# Development of an intermediate foodstuff from freshwater fish in China

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#### Abstract

Frozen surimi is an intermediate foodstuff with high potential for long shelf life, for distribution over a wide area, and for the production of various texturized products. Hence, a vast quantity of cultured freshwater fish should be developed and utilized as new materials in surimi and surimi-based products in the near future.

According to three-dimensional contour maps showing the gel-forming properties of surimi, eight species of freshwater fish surimi were classified into two types: the 'V-valley-type' surimi (silver carp, big-head carp, Chinese snake-head and blunt-snout bream) shows easy setting, low resistance to gel collapse, high enhancement ability with two-step heating, and narrow optimum heating temperature and time area. These characteristics are similar to those of walleye pollack surimi. In contrast, the 'plateau-type' surimi (tilapia, grass carp, mud carp and common carp) is difficult to set, highly resistant to gel collapse, has no enhancement ability with two-step heating temperature and time area. As well, there are seasonal changes in the gelling properties of silver carp surimi, with the setting ability of surimi gel being higher in winter and lower in summer. In addition, myofibrillar protein thermal stability of silver carp surimi is higher in summer and lower in winter. Therefore, the setting ability of surimi gel is evidently affected by myofibrillar protein thermal stability, i.e. the bigger the inactivation rate of myofibrillar protein, the higher the rate of setting.

Sensory evaluations were conducted on kamaboko gels prepared from silver carp (SC) surimi and from walleye pollack (WP) surimi for comparison. Different specimens of kamaboko gel, produced with or without the extract from silver carp muscle (Esc) or walleye pollack muscle (Ewp), were tested for their odor, flavor, texture, whiteness and overall acceptability by a panel of college-age students in three locations. According to the sensory evaluation results in Kyoto, there were significant differences (P < 0.05) between SC kamaboko gel and WP kamaboko gel for all sensory evaluation indices. Compared to WP kamaboko gel, SC kamaboko gel was unacceptable to Japanese consumers, especially due to its odor. When the same investigations were made in Shanghai (a coastal area) and Wuhan (an inland area) of China, the former showed the same pattern as observed in Kyoto, but to a lesser extent; the latter exhibited a different pattern than in Kyoto, with fewer differences between all of the test specimens. Students from inland China found the marine fish meat gels and the freshwater fish meat gels to be equally acceptable. Sensory scores increased slightly when Ewp was added to kamaboko gels—in particular, the odor scoring of the SC kamaboko gel.

## Introduction

A new source of animal protein appeared in the latter half of the 20th century: freshwater fisheries resources produced mainly by aquaculture technology in inland water regions of China. China's freshwater fisheries catch has increased rapidly—more than ten-fold in the last 50 years—reaching more than 10 million t, which is equivalent to about 10% of the total world catch. However, freshwater fisheries resources lack diversity of consumption compared to ocean-based resources because the transportation and processing technologies of the freshwater fisheries remain behind those of ocean-based resources (Qian 1994; MOA 2000).

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To address the postharvest problems associated with this sector, China (Shanghai Fisheries University, SFU) and Japan (Japan International Research Center for Agricultural Sciences, JIRCAS) initiated a collaborative research project entitled 'Development of Technology for Utilization and Processing of Freshwater Fisheries Resources' (Fukuda and Uno 1998; Fukuda et al. 2000). Originally a Japanese term, 'surimi' is an intermediate foodstuff with high potential for production of a wide array of texturized products, such as imitation crab, and has a long frozen shelf life. Surimi is generally manufactured using simple technology which involves washing minced meat with water, then dehydrating it and mixing it with cryoprotectants. The initial aim of the surimi development research was to identify the gel-forming properties and acceptability of surimi made mainly from aquacultured freshwater fish in China—comparing it with walleye pollack surimi, a typical commercial marine fish surimi that accounts for more than 60% of world surimi production.

