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Effect of Washing Conditions on the Removal of Lipid from the Fatty Fish Escolar (*Lepidocybium flavobrunneum*) Meat

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Abstract: This study was carried out to explore methods to decrease the lipid content to less than 1% by washing escolar (*Lepidocybium flavobrunneum*) meat as well as the possibility of utilizing the obtained washed meat. For first washing, the escolar meat was washed with an alkaline solution under a different cooling condition. The once-washed meat was washed with palmitic sucrose ester (P-1670) solution in order to lower the lipid efficiently. The washed meat obtained was heated to prepare a gel whose quality was determined. When the temperature was maintained below 1°C during the first washing with an alkaline solution, the lipid and wax contents were 1.8 and 1.0%, respectively. After the once-washed meat was washed with a 0.25% (w/v) P-1670 solution, the lipid and wax contents decreased to 0.91 and 0.32%, respectively. While L/P and W/P ratios were 0.07 and 0.03, respectively. The strength and whiteness of the gel prepared from washed escolar meat were better than those of the gel prepared from commercial bigeye snapper surimi (SA grade). Thus, there is great potential to promote escolar as raw material for surimi production.

Key words: Escolar, surimi, lipid, wax, alkaline solution, sucrose ester

INTRODUCTION

Escolar (Lepidocybium flavobrunneum) Aburasokomutsu in Japanese is an underutilized fatty fish from the family Gempylidae and is widely distributed in the tropical and temperate seas of the world (Nakamura and Parín, 1993). This fish is frequently caught during tuna longline fishing and is discarded immediately (Milessi and Defeo, 2002). In 2003, escolar catch accounted for 16, 501 tons of the total bycatch species in longline fishing conducted by the Southern and Western Tuna and Brillfish Fishery (Lynch, 2004). This species does not metabolise wax ester that naturally occurs in its diet; Consequently, the wax ester is stored in its body. The lipid content of escolar meat is 18-22% and the lipid contains more than 90% wax ester (Karl and Rehbein, 2004; Nichols et al., 2001). If consumed, the wax ester causes diarrhoea and other acute gastrointestinal symptoms (Gregory, 2002; Kan et al., 2000; Shadbolt et al., 2002; Yohannes et al., 2002). Therefore, the use of escolar meat as food is prohibited in Japan.

Surimi is prepared from minced fish meat which is washed several times to produce a white, odourless, bland product and is mixed with cryoprotectants before freezing (Park and Morrissey, 2000). Recently, the new technology

of surimi manufacture and preservation of surimi products have been developed rapidly for supporting the increasing of population. The Alaska pollack is mainly used as raw material for surimi production, accounting for 50-70% of the total surimi production, but its quantity has been decreasing steadily (Park and Morrissey, 2000), there is a great interest in using the low value underutilized fish species such as escolar for surimi production. Recently, many other species, e.g., small pelagic fish, have been considered as a source of raw material for surimi production. However, producing surimi from small pelagic species such as sardine and mackerel is associated with drawbacks such as low pH and high lipid content of the meat. To resolve these problems, many researchers have used alkaline solutions in the meat-washing process in surimi production. Hultin and Kelleher (2000) also used an alkaline solution to obtain surimi of a better quality, particularly a high gel strength, white colour and the right flavour. Moreover, Undeland et al. (2002) reported that a washing process that includes acidic and alkaline solutions potentially overcomes some of the problems caused by the natural characteristics of the meat obtained from pelagic species.

In order to utilize escolar meat as raw material in surimi production, it is necessary to reduce the lipid and wax contents of the meat. If the greater part of the wax could be removed from the meat by alkaline washing, the escolar meat could be considered a potential raw material resource for seafood products, which are presently prepared largely from the Alaska pollack. Moreover, this step would enhance the economic value of the underutilized escolar fish. Therefore, in this study, we investigated the factors affecting the efficient removal of lipids for obtaining low-wax surimi with lipid content less than 1% and a lipid/protein (L/P) ratio of less than 0.10. In addition, we evaluated the quality of the heat-induced gel prepared from the washed meat.

MATERIALS AND METHODS

Materials: Frozen escolar (30-40 kg) was provided by the Ogasawara Fisheries Research Center and transported to the Laboratory of Aquatic Product Utilization, Kochi University. The fishes obtained were immediately cut with a mechanical saw into pieces of 4 cm thickness, packed in polyethylene bags and stored at -50°C until use.

Palmitic sucrose ester (P-1670) was purchased from Mitsubishi-Kagaku Foods Corporation (Tokyo, Japan). Other reagents were purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan).

