¹⁴⁾ Milkfish is an important fish species cultured in Taiwan. Its dorsal muscles are usually utilized as minced fish raw material and Sp-P is lost in wash water during processing. In this study, the recovered Sp-P was added to meat pastes to investigate the contribution to thermal gelation. The effect of Sp-P on *suwari* and *modori* was also determined.

Materials and Methods

Materials

Milkfish *Chanos chanos* weighing about 600 g was purchased from a local retail store, and was kept in ice before preparation of test samples. All chemicals used were of reagent grade.

Sarcoplasmic Protein (Sp-P) Preparation

Ordinary muscle carefully excised from milkfish was minced in a chopper (hole size 3 mm). Five vol. of cold phosphate buffer ($I=0.05$, pH 7.0) was added to the minced meat, then homogenized for 3 min. The supernatant obtained by centrifugation at $12,000 \times g$ for 15 min at 4°C

was collected as Sp-P solution.

Myofibrillar Protein Preparation

The minced milkfish muscle in 0.09 M KCl–5 mM EDTA–0.039 M borate buffer (pH 7.0) was homogenized for 5 min. After centrifugation at $10,000 \times g$ for 30 min, the supernatant was discarded. This process was repeated four times. The sediment obtained was used as myofibrillar protein.

Ultrafiltration Operation

Sp-P was recovered and concentrated by an ultrafiltration unit designed by Yeameei Co. of Taiwan. All conditions were the same as that used by Ko *et al.*¹⁴⁾

Meat Paste Preparation

Fish muscles were minced in a chopper (hole size 3 mm), and washed twice with 10 vol. of 0.3% NaHCO_3 –0.15% NaCl to remove blood and water-soluble components. It was then centrifuged at $6,500 \times g$ for 60 min to remove most of the moisture. The washed meat with or without Sp-P addition according to experimental conditions was adjusted at a fixed pH and desired moisture content. It was then ground manually with 2.5% NaCl of the total amount in a mortar at 4°C for 10 min to prepared meat paste.

Thermal Gelation Test

The meat pastes were sealed in stainless steel tubes (diameter 1.3 cm $\times$ height 1.3 cm), then heated in a water bath at various temperatures.

Gel Strength Measurement

A rheometer (San Kagaku CR-200D, Japan) with a ball type plunger of diameter 0.7 cm, table speed of 10 cm/min and force of 2,000 g was used. The gel strength was defined as the maximum stress (g) at the breaking point of a sample. Each measurement was made on five samples. Data were analyzed using the Statistical Analysis System.¹⁵⁾ The General Linear Model procedure for regression analyses,

ANOVA procedure for analysis of variance and Duncan's multiple range test for ranking the means were used.

Setting Index and Disintegration Index Calculation

Setting and disintegration indices were calculated according to the equation of Shimizu *et al.* as follows:¹⁶⁾

$$\text{Setting index} = G.S_{40^{\circ}\text{C}, 2\text{h}} / G.S_{60^{\circ}\text{C}, 20\text{min}}$$

$$\text{Disintegration index}$$

$$= (1 - G.S_{60^{\circ}\text{C}, 2\text{h}} / G.S_{60^{\circ}\text{C}, 20\text{min}}) \times 100$$

G.S = gel strength

Protein Determination

Protein concentration was determined by the biuret method with calibration using the Kjeldahl method.¹⁷⁾

Results and Discussion

Washed and unwashed meat pastes were adjusted to 80% moisture content and pH 6.8 with the addition of 2.5% NaCl. As shown in Fig. 1, the gel strength for washed meat pastes was 1.4–2.0 times that of unwashed paste. In the production of surimi, fish meat is generally washed to improve the gel strength and to obtain a good whiteness.⁵⁾ The increase in gel strength is due to the concentration of myofibrillar proteins rather than Sp-P removal.

Sp-P dissolved in the wash water was recovered by an ultrafiltration unit with a membrane of MWCO 30,000. The effect of Sp-P on the gel strength of washed and unwashed milkfish meat pastes was investigated. Whether the meat

pastes were washed or not, as shown in Table 1, gel strength clearly increased. Though there is no significant difference in breaking strain, Figs. 2 and 3 show that the higher the Sp-P concentration, the higher the gel strength for both meat pastes and myofibrillar proteins.

Figure 4 shows that, for milkfish meat pastes, *suwari*, a setting phenomenon, occurred at 40–50°C, and *modori*, a disintegration phenomenon, occurred at 60–70°C. The temperature ranges of *suwari* and *modori* were not affected by the content of Sp-P in the meat pastes.

Figure 5 shows the temperature-gelation curves of washed milkfish meat pastes with or without Sp-P addition. A *suwari* index of 41 at 40°C and a *modori* index of 37 at 60°C were obtained for the meat paste without Sp-P addition. As also shown in Fig. 5, the index of meat pastes with Sp-P addition (10 mg/g) changed from 41 to 51 and from 37 to 33 for *suwari* and *modori*, respectively. The finding shows the promotion effects by Sp-P on *suwari* and minor inhibition effects on *modori*.

The Sp-P not only had no effect on the temperature range for gelation of milkfish meat paste, but also contributed to its thermal gelation properties. This probably results from

Fig. 1. Relationship between gel strength and moisture content of washed and unwashed milkfish meat paste.
Heating condition: 90°C, 20 min.

Table 1. Effect of sarcoplasmic protein (Sp-P) on the gel strength of washed and unwashed milkfish meat pastes*

Treatment	Gel strength (g)	
	Washed	Unwashed
Sp-P addition (3.2 mg/g)	675.6 ^a	436.6 ^c
Water addition	593.2 ^b	342.8 ^d

* Minced meat pastes (moisture content 83%, pH 6.8, NaCl 2.5%) were treated at 90°C for 20 min before measuring gel strength.

^{a-d} Means not followed by the same letter are significantly different from each other by Duncan's Multiple Range Test at the $p < 0.05$ level.

Fig. 2. Effect of sarcoplasmic protein concentrate on gel strength of washed milkfish meat paste.

Heating condition: 90°C, 20 min.

Different letters (a-e) adjacent to a value indicate a significant difference ($p < 0.05$) among values.

Fig. 3. Effect of sarcoplasmic protein concentrate on gel strength of myofibrillar protein.

Heating condition: 90°C, 20 min.

Different letters (a-e) adjacent to a value indicate a significant difference ($p < 0.05$) among values.

Fig. 4. Effect of setting temperature and time on gel strength of washed milkfish meat paste. The meat pastes (moisture content 83%, pH 6.8, NaCl 2.5%) were set at 40–70°C for 5–120 min.

Fig. 5. Temperature-gelation curves of washed milkfish meat paste. The meat pastes with or without Sp-P addition (moisture content 83%, pH 6.8, NaCl 2.5%) were treated at different temperatures for 20 min or 2 h before measuring gel strength.

A: without Sp-P addition. B: with Sp-P addition.

Different letters (a–h) adjacent to a value indicate a significant difference ($p < 0.05$) among values.

its binding to myofibrillar proteins.¹⁾ From the standpoint of thermal gelation and water pollution, Sp-P removal is not necessary. On the other hand, the function of the Sp-P molecular weight below 30,000 and the characteristics of kamaboko gel with Sp-P addition must be investigated in detail. Whether or not the Sp-P of other fish species has the same effect also needs further research.

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