# **Materials and methods**

#### Materials

Silver carp (Hypophthalmichthys molitrix), bighead carp (Aristichthys nobilis), grass carp (Ctenopharyngodon idellus), common carp (Cyprinus carpio), Chinese snake-head (Ophiocephalus argus), blunt-snout bream (Megalobrama amblycephala), tilapia (Tilapia nilotica) and mud carp (Cirrhinus molitorella) were used as frozen surimi materials in this study. Walleye pollack (Theragra chalcogramma) surimi (SA grade), made by Nichiro Co. Ltd (Tokyo, Japan), was supplied by the Industrial Research Center of Ehime prefecture in Japan.

Extracts of silver carp meat (Esc) or walleye pollack meat (Ewp) were prepared using a hot water extraction method. Fresh, white dorsal meat was homogenized with three volumes of distilled water, and extracted for 30 min in a boiling water bath. After being cooled, the supernatant was centrifuged (10,000g for 15 min) and filtered. The final extract was made up to equal quantities of raw meat with evaporation.

#### Manufacture of frozen surimi

Freshwater fish were purchased alive, and minced meat was separated immediately from the headed, gutted and cleaned fish fillets through a belt drum type meat separator. The minced meat was washed four times in four times its weight of cold water. After the third washing cycle, a strainer was used to remove residual black skin, fine bone, and scale. For the last washing process, a 0.3% NaCl solution was used to aid the removal of the water. The water-washed meat was dehydrated in a centrifuge. Sugar, sorbitol, and polyphosphates as cryoprotectants were mixed into the dewatered meat at levels of 4, 4, and 0.3%, respectively. The prepared surimi was frozen at  $-40^{\circ}$ C for about 20 h and subsequently stored at  $-30^{\circ}$ C.

#### Preparation of heat-induced gel

To compare the gel-forming properties of the surimi manufactured from different freshwater fish species, the chemical composition of all surimi was made as similar as possible. That is, the contents of moisture, protein, and cryoprotectants in the half-thawed surimi were adjusted to 78.0%, 14.7%, and 7.3%, respectively, by adding water and the cryoprotectants as necessary. The surimi thus obtained was mixed with 3% NaCl at a controlled temperature of 0-5°C, using a high-speed, vacuum and cooling mixer (Steph Co.). The saltground surimi was stuffed into polyvinylidene chloride casing tubes, 22 mm in diameter and 70 mm in length. and heated for a maximum of 10 h in a water bath at 30, 40, 50, 60, 70 or 85°C. This was defined as the 'firststep heat-induced gel' (one-step heating gel or setting gel). A portion of the first-step heat-induced gel was then heated at 85°C for 30 minutes. This was defined as the 'second-step heat-induced gel' (two-step heating gel or cooked gel).

#### Assessment of gel strength

The breaking force and deformation of the heatinduced gel was measured using a rheometer (Type EZtest, Shimadzu Co. Ltd) with a spherical plunger of diameter 5 mm. For the measurements, the casing film was removed from the gel, then cylindrical samples of heat-induced gel, 22 mm in diameter and 30 mm in height, were prepared. The penetration speed of the plunger was established as 60 mm/min.

#### Calculation of gel disintegration rate

The first-order disintegration rate constant  $(K_D, s^{-1})$ of the gel breaking force was calculated using the following equation,  $K_D = (\ln G_0 - \ln G_1) \cdot 1/t$ , where  $G_0$ and  $G_1$  were the breaking force (g) before and after heating for t sec.

#### Preparation of myofibrils from surimi

Myofibrils were prepared from silver carp surimi as described by Katoh et al. (1979), with some modifications. Firstly, chopped surimi (5 g) was washed twice with six volumes of 0.1 M KCl, 20 mM Tris-HCl (pH 7.5). Washed surimi was then homogenized four times for 1 min each time at 12,000 rpm with an interval of 30 seconds in the above medium and centrifuged at 3°C and 8000g for 10 min. The supernatant was discarded, and this washing procedure was repeated four more times. The final suspension in the above medium was filtered through a layer of gauze to remove connective tissue contained in the surimi. The filtrate was used as the myofibril suspension. Protein concentration was measured by the biuret method, using bovine serum albumin as a standard.