Washing conditions: Ishiuchi et al. (1994) subjected escolar meat to alkaline bleaching in vacuum and reported that the lipid content could be decreased from 20.0% in unwashed meat to 5.8% in washed meat in case of a big fish (body weight, 9 kg) and from 19.2 to 3.1% in case of a small fish (body weight, 5 kg). This result suggested that alkaline bleaching in vacuum could not be applied to escolar meat because our objective was to investigate the factors affecting the efficient removal of lipids for obtaining low-wax surimi with lipid content less than 1% and a lipid/protein (L/P) ratio of less than 0.10. Chen et al. (1997) reported that pulverization of meat in an alkaline solution is effective for removal of lipids from mackerel meat. Therefore, in our experiments, escolar meat was pulverized in an alkaline solution by using a homogenizer (AM-1; Nihon Seiki Co., Ltd., Japan).

Washing conditions such as washing time, washing temperature, the meat/solution ratio and washing cycles were as follows. After thawing the frozen fish overnight at 5°C, the skin and red flesh were removed and the white flesh was chopped and passed through a 5 mm plate to obtain minced meat.

For effect of washing time, the minced meat was washed with 4 volumes of a cold alkaline solution (0.2% NaHCO₃ and 0.15% NaCl, ionic strength (I) = 0.05) at 5,000 rpm for 90 sec or 180 sec in a homogenizer with cooling in ice or ice-salt. The homogenate was filtered through

a 2 mm stainless steel sieve to separate the connective tissue. The washed meat was collected after centrifugation (CR 21E; Hitachi Koki, Tokyo, Japan) at 8,000 x g at 5°C for 15 min.

For effect of washing temperature, 19 samples of washed meats were prepared as mentioned above. After washing, the temperature of the homogenate was immediately measured using a digital thermometer (ND 500; Chino Corporation, Tokyo, Japan).

When the effect of the washing cycle (first, second and third cycles) at the different mate/solution ratio (1:2, 1:3 and 1:4) on the removal of lipids was investigated, once-washed meat was obtained by homogenization with an alkaline solution (0.2% NaHCO₃ and 0.15% NaCl) at 5,000 rpm for 90 sec with cooling in ice-salt as mentioned above. Twice- and thrice-washed meats were obtained by homogenization with 0.3% NaCl (I = 0.05) solution with cooling in ice before centrifugation. Twice washed meat was also filtered before centrifugation.

Effect of sucrose and palmitic sucrose ester on lipid removal: To decrease the lipid and wax contents efficiently, the second washing condition was further studied using sucrose and palmitic sucrose ester (P-1670; food-grade surfactant) solutions.

Effect of sucrose, after alkaline washing, the meat was further washed with 4 volumes of sucrose solution (at concentrations of 0, 1, 2, 4, 8 and 16%) containing 0.3% NaCl at 5,000 rpm for 90 sec with cooling in ice. To obtain washed meat, the sample was filtered through a 2 mm stainless steel sieve and centrifuged as mentioned above.

For P-1670 solution washing (at concentrations of 0, 0.05, 0.1, 0.25, 0.5, 1.0 and 2.0%), the meat washed once with an alkaline solution was also performed as the above mentioned sucrose solution washing.

In order to decrease the moisture content of meat washed with a P-1670 solution, the effect of NaCl on lipid removal during washing with the P-1670 solution was also determined in this study. P-1670 solutions of different concentrations (0, 0.125, 0.25 and 0.5%) containing 0.3% NaCl were used for washing the once-washed meat with cooling in ice in the second washing. The process was performed as the same P-1670 solution washing without 0.3% NaCl.

Chemical analysis: The washed meat obtained was analyzed for protein, lipid and wax contents, including determination of residual P-1670 in the twice-washed meat. The changes in the L/P and wax/protein (W/P) ratios of washed meat were also analyzed in this study. For unwashed meat, the proximate composition (moisture, crude ash, crude protein and crude lipid) and lipid composition were also determined.

Methods of the Association of Official Analytical Chemists (AOAC, 1995) were adopted to determine the levels of moisture, ash and crude protein (N×6.25). Crude lipid was extracted with chloroform and methanol (2:1, v/v) according to the method of Bligh and Dyer (1959).

While studying wax toxicity by an animal feeding test, Arai and Kinumaki (1977) examined the nutritive value of kamaboko prepared from the washed meat of a bericoid fish (*Hoplostethus* sp.) containing large amounts of wax. They reported that such kamaboko with lipid and protein contents of 1.8 and 17.3%, respectively, had good nutritive value as a protein source in the animal feeding test and abnormal symptoms such as seborrhoea were not observed. In this case, the L/P ratio of kamaboko was approximately 0.10 (1.8/17.3); therefore, we defined surimi with an L/P ratio of less than 0.10 as a less fatty surimi.

Lipid composition analysis: The wax, phospholipid and P-1670 contents were analyzed using a thin-layer chromatography-flame-ionization detector (TLC-FID) analyzer (Iatroscan MK-5; Iatron Co., Tokyo, Japan).

