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Effect of Washing and Salt Concentration on the Gel Forming Ability of Two Tropical Fish Species

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ABSTRACT

Effect of washing solution, washing period and salt concentration on the gel properties of silver carp (*Hypophthalmichthys molitrix*) and pangas (*Pangasius hypophthalmus*) were investigated. Both silver carp and pangas mince were washed with 0, 0.05, 0.1, 0.15 and 0.2% NaCl for different washing period (5, 10, 15 minuts). The results showed that to obtain a good quality surimi fish mince required to be washed once with 0.1% NaCl and washing time should be limited within 10 minutes (7 min agitation, 3 minutes settling). To study the heating process for both fishes, the optimum setting temperature to obtain highest gel strength was found at 50°C. A two-step heating process (50°C for 2 hours prior to heating at 80°C for 30 minutes) gave the good gelling performance than one-step heating process (50°C for 2 hrs). To investigate the salt concentration on the gel functionalities of both fish minces, different amount of salt (1.0, 2.0, 3.0 and 4.0% NaCl) were used. Addition of 3% NaCl to the fish minces during grinding showed the highest gel strength regardless of heating schedule. The over all study shows that both tropical fresh fish can be utilized effectively for the preparation of surimi as raw material.

Key Words: Surimi; Gel forming ability; Washing; Salt concentration

INTRODUCTION

Surimi is a mechanically deboned, washed and dewatered minced fish flesh and a semi-purified concentrate of myofibrillar protein from fish muscle. When fish or meat muscle is ground with salt, it forms a viscous sol. The sol turns to an elastic gel upon heating. The thermal gelation of the sol is responsible for the elasticity of comminuted fish meat gel products (Sano *et al*., 1988). Washing is very important step in surimi processing. Washing is necessary to remove water-soluble substances, mainly sarcoplasmic proteins, fat and other undesirable materials like pigments. The removal of sarcoplasmic proteins concentrates myofibrillar proteins, which is the primary component in the formation of three-dimensional gel structure, responsible for the gel forming ability of surimi. The number of washing cycles and the volume of water vary with fish species, freshness of fish, type of washing unit and the desired quality of the surimi (Hall & Ahmad, 1997).

A somewhat crumbly fish flesh mince (or raw surimi) becomes a viscous sol or paste upon grinding or comminution with salt. Such a paste cannot be obtained in the absence of salt even if comminution is carried out for many hours. The main function of the salt is to help solubilize the myofibrillar protein to improve gel formation. Both myosin and actomyosin have dominant roles in surimi gelation and show species specificities with regard to gelation properties (Shimizu *et al*., 1983; Numakura *et al*., 1985). Although surimi is generally produced from cold-water marine species (Lanier, 1986; Kellher *et al*., 1992; Hultin & Kim, 1994). However, freshwater fish are excellent sources of high quality protein since they are well balanced in essential amino acids and highly digestible (Karmas & Lauber, 1987). The surimi making ability of many freshwater species could be upgraded by manipulating processing techniques (Onibala *et al*., 1997). Very recently, some investigations have been done on the quality of the mince of freshwater fish for the manufacture of surimi (Ismond & Tonogai, 1994; Lin & Morrissey, 1995; Kim *et al*., 1996). However, the information about the tropical freshwater fish as surimi raw material is very scanty.

Silver carp, a Chinese fast growing carp, and Thai pangas are extensively cultured in Bangladesh. These two fishes has a great aquaculture potential in Bangladesh due to its very high growth rate compared to other popular major carps. But in the peak season, the market price of these fishes often decline due to abundance of their production. This abundant catch of two fish species might be utilized as an alternative source of surimi raw material. But, till now, there is no information concerning the effect of washing and salt concentration on the gelling properties of silver carp and pangas fish mince. Therefore, the purpose of the study was to investigate the effect of washing and salt concentration on the gel-forming characteristics of silver carp and pangas fish mince in the tropical region. This study will generate the information on the tropical freshwater surimi technology and give the possibility of utilization of these tropical fishes as an effective source of surimi raw material.