### Assay of myofibrillar ATPase activity

Ca<sup>2+</sup>-ATPase was assayed at 25°C in a medium of 0.5 M KCl, 5 mM CaCl<sub>2</sub>, 25 mM Tris-HCl (pH 7.0) and 1 mM adenosine triphosphate (ATP). Liberated inorganic phosphate (P<sub>i</sub>) was determined by the Fiske and Subbarow method. The specific activity was expressed in  $\mu$ mol P<sub>i</sub>/min/mg of myofibrils. All samples were measured in duplicate or triplicate and their average values taken for statistical analysis.

#### Thermal stability of myofibrils

The myofibril suspension was heated at 40°C, and the residual Ca<sup>2+</sup>-ATPase activities were measured. The first order inactivation rate ( $K_D$ ) was estimated routinely (Hashimoto and Arai 1978). The measurements were performed in duplicate or triplicate.

#### Sensory evaluation

Sensory evaluation of 'kamaboko' (fish cake) gels prepared from different surimi samples was conducted by more than 60 university students specializing in food science and studying either at the Faculty of Agriculture, Kyoto University, Japan, Shanghai Fisheries University, Shanghai (coastal area in China), or Huazhong Agricultural University, Wuhan (inland area in China). For testing, casing gels were cut into bite-sized (3 mm) samples and, after being warmed slightly (around 25°C), all samples were served to the students randomly and at times other than during meal hours. All panels were asked to score four characteristics (odor, taste, texture, and whiteness), as well as overall desirability using a 5point hedonic scale (1, dislike extremely; 2, dislike moderately; 3, neither like nor dislike; 4, like moderately; and 5, like extremely). A score of 3 was the between division acceptable and unacceptable (Yoshikawa 1969; Furukawa 1994; Sheng et al. 1996).

#### Whiteness measurement of heated gels

Before conducting the penetration test, whiteness of the cylindrical cross-sectioned samples was measured immediately with a spectrophotometer (CM-2002, Minolta Co. Ltd, Osaka, Japan) for the values of L\*, a\*, and b\* (CIE Laboratory system) to the first decimal place. Whiteness (Park 2000), as an index for the general appearance of the test specimen, was calculated as: Whiteness =  $100 - [(100 - L^*)^2 + a^{*2} + b^{*2}]^{0.5}$ .

### Statistical analysis

The StatView for Windows (version 4.58, Abacus Concepts Inc., Cary, NC, USA) was used for data anal-

ysis. Analysis of variance was performed to compare the mean hedonic scores of all sensory indices among the different surimi gels. In all cases, the criterion for statistical significance was set at P < 0.05.

# **Results and discussion**

## Gel-forming properties demonstrated by threedimensional contour map

The breaking strength of the salt-ground surimi samples from the freshwater fish and walleye pollack that were heated at various temperatures and times were presented as three-dimensional contour maps. The shapes at intermediate temperatures differed greatly and were classified into two types.

Figure 1 shows typical contour maps of an easy-todisintegrate surimi gel and a difficult-to-disintegrate surimi gel when heated at an intermediate temperature (around 60°C). Four kinds of surimi made from silver carp, big-head carp, Chinese snake-head, and bluntsnout bream, and the walleye pollack surimi were classified into the former group. The breaking force of those gels during heating at 60°C in a water bath reached a maximum of about 400 g after 10 minutes. Those gels began to disintegrate when the breaking force began to decrease after 30 minutes, and lost their characteristics when the breaking force decreased from 200 to 100 g after 60 minutes.