For wax and phospholipid contents, one aliquot of chloroform extract containing 50 mg lipid was spotted on silica gel (Chromarods-SIII) using a mini-cap disposable capillary (1 μ L). The spotted sample on the rods was developed using a solvent system comprising hexane/diethyl ether/acetic acid (80:20:1, v/v [%]). The rods were maintained at 110°C for 10 min after development. The dried rods were scanned using a TLC-FID analyzer to quantify the values of separated substances.

For P-1670 content, the method was done as the abovementioned wax and phospholipid analysis. A solvent system comprising chloroform/methanol/water (65:25:4, v/v [%]) was used for separation of P-1670.

Lipid distribution: The lipid distribution in the fish muscle was assayed according to the method of Thakur *et al.* (2003). A fish fillet was fixed in formalin solution containing 10% formalin and 2% calcium acetate and kept in a cold room for 1 month. Next, the formalin-fixed fillet was washed in flowing tap water for 3 h and cut into slices of 1 cm thickness. The sliced sample was rinsed in 60% isopropanol for 1 h, treated with a staining solution for lipids (Sudan II) for 20 min and rinsed again in 60% isopropanol for 3 h (solution changed 3 times). Then, the sample was washed in flowing tap water for 10 min and the sample with stained lipids was photographed.

Gel preparation: The washed meat obtained (to which were added 4% sorbitol, 4% sucrose, 0.1% sodium tripolyphosphate and 0.1% sodium pyrophosphate) or

commercial bigeye snapper surimi (SA grade) were chopped using a food processor (MK-K48; National, Tokyo, Japan) with 3.0% NaCl at 5°C for 2 min. The moisture content of both gels was adjusted to 80% with cold water. The pastes were stuffed into stainless steel tubes (diameter, 3.1 cm; height, 3.0 cm), wrapped in polyvinylidene film and heated at 80°C for 20 min. After heating, the gels were immediately cooled in ice water and maintained at 5°C for 24 h before gel strength measurement.

Gel strength measurement: Gels were equilibrated and tested at room temperature. The gels were sliced and cut into 5 ring-shaped samples of 0.5 cm thickness. The breaking force (g cm⁻²) and deformation ($\Delta L/L_0$) of the gels were measured by the stretching procedure using a rheometer (CR-200D; Sun Scientific Co., Ltd.) according to the method of Shimizu *et al.* (1981). The gel strength (g cm⁻²) was calculated by multiplying the breaking force (g cm⁻²) and elongation ($\Delta L/L_0$).

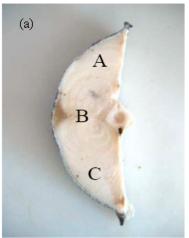
Whiteness measurement: The colour of surimi gels was determined using a Minolta chromameter (CR-300; Konica Minolta Co., Osaka, Japan). The L* (lightness), a* (redness/greenness) and b* (yellowness/blueness) values were measured and whiteness was calculated as described by Fujii *et al.* (1973) as follows:

Whiteness =
$$100 - [(100 - L^*)^2 + a^{*2} + b^{*2}]^{0.5}$$

Statistical analysis: All measurements were performed at least in duplicate and all results were expressed as mean values. For effect of washing conditions on lipid removal, the data was analyzed using SPSS software (SPSS 11.5 for windows, SPSS Inc., Chicago, IL) by the Duncan's multiple range test to compare the differences among means. To evaluate the level of significance of gel strength and whiteness, student's t-test was employed in this study. Significance was defined at p<0.05. Liner regression analysis was performed using Microsoft Excel version 2003.

RESULTS AND DISCUSSION

Proximate composition and lipid distribution: Proximate composition of different parts of the escolar muscle (predorsal, dorsal and tail) is shown in Table 1. Escolar meat from the above mentioned parts contained high levels of lipids, which were composed predominantly of wax with only minor amounts of phospholipids. The lipid and wax contents ranged from 21-25 and 20-25%, respectively,



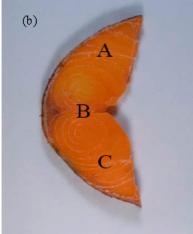


Fig. 1: A transverse section of muscle from the escolar *Lepidocybium flavobrunneum* stained for the distribution of total lipids and parts of the escolar muscle used for chemical analysis. A is the upper part, B is the middle part and C is the lower part of the dorsal. (a) Sample before staining and (b) Stained sample

Table 1: Proximate composition of the meat of the escolar Lepidocybium flavobrumsum