MATERIALS AND METHODS

Fish. Fresh Silver carp (*Hypophthalmichthys molitrix*) and Pangas (*Pangasius hypophthalmus*) were collected from the local fishermen at the retail fish market, Mymensigh Mechua Bazar. The fishes were transported to the laboratory in proper iced condition in insulated icebox (Cosmos Ltd., Seoul, Korea 20 kg capacity). The average size of silver carp was 30.47± 1.24 cm and 1.0 ± 0.39 kg and Pangas was 33.56± 3.28 cm and 1.3 ± 0.43 kg.

Preparation of meat paste. On arrival at the laboratory the fishes (each size: 1.0 - 1.5 kg) were decapitated and gutted before washing by chilled fresh water. Sufficient time was lapsed to drain out excessive blood. The washed fish was filleted very carefully eliminating scales, skin, red muscles, belly flaps and kidney tissue. The fillet was deboned and minced by a manually operated meat mincer kept in cold condition previously (4^oC). Remaining bones and connective tissue fibers were removed from the meat by fine mesh sieve. The minced meat was washed with various washing solutions at variable washing frequencies and period at around 5^oC. For washing, the mince was stirred in 4 volumes of the washing solution for 5, 10 and 15 min for agitation and settling down before leaching. The meat was drained and pressed in a nylon bag after leaching and excess water was removed (15-20 kg/cm^2 for 20 min each time). The washed minces were ground with 3 % NaCl in a mortar for 20 min at below 10^oC with ice. The paste was carefully stuffed into polyethylene tube (2.8 cm diameter, 12.0 cm long). Two ends of the tube were wrapped by plastic paper before chilling in iced-water (4^oC). All procedures for mince preparation to final filling the paste were done at around 5-8^oC.

Proximate analysis. To characterize the mince types of two fishes moisture, ash, crude fat and crude protein were determined by methods of the Association of Official Analytical Chemists (1980). Results were mean values of three determinations and expressed as percentage muscle mince.

Washing procedure of the mince. The fish was cleaned with water, beheaded, eviscerated and minced by a mechanical mincer through a 1 mm orifice diameter so that all bones were removed from the muscle. The mince was washed with variable levels of NaCl (0, 0.05, 0.1, 0.15 and 0.2%). The mince was stirred in 4 volumes of washing solution with 1% NaCl for 5 minutes (3 min agitation + 2 min settling), 10 minutes (6 min agitation + 4 min settling) and 15 minutes (9 min agitation + 6 min settling). The meat was drained in a nylon bag after leaching and excess water was removed by pressing (15 kg/cm^2 for 15 min each time).

Gel preparation. The unwashed and washed minces were ground with 3% NaCl in a mortar for 20 min at 4°C. Required amount of cold distilled water was added to adjust the final moisture content to 80%. The paste was carefully stuffed into polyethylene tube (2.8 cm diameter, 12.0 cm long). Two ends of the tube were wrapped by parafilm and plastic paper before chilling in iced-water to retard changes in the meat until heating. All the procedures from handling fish to filling the paste were done at around 5°C. The paste in the tube was heated differently to produce gel. In one-step heating, the salt-ground pastes were heated in three different water baths at the temperatures of 40, 50, and 60°C for 2 hours. In two-step heating, the pastes once heated at 40, 50 and 60°C for 2 hours were further cooked at 80°C for 30 min. The resulting gels were immediately cooled in iced water for 30 min and kept at room temperature (24-25°C) for 1 hour before any measurement.

Puncture test. Puncture test was carried out on the gel sample (20 mm thickness) at a table speed of 6-cm/ min using a simplified Rheometer (Nowsad *et al*., 2000). Gel strength was expressed as the product of breaking strength (g) and deformation (cm)

Statistical analysis. In each replication, triplicate gel samples were tested. One-way analysis of variance (Steel & Torrie, 1980) was used to compare treatment effects. Least significant difference at $P < 0.01$ and significant F ratio were used to determine significant differences between mean values.