The disintegration of those gels began to extend to not only 60°C, but the circumference temperature of 60°C as the heating time increased. Furthermore, the characteristic 'V-valley' became deeper and fanshaped. The V-valleys of silver carp, big-head carp, and Chinese snake-head spread from 45°C to 70°C, but those of the walleye pollack and blunt-snout bream were located at slightly lower temperatures. This suggested that the gel disintegration phenomenon influence existed not only in a narrow temperature range, but also the heat-induced gel formation of the lowtemperature side and the high-temperature side. Funatsu et al. (1996) reported that a myosin heavy chain was assumed to be derived from some low molecular components at the same time as a crosslinked myosin heavy chain was formed in the setting gel of the walleye pollack surimi. In the above freshwater fish surimi, the selection of the heating temperature is important for forming an elastic gel, since the collapsing area of the gel is relatively wide.

Surimi made from tilapia, grass carp, mud carp, and common carp was classified into a disintegrationresistant group. The breaking force of each gel heated to 60°C reached a maximum just like the easy-todisintegrate surimi gel 10 min after heating. Although the breaking force subsequently decreased after 30 minutes, the decrease was obviously smaller than that of the former five species. The breaking force of tilapia and grass carp surimi was maintained at about 300 g 3 h after heating. Those contour maps showed plateaus, and the V-valley was not observed even 5 h after heating. Therefore, the latter four species' surimi was called 'plateau-type' surimi, and had a wide optimal heating temperature and time range; the area where the breaking force of tilapia surimi exceeded 450 g was obviously wider than that of walleye pollack.

### Gel disintegration rate during heating at 60°C

The decrease in gel breaking force that occurred at 60°C heating was analyzed according to the first-order equation, and the disintegration rates calculated from the slopes of the lines are shown in Table 1. Tilapia, grass carp, and blunt-snout bream were classified into the 'slow' group; walleye pollack was classified into the 'middle' group; and silver carp, Chinese snakehead, and big-head carp were classified into the 'fast' group. The gel disintegration rates of the silver carp and big-head carp were about 15 times as fast as that of the tilapia.

#### Enhancement of gel strength by two-step heating

The two-step heating technique has been employed from ancient times in Japan to manufacture elastic food products from fish protein. In this study, the most remarkable enhancement of the gel strength by twostep heating was found in the walleye pollack surimi. The breaking forces of the first-step heating gels and the second-step heating gels were 448 g and 884 g, respectively. The setting and the two-step heating effects for gels of silver carp, big-head carp, and bluntsnout bream were observed, although the degree of those was small and required longer heating times in the first step compared with walleye pollack. However, since formation of the setting gel was not marked in common carp, tilapia, grass carp, and Chinese snakehead, the enhancement of two-step heating was not pronounced in those surimi. As described above, the two-step heating effect relates clearly to the formation of the setting gel (Numakura et al. 1985).

 Table 1.
 Disintegration rate constants of surimi gels heated at 60°C.

Fish	Disintegration rate constant $K_D \times 10^5 (s^{-1})$	Ratio
Tilapia	2.5	1.0
Grass carp	3.8	1.9
Brunt snout bream	4.7	1.9
Common carp	5.5	2.2
Mud carp	6.7	2.7
Walleye pollack	14.2	5.7
Silver carp	36.3	14.5
Chinese snake-head	36.3	14.5
Big-head carp	39.7	15.9

# Gel-forming ability of silver carp surimi manufactured in four seasons

Seasonal changes in setting ability at 30°C heating of silver carp surimi are shown in Figure 2. There were marked differences in the rate of breaking force required in the different seasons. The order of setting ability of the surimi gel samples was March (notable increase in breaking force of the gel), followed by

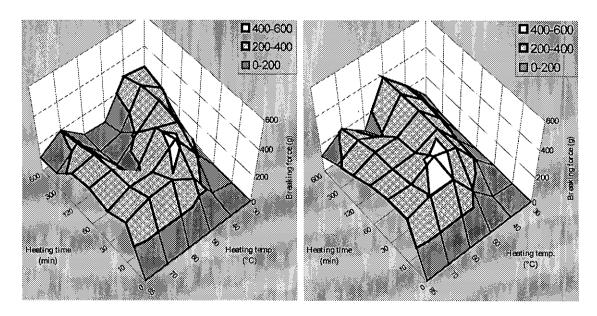


Figure 1. Contour maps for the easy-to-disintegrate silver carp surimi gel (V-valley-type surimi, left) and the difficult-to-disintegrate grass carp surimi gel (plateau-type surimi, right) by heating at an intermediate temperature (around 60°C).