	Moisture	Protein	Lipid	Ash	Wax	Phospholipid	Lipid/Protein	Wax/Protein
Samples	(%)	(%)	(%)	(%)	(% of lipid)	(% of lipid)	ratio	ratio
Predorsal								
A	64.8±0.72	17.0±0.16	22.3±0.41	0.84±0.00	21.3±0.21	0.33±0.02	1.30±0.02	1.25±0.03
В	62.2±1.22	16.6±0.05	23.9±0.58	0.83±0.00	23.0±0.00	0.30±0.01	1.44±0.01	1.38±0.02
C	63.7±0.34	16.4±0.12	21.1±1.53	0.85±0.02	20.1±0.74	0.28±0.02	1.28±0.05	1.22±0.02
Dorsal								
A	63.2±0.19	15.6±0.30	22.8±0.76	0.79±0.00	22.0±0.34	0.22±0.01	1.46±0.03	1.41 ± 0.03
В	61.8±0.14	16.8±0.86	23.0±0.65	0.79±0.02	22.2±0.55	0.20±0.01	1.37±0.04	1.32±0.02
C	61.3±0.52	15.7±0.19	25.4±0.34	0.80±0.02	24.7±0.12	0.18±0.02	1.62±0.02	1.58±0.01
Tail								
A	63.3±0.00	17.0±0.26	21.9±0.43	0.85±0.00	21.2±0.96	0.21±0.02	1.29±0.01	1.25±0.02
В	63.5±0.02	17.0±0.02	22.5±0.62	0.79±0.00	21.7±1.44	0.22±0.04	1.32±0.03	1.27±0.03
C	62.8±0.12	16.4±0.33	24.3±0.19	0.84±0.00	23.0±0.05	0.30±0.01	1.48±0.04	1.40±0.01

The parts of escolar muscle used for chemical analysis: A is the upper part, B is the middle part and C is the lower part of the predorsal, dorsal and tail. Values are given as mean \pm SD (n=2)

while the phospholipid content ranged between 0.2 and 0.3%. Similar results were noted by Karl and Rehbein, (2004) and Nichols et al. (2001) in a study on the lipid composition of escolar fish. The moisture, protein and ash contents ranged from 61-65, 16-17 and 0.8-0.9%, respectively. The L/P and W/P ratios ranged from 1.3-1.6 and 1.2-1.6, respectively.

The lipid distribution in the escolar muscle is shown in Fig. 1. In the present lipid-staining method (Thakur $et\ al.$, 2003), the lipids deposited in the muscle were stained red. As shown in Fig. 1b, the escolar muscle was stained red by comparison with Fig. 1a, indicating that it had high lipid content with lipids distributed throughout the muscle.

Effect of washing time on lipid removal: The washing time had a definite impact on lipid removal. The residual lipid content of washed meat subjected to cooling in ice during washing for 90 and 180 sec was 3.21 and 3.79%, respectively (Fig. 2). The temperature of the homogenate after washing with cooling in ice was 6.3°C for

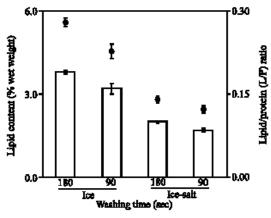


Fig. 2: Effect of washing time on lipid removal under cooling with ice and ice-salt. Error bars represent standard deviations (n = 3). (□) Lipid (%) and (●) L/P ratio

the 90 sec washing and 9.1°C for the 180 sec washing. This result suggested that the decrease in the lipid

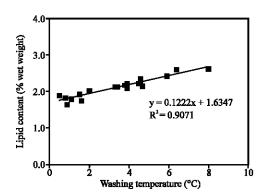


Fig. 3: Effect of washing temperature on lipid removal

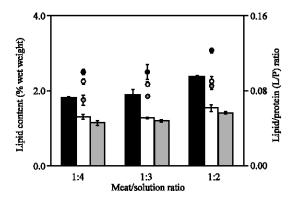


Fig. 4: Effect of the meat/solution ratio and washing cycle on lipid removal and the lipid/protein ratio. Error bars represent standard deviations (n = 2). (■), (●) First washing cycle; (□), (○) Second washing cycle; (■) (●)Third washing cycle; (■), (□), (□) Lipid (%) and (●) and (○), (●) L/P ratio

content of the washed meat might be related to the temperature during washing. Therefore, we applied the cooling in ice-salt condition during washing for 90 sec and 180 sec. The temperature of the homogenate after washing for 90 sec was 2.8°C and 180 sec was 5.7°C. When the temperature of the sample was kept low by cooling in ice-salt during washing, there was a marked removal of lipids from the meat. The residual lipid content of the sample subjected to cooling in ice-salt during washing was lower than that of sample subjected to cooling in ice during washing (p<0.05). The temperature was also effective against L/P ratio. When the temperature of the sample was kept by cooling in ice-salt during washing, the L/P ratio was lower than that of by cooling in ice (p<0.05). These results suggested that the washing temperature affects the removal of lipids.