RESULTS AND DISCUSSION

Proximate composition. The proximate composition of both washed and unwashed meat of silver carp is presented in Table I. Silver carp is a high-moisture and low protein-lipid containing fish. Unwashed mince of silver carp had moisture 80.9%, protein 15.61%, lipid 3.1% and ash 0.93%. After washing the chemical composition was somewhat changed. Lipid is very important, as far as surimi concerned, because of its interference with the gel formation. The significant reduction in lipid content after washing could be explained by the fact that lipid substances were removed effectively by washing. Washing increased the moisture content and slightly protein contents and decreased the ash contents. In both fishes, it was clearly found that washed muscles have tendency to retain some water during washing even washed with salt water.

Lower moisture and slightly higher lipid content was found in pangas muscle than that of silver carp (Table I & Table II). In this study, we used moderate sized of pangas fish of one year or less than one year. Both fish were collected at end of April. Therefore, both fishes might have been harvested in the peak feeding period before spawning. Generally, fish harvested during the feeding period produced the highest quality surimi. During this period, fish muscle has the lowest moisture content as well as the highest total protein. However, Sylvia *et al*. (1994) reported that the highest moisture and lowest protein content of Pacific whiting was in April. Proximate composition of fish species varies according to the factors such as nutrition, living area, and fish size, catching season, seasonal and sexual variation as well as other environmental condition (Pacheco-Aguilar *et al*., 2000). For silver carp NPN value

Table I. Proximate composition, NPN and pH of silver carp mince

Mince	Moisture	Protein	Lipid	Ash	NPN	pH
Unwashed	80.9 ± 1.44	15.61 ± 0.82	3.1 ± 0.17	0.93± 0.09	0.43 ± 0.05	6.8 ± 0.22
Washed	82.45 ± 1.34	16.12 ± 0.53	0.63 ± 0.08	0.56 ± 0.07	0.32 ± 0.03	6.9 ± 0.42

Results were the mean ± S.E. of three individual measurements

Table II. Proximate composition, NPN and pH of pangas mince

Mince	Moisture	Protein	Lipid	Ash	NPN	pH
Unwashed	78.6 ± 2.14	16.5 ± 0.88	6.8 ± 0.39	0.78± 0.06	0.35 ± 0.04	6.6 ± 0.37
Washed	80.45 ± 1.62	16.8 ± 0.64	0.59 ± 0.05	0.43 ± 0.08	0.27 ± 0.03	6.7 ± 0.29

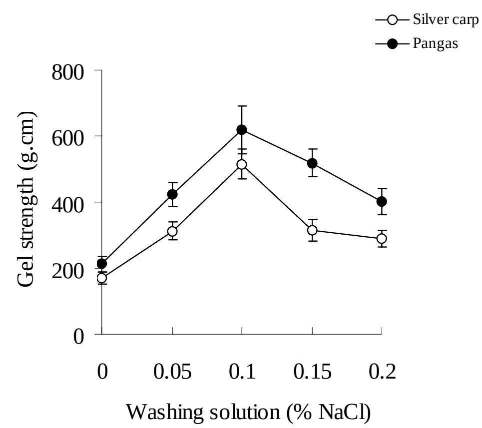
Results were the mean ± S.E. of three individual measurements

was 430 mg and 320 mg/100g (Table I) where as in Pangus NPN value was 350 mg and 270 mg/ 100 g (Table II) for unwashed and washed mince, respectively. Sakaguchi *et al.* (1982) reported the initial NPN values of 218 and 591 mg/100g in red and ordinary muscle of yellow fin tuna (*Seriola quinqueradiata*). However**,** Shimidu (1961) found NPN content in sardine varies from 500 to 600 mg/100 g. Our results are almost similar to those reported above. NPN in fish muscle is composed of low molecular weight amines, free amino acids and peptides (Riaz & Quadri, 1985). The results in this study indicated that washing procedure reduces the above low molecular substances in both silver and pangas fish meat.

The pH of both fish muscle was about neutral because of the pre-rigor prime quality fresh fish were used. Good quality gel cannot be formed if the muscle is outside the pH range 6.0-8.0 (Shimizu, 1975). Hashimoto