November, September and June, i.e. the setting ability of surimi gel was higher in winter and lower in summer. Hence, it was concluded that silver carp clearly had an ability to set, and that the setting profile for the surimi was season-dependent.

#### Comparison of the myofibrillar thermal stability of surimi samples prepared in different seasons

Figure 3 shows changes in the myofibrillar thermal stability of silver carp surimi over four seasons. The inactivation rate constant,  $K_{D}$ , calculated from the slope of the straight line in logarithmic plots related to Ca-ATPase activity of myofibrillar protein versus incubation time, was used as the index of myofibrillar thermal stability, and the higher the  $K_D$  value, the lower myofibrillar thermal stability. There were different  $K_D$  values of myofibrillar protein among silver carp surimi prepared in different seasons. The order of myofibrillar thermal stability was September (slight decrease in

ATPase activity), June, March, and November (remarkable decrease in ATPase activity), i.e. myofibrillar thermal stability is higher in summer and lower in winter. This result was in accordance with previous reports (Guo and Watabe 1993; Wang et al. 1997). Therefore, the setting ability of surimi gels is evidently affected by myofibrillar protein thermal stability—the greater the inactivation rate of myofibrillar protein, the higher the rate of setting.

### Comparison of acceptability to Chinese and Japanese consumers of kamaboko gels prepared from silver carp surimi and walleye pollack surimi

The sensory evaluation results indicated that there were significant differences (P < 0.005) for all sensory characteristics (odor, taste, texture, and whiteness), as well as overall desirability between silver carp kamaboko gel and walleye pollack kamaboko gel

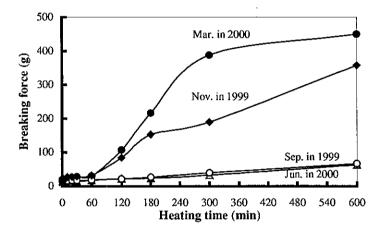


Figure 2. Seasonal changes in the setting ability of silver carp surimi heated at 30°C.

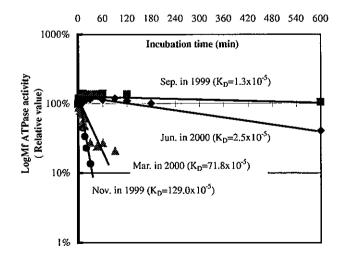


Figure 3. Seasonal changes in the myofibrillar thermal stability of silver carp surimi in different seasons (see text for details).

(Figure 4a) when evaluated by the Japanese consumers. The mean scores of silver carp kamaboko gel were all less than 3, whereas those of walleye pollack kamaboko gel were all more than 3. Thus, silver carp kamaboko gel was rated much lower in terms of odor and overall acceptability than walleye pollack kamaboko gel. Kamaboko gel derived from freshwater fish surimi was unacceptable to the Japanese, most probably because of its odor.

The same sensory tests accomplished in Shanghai (coastal area of China) and Wuhan (inland area of China) showed that the kamaboko gel made from walleve pollack surimi also obtained higher sensory scores than that from silver carp surimi (Figure 4b,c). However, silver carp kamaboko gel obtained higher sensory scores in China than in Japan, especially in its odor and overall desirability (Figure 4). Thus, kamaboko gels made from both walleye pollack and silver carp surimi were acceptable for Chinese consumers in Shanghai and Wuhan. The Chinese who live in the inland area especially are accustomed to and/or like to eat freshwater fish. The above findings indicate that there were some differences in the acceptability of fish food products, which are apparently influenced by people's eating habits and where they live.