In this experiment, a lower washing temperature (cooling in ice-salt) appeared to prevent the melting of lipids and assist in the formation of dense lipid particles. These particles had a lower density than the washing solution; therefore, the lipid particles floated on the surface of the washing solution. On the other hand, compared to the longer washing time, the shorter washing time was better suited for maintaining a low temperature throughout the process. When washing was performed for a longer time, the temperature rapidly increased, resulting in a lipid emulsion; therefore, the lipids could not separate from the solution. As a consequence of the longer washing time and higher temperature, a fraction of the lipids located at the surface might be returned to the washing solution.

To clarify the effect of temperature on lipid removal, alkaline washing was performed under different temperature conditions. Figure 3 shows the correlation between the lipid content of washed meat and the temperature of the homogenate immediately after washing. A significant positive correlation was observed between the residual lipid content and the temperature of the homogenate (p<0.01). On the other hand, no significant correlation was observed between the recovery of washed meat and the temperature of the homogenate immediately after washing (data not shown). Based on this result, it was confirmed that washing at a lower temperature was effective for removal of lipid from escolar meat and that the lipid and wax contents decreased to appropriate 1.8 and 1.0%, respectively, when the temperature was maintained below 1°C.

Effect of meat/solution ratio and washing cycle on lipid removal: The overall meat/solution ratio generally used by surimi manufacturers during washing ranges from 1:4-1:8 (Park and Morrissey, 2000). The effect of the meat/solution ratio and washing cycle on lipid removal from escolar meat was investigated. As shown in Fig. 4, the lipid content of washed meat and the L/P ratio decreased as the number of washing cycles increased. In each washing cycle, more lipids could be removed from the muscle at meat/solution ratios of 1:3 and 1:4 than at a meat/solution ratio of 1:2. No notable decrease in lipids was observed between washing at meat/solution ratios of 1:3 and 1:4 for each cycle. This result indicated that washing cycles with meat/solution ratios of 1:3 or 1:4 were effective for removal of lipids. According to Roussel and Cheftel (1988) and Ishikawa et al. (1977), lipid removal increased markedly with the number of washing cycles. After the third washing, the lipid content of sardine surimi prepared by manual and automatic washing processes decreased by 15-20% (Roussel and Cheftel, 1988) and 79.3% lipids were lost after the fourth washing

Table 2: Effect of various concentrations of palmitic sucrose ester (P-1670) solution on lipid removal, wax, other component of lipid, residual of P-1670, lipid/protein (L/P) and way/protein (W/P) ratios

Concentration of	Lipid	Wax	Other	Residual of	Lipid/protein	Wax/protein
P-1670, (%, w/v)	(%)	(%)	component (%)	P-1670 (%)	ratio	ratio
0	1.40 ± 0.04^{b}	$0.81\pm0.05^{\rm f}$	0.58 ± 0.03^{d}	-	0.09±0.01°	0.05±0.00°
0.05	1.17 ± 0.03 ab	0.61 ± 0.02^{e}	0.52±0.01°	0.03 ± 0.01^a	0.09±0.00°	$0.05\pm0.00^{\circ}$
0.1	0.99 ± 0.02^a	0.44 ± 0.08^{d}	0.43 ± 0.01^{b}	0.06 ± 0.01^a	0.08 ± 0.00^a	0.04 ± 0.00^{d}
0.25	0.91 ± 0.05^a	$0.32\pm0.01^{\circ}$	0.38 ± 0.05^a	0.21 ± 0.03^a	0.07±0.01a	$0.03\pm0.00^{\circ}$
0.5	1.25 ± 0.07^{ab}	0.27 ± 0.05^{bc}	0.48 ± 0.01^{bc}	0.55 ± 0.03^{b}	0.11±0.01 ^a	0.02 ± 0.01^{b}
1.0	$1.95\pm0.11^{\circ}$	0.22 ± 0.00^{ab}	0.48 ± 0.05^{bc}	$1.22\pm0.13^{\circ}$	0.19 ± 0.01^{b}	0.02 ± 0.00^{ab}
2.0	2.60 ± 0.54^{d}	0.16±0.04 ^a	$0.52\pm0.04^{\circ}$	1.94 ± 0.46^{d}	0.26±0.05°	0.02 ± 0.01^a

Values are given as mean±SD (n = 3). Different letter(s) within a column indicate significant differences (p<0.05)

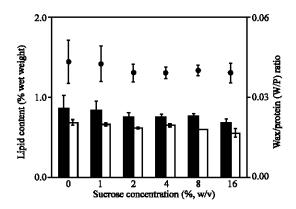


Fig. 5: Effect of various concentrations of sucrose solution on lipid removal and the wax/protein ratio.

Error bars represent standard deviations (n = 2).

(■) Wax (%), (□) Other components of lipids (%) and (●) W/P ratio

(Ishikawa *et al.*, 1977). In our experiment, the lipid content of thrice-washed meat processed at meat/solution ratios of 1:3 and 1:4 was 1.20 and 1.16%, respectively and approximately 95% of the initial lipids were removed by 3 washings.

The meat/solution ratio affected the recovery of the washed meat. When meat was washed at meat/solution ratios of 1:4, 1:3 and 1:2, the recovery rates of meat were 50, 43 and 39%, respectively.

Based on these results, washing at a meat/solution ratio of 1:4 appeared to be best condition that could be used for removing lipids from meat in this experiment and less fatty washed meat (lipids, 1.16%; proteins, 15.3% and L/P ratio, 0.07) could be obtained by 3 washings.

Effect of sucrose and P-1670 on lipid removal from meat subjected to alkaline washing: In order to determine a more efficient method of decreasing the lipid and wax contents, the once-washed meat was washed with various concentrations of sucrose and P-1670 solutions. Sucrose is a disaccharide and is usually used as a cryoprotectant in surimi processing. Sucrose esters are nonionic

surfactants; sucrose esters with various hydrophilic-lipophilic (hydrophilic-lipophilic balance, HLB) properties can be produced by using different fatty acids varying in their lipophilic chain length (Garti *et al.*, 1999). These surfactants are used in different industries, including pharmaceuticals, food processing, detergents, agriculture and so on (Garti *et al.*, 1999). P-1670 (HLB = 16) is approximately 70% pure monoester with palmitic acid. It has been widely used in wheat products, dairy products and substitutes, processed fats and oils, other edible substances and for miscellaneous uses.

Figure 5 shows the effect of various concentrations of sucrose solution containing 0.3% NaCl on lipid removal from meat subjected to alkaline washing. The lipid content of once-washed meat obtained by alkaline washing was 1.85%. As the sucrose concentration increased, the lipid content of the washed meat slightly decreased. The lipid content of washed meat obtained by washing with 0.3% NaCl solution (0% sucrose) was 1.55%. As the sucrose concentration in the 0.3% NaCl solution increased from 1-16%, the residual lipid content of twice-washed meat decreased from 1.49-1.24%. This result suggested that the addition of sucrose has a slight effect on lipid removal.

Table 2 shows the effect of P-1670 on the removal of lipids from once-washed meat obtained by alkaline washing. The lipid and wax contents of washed meat obtained by washing with a cold 0.3% NaCl (0% P-1670) solution were 1.40 and 0.81%, respectively. The lipid content decreased from 1.17-0.91% (p>0.05) as the P-1670 concentration increased from 0.05-0.25%; subsequently, the lipid content increased from 1.28-2.60% (p<0.05) with an increase in the P-1670 concentration (0.5-2.0%). On the other hand, the wax content of washed meat markedly decreased from 0.61-0.16% with an increase in the P-1670 concentration (p<0.05). The changes in the L/P and W/P ratios values were similar to those in the lipid and wax contents, respectively. In contrast, the residual P-1670 in the washed meat increased as the P-1670 concentration increased, particularly at concentrations greater than 0.25% (from 0.5-2.0%). The contents of other components of lipids ranged between 0.38 and 0.58%. These results

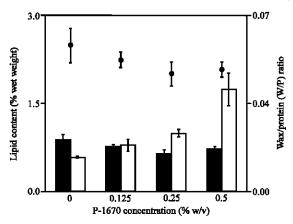


Fig. 6: Effect of various concentrations of palmitic sucrose ester (P-1670) solution containing 0.3%
NaCl on lipid removal and the wax/protein ratio.
Error bars represent standard deviations (n = 2).
(■) Wax (%), (□) Other components of lipids (%) and (●) W/P ratio

suggested that lipid and wax were removed more efficiently with the P-1670 solution than with the 0.3% NaCl solution (control).

Although wax removal after washing with 0.5-2.0% P-1670 solution was significantly greater than that with 0.25% P-1670 solution (p<0.05), the residual P-1670 content of washed meat with 0.5-2.0% P-1670 solution was significantly higher than that washed meat with 0.25% P-1670 solution (p<0.05) and the washed meat obtained was softer with the 0.5-2.0% P-1670 solution than with the 0.25% P-1670 solution. On the other hand, after washing with a 0.05-0.1% P-1670 solution, the residual P-1670 was lower than that remaining with the 0.25% P-1670 solution. However, the contents of wax and other components of lipids were significantly higher than those obtained with the 0.25% P-1670 solution (p<0.05). These results suggested that the use of the 0.25% P-1670 solution was appropriate for lipid removal from escolar meat. The lipid and wax contents of meat washed with the 0.25% P-1670 solution were 0.91 and 0.32%, respectively and the L/P and W/P ratios were 0.07 and 0.03, respectively.