# Effects of fish muscle extracts on the acceptability of kamaboko gels

In view of the results discussed in previous sections. odor and taste were considered to be the principal factors influencing the acceptability of kamaboko gels. Thus, it was predicted that different results would be obtained if kamaboko gel specimens were formed with or without the addition of muscle extract from walleye pollack (Ewp). As listed in Table 2, there was a slight increase in the sensory scores given in terms of odor, taste, texture, whiteness, and overall desirability of kamaboko gel when extract was added to silver carp kamaboko gel. The notable results were the significantly higher odor scorings (P < 0.05) given to silver carp surimi gel in which the extract of walleye pollack had been added as compared with the control (no addition of extract). In general, to improve the acceptability of silver carp kamaboko gel, the addition of its original and/or marine fish muscle extract is necessary.

According to the results of the mechanical measurements done on samples' gel properties (Table 3), there were also significant differences (P < 0.05) between silver carp kamaboko gel and walleye pollack kamaboko gel in terms of the breaking force and whiteness, whereby walleye pollack sample gel had higher

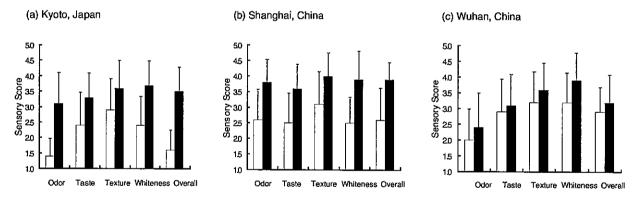


Figure 4. Comparison of acceptability of kamaboko gels derived from (□) silver carp surimi and from (■) walleye pollack surimi by Japanese and Chinese consumers. Values are shown as the mean ± standard deviation.

 Table 2. Effect of walleye pollack extracts on the acceptability of kamaboko gels derived from silver carp surimi and from walleye pollack surimi in Kyoto, Japan.

Sample	Odor	Taste	Texture	Whiteness	Overall desirability
SC-0	$1.4 \pm 0.6a$	2.4 ± 1.1a	2.9 ± 1.0a	2.4 ± 0.9a	1.6±0.6a
SC-Ewp	$1.9 \pm 1.0 \mathrm{b}$	$2.5 \pm 0.8a$	3.0 ± 0.9a	$2.9\pm0.9b$	$2.4 \pm 0.8b$
WP-0	$3.1 \pm 1.0c$	$3.3 \pm 0.8$ b,c	$3.6 \pm 0.9$ b,c	$3.7\pm0.8c$	$3.5\pm0.8c$
WP-Ewp	$3.1\pm0.9c$	$3.2 \pm 1.0c$	$3.6 \pm 1.0c$	$3.8\pm0.7c$	$3.4 \pm 0.8c$

Notes: Means with different letters are significantly different (P < 0.05), and those with the same letter represent no significant difference (P > 0.05) within a subgroup of a column.

Data are shown as mean  $\pm$  standard deviation (SC = silver carp surimi, WP = walleye pollack surimi, Ewp = walleye pollack extract added, 0 = no addition of extract).

values for both. Furthermore, a higher breaking force was observed in silver carp surimi gel to which the extract of walleye pollack had been added compared with the control. These physical results are in accordance with the aforementioned organoleptic data.

### Gel-forming property and acceptability of silver carp surimi gels as affected by the addition of trimethylamine N-oxide

Walleye pollack muscle and other marine products are rich in trimethylamine *N*-oxide (TMAO) and trimethylamine (TMA)—a dominant component of marine fishy odor (Tokunaga 1982). In contrast, there is no TMAO in silver carp muscle, and freshwater fishy odor comprises compounds other than TMA. It is of interest, therefore, to investigate the effect of adding TMAO on the quality of freshwater fish surimi gels.