P-1670 has an HLB value of 16 and dissolves in water better than lipids. It can be used as a surfactant for reducing the surface tension and viscosity of a solution. This property of P-1670 facilitated the removal of lipids from the solution and the flocculation of lipids on the surface.

The moisture content of washed meat obtained using the P-1670 solutions was higher than that obtained using the control solution. The moisture content of meat washed with 0.3% NaCl was 84% while that of meat washed with P-1670 was approximately 88%. In surimi

production, 0.1-0.3% NaCl solution is used as a solvent for washing in order to dewater the washed meat easily; washing in the absence of salt causes the meat to be swell, resulting in a high moisture content in the final product (Park and Morrissey, 2000). In this study, the P-1670 solution did not contain any salt. Thus, the high moisture content of meat washed with the P-1670 solution might be due to the absence of NaCl in the solution.

In order to decrease the moisture content of meat washed with a P-1670 solution, the effect of NaCl on lipid removal during washing with the P-1670 solution was studied as shown in Fig. 6. The lipid content increased as the P-1670 concentration increased; the lipid content was considerably higher with the 0.5% P-1670 solution in particular than with the control solution and the other P-1670 solutions (0.125 and 0.25%). The wax content decreased as the P-1670 concentration increased, with the exception of the 0.5% P-1670 solution that increased the wax content by a small amount. However, the wax content observed with all P-1670 solutions was considerably lower than that obtained with the control solution. In contrast, the content of other components of lipids was higher in washed meat prepared from all P-1670 solutions, particularly the 0.5% P-1670 solution, than in that prepared from the control solution. The W/P ratio was clearly lower with all P-1670 solutions than with the control solution. This result was very different from that obtained with P-1670 solutions (0.25 and 0.5%) not containing 0.3% NaCl (Table 2). The contents of lipids and other components of lipids in washed meat prepared from P-1670 solutions containing 0.3% NaCl were considerably higher than those in washed meat prepared from P-1670 solutions without 0.3% NaCl. P-1670 becomes partially insoluble in the presence of salt. This might affect removal of lipids. These results suggested that washing with a P-1670 solution containing 0.3% NaCl was not effective for lipid removal from meat subjected to alkaline washing.

Gel characteristics of washed meat treated with P-1670:

The texture (gel strength; Fig. 7a) and colour (whiteness; Fig. 7b) of washed meat were evaluated. Washed meat was prepared by washing meat with alkaline solution, followed by washing with 0.25% P-1670 solution. Kamaboko gel was prepared by heating at 80°C for 20 min. In Thailand, bigeye snapper is often used as raw material for surimi production. Commercially available kamaboko gel prepared from frozen bigeye snapper surimi (SA grade) was used for comparison. The strength of the gel prepared from washed escolar meat was higher than that of the commercial gel prepared from bigeye snapper surimi (p<0.05). The whiteness of the gel prepared from washed escolar meat was also higher than that of the commercial

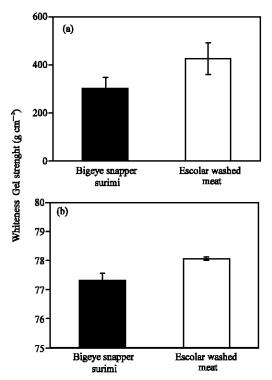


Fig. 7: Gel strength (g cm⁻²) (a) and whiteness (b) of gels prepared from washed escolar meat and bigeye snapper surimi by heating at 80°C for 20 min. Error bars represent standard deviations (n = 5 for gel strength and n = 3 for whiteness)

gel prepared from bigeye snapper surimi (p<0.05). Considering the strength and whiteness of the gel prepared from washed escolar meat, there is great potential to promote this fish as raw material for surimi production. The detailed characteristics of gel formation from washed escolar meat will be reported elsewhere.

CONCLUSIONS

The removal of lipids and decrease in wax in escolar meat appears to be greatly affected by temperature; it is best to maintain the temperature below 1°C during the first washing with alkaline solution. To lower the lipid and wax contents, the use of a P-1670 solution appears to be an alternative method of lipid removal after the first washing. As the demand for surimi continues, other fish species will be required as raw material for surimi production. Since the escolar is an underutilized fish and the quality (gel strength and whiteness) of the gel prepared from washed escolar meat was better than that prepared from commercial bigeye snapper surimi (SA grade), there is great potential to use this fish as raw material for surimi production.