The results (Table 4) show that the acceptability of kamaboko gels with the addition of TMAO had a higher score for its odor compared to the control T-0 (no addition of TMAO), presumably as a result of TMA derived from the TMAO additive. Meanwhile, the breaking force of the gels had higher values than the control. Treatment T-2 was the most effective of the

additive amounts. The results indicate that the acceptability of the silver carp surimi gel can be improved with the addition of TMAO.

Baskakov and Bolen (1998) reported that organic osmolytes, such as TMAO, have an extraordinary capability to force two thermodynamically unfolded proteins to fold to native-like species having significant functional activity, and maintains the structure and function of cellular proteins in organisms exposed to denaturing environmental stresses. In addition, Anthoni et al. (1990) also illustrated that TMAO and taurine have both been shown to stabilize enzymes against thermal denaturation and the former acts as a cryoprotectant during frozen storage. All of these findings may provide some explanation of the effect of TMAO on heat-induced gelling.

## Conclusion

THE present study demonstrated that the gel-forming properties of gels of freshwater fish surimi differ between species, heating temperatures/times and processing seasons. The acceptability of kamaboko gels also differs between Chinese and Japanese

 Table 3. Effect of walleye pollack extracts on the texture and whiteness of kamaboko gels derived from silver carp surimi and from walleye pollack surimi.

Item	SC-0	SC-Ewp	WP-0	WP-Ewp
Breaking force (g)	478 ± 48a	482 ± 41a	587 ±62b	568±57b
Breaking strain (mm)	13.1 ± 0.9a	12.8 ± 0.9a	12.9 ± 0.7a,b	$12.2 \pm 0.6b$
Whiteness	77.2 ± 0.8a	77.6 ± 1.1a	$81.1 \pm 0.6b$	80.2±0.5b

Notes: Means with different letters are significantly different (P < 0.05), and those with the same letter represent no significant difference (P > 0.05) within a subgroup of a row.

Data are shown as mean  $\pm$  standard deviation (SC = silver carp surimi, WP = walleye pollack surimi, Ewp = walleye pollack extract added,  $0 = n_0$  addition of extract).

Table 4. Effects of the addition of trimethylamine N-oxide (TMAO) on the quality of silver carp surimi gel.

Addition of TMAO	T-0	T-1	T-2	T-3
Gel strength				·
Breaking force (g)	431 ± 96a	473 ± 78a	$490 \pm 89b$	473 ± 119a
Breaking strain (mm)	$11.7 \pm 1.3$	$11.7 \pm 1.4$	$11.9 \pm 1.1$	$11.1 \pm 1.7$
Sensory evaluation				
Odor	$2.1 \pm 0.9a$	$2.4 \pm 0.5a$	$3.0 \pm 0.5b$	$2.4\pm0.7a$
Taste	$2.7 \pm 0.7a$	$2.7 \pm 1.1 \mathrm{ac}$	$2.9 \pm 1.1 bc$	$3.3 \pm 1.2 ac$
Texture	3.4 ± 1.2ab	3.7 ± 1.3ab	2.7 ± 0.9a	$3.2 \pm 0.8b$
Whiteness	3.1 ± 0.9a	3.8 ± 1.0a	$2.7\pm0.7b$	3.6 ± 0.7a
Overall desirability	$2.3 \pm 1.0$	$2.6\pm0.9$	$2.9\pm0.9$	$2.9 \pm 0.9$

Notes: T-0, T-1, T-2, and T-3 are increasing amounts TMAO added, i.e. 0, 167, 333, 500 mg/100 g (surimi weight basis), respectively. Heating conditions of gels were at 30°C setting for 1 h and subsequently at 85°C heating for 30 min. Means with different letters are significantly different (P < 0.05), and the same letter represents no significant difference (P > 0.05) within a subgroup of a row. Data are shown as mean  $\pm$  standard deviation.

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