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REFERENCES

- AOAC, 1995. Official Methods of Analysis. 16th Edn. American Association of Analytical Chemists, Washington, DC.
- Arai, K. and T. Kinumaki, 1977. Feeding test on nutritive value of kamaboko (fish jelly product) made from fish meat containing wax. Bull. Tokai Reg. Fish. Res. Lab., 91: 93-99.
- Bligh, E.G. and W.J. Dyer, 1959. A rapid method of total lipid extraction and purification. Can. J. Biochem. Physiol., 37 (8): 911-917.
- Chen, H.H., E.M. Chiu and J.R. Huang, 1997. Color and gel-forming properties of horse mackerel (*Trachurus japonicus*) as related to washing conditions. J. Food Sci., 62 (5): 985-991.
- Fujii, Y., K. Watanabe and Y. Maruyama, 1973. Relation between the ATP breakdown in ice-stored Alaska Pollack meat and the quality of frozen surimi. Bull. Tokai Reg. Fish. Res. Lab., 75: 7-11.
- Garti, N., V. Clement, M. Leser, A. Aserin and M. Fanun, 1999. Sucrose ester microemulsions. J. Mol. Liq., 80 (2-3): 253-296.
- Gregory, J., 2002. Outbreaks of diarrhoea associated with butterfish fish in Victoria. Commun. Dis. Intel., 26 (3): 439-440.
- Hultin, H.O. and S.D. Kelleher, 2000. Surimi Processing from Dark Muscle Fish. In: Surimi and Surimi Seafood, Park, J.W. (Ed.). Marcel Dekker, Inc., New York, pp. 59-77.
- Ishikawa, S., K. Nakamura and Y. Fujii, 1977. Fish jelly product (kamaboko) and frozen minced meat (frozen surimi) made of sardine. I. Freshness and handling of the fish material affecting its kamaboko forming ability. Bull. Tokai Reg. Fish. Res. Lab., 90: 59-66.
- Ishiuchi, Y., K. Matsuoka, M. Shiba, F. Nishioka, K. Masaki and Y. Murata, 1994. Utilization of escolar and oilfish meat as materials of fish jelly products. In: Proceedings of the 6th Research Promotion Meeting of Fisheries Products, National Research Institute of Fisheries Science, Fisheries Research Agency, Yokohama, Japan.
- Kan, K., H. Ushiyama, T. Shindo, S. Uehara and K. Yasuda, 2000. Outbreak of histamine poisoning due to ingestion of fish, Aburasokomutsu (*Lepidocybium flavobrunneum*). Yakugaku Zasshi, 41 (2): 117-121.

- Karl, H. and H. Rehbein, 2004. Butterfish on the German market. Dtsch. Lebensm.-Rundsch., 100 (5): 176-184.
- Lynch, A.W., 2004. Southern and Western Tuna and Billfish Fishery Data Summary 2003. Logbook Program, Australian Fisheries Management Authority, Canberra.
- Milessi, A.C. and O. Defeo, 2002. Long-term impact of incidental catches by tuna longlines: The black escolar (*Lepidocybium flavobrunneum*) of the Southwestern Atlantic Ocean. Fish. Res., 58 (2): 203-213.
- Nakamura, I. and N.V. Parín, 1993. FAO Species Catalogue. Snake Mackerels and Cutlassfishes of the World (Families Gempylidae and Trichiuridae). FAO, Rome, Italy.
- Nichols, P.D., B.D. Mooney and N.G. Elliott, 2001. Unusually high levels of non-saponifiable lipids in the fishes escolar and rudderfish: Identification by gas and thin-layer chromatography. J. Chromat. A, 936 (1-2): 183-191.
- Park, J.W. and M.T. Morrissey, 2000. Manufacturing of Surimi from Light Muscle Fish. In: Surimi and Surimi Seafood, Park J.W. (Ed.). Marcel Dekker, Inc., New York, pp. 23-58.

- Roussel, H. and J.C. Cheftel, 1988. Characteristics of surimi and kamaboko from sardine. Int. J. Food Sci. Tech., 23 (6): 607-623.
- Shadbolt, C., M. Kirk and P. Roche, 2002. Editorial: Diarrhoea associated with consumption of escolar (rudderfish). Commun. Dis. Intel., 26 (3): 1-5.
- Shimizu, Y., R. Machida and S. Takanemi, 1981. Species variations in the gel forming characteristics of fish meat paste. Nippon Suisan Gakkaishi, 47 (1): 95-104.
- Thakur, D.P., K. Morioka, Y. Itoh and A. Obatake, 2003. Lipid composition and deposition of cultured yellowtail *Seriola quinqueradiata* muscle at different anatomical locations in relation to meat texture. Fish. Sci., 69 (3): 487-494.
- Undeland, I., S.D. Kelleher and H.O. Hultin, 2002. Recovery of functional protein from herring (*Clupea harengus*) light muscle by acid or alkaline solubilization process. J. Agric. Food Chem., 50 (25): 7371-7379.
- Yohannes, K., C.B. Dalton, L. Halliday, L.E. Unicomb and M. Kirk, 2002. An outbreak of gastrointestinal illness associated with consumption of escolar fish. Commun. Dis. Intel., 26 (3): 441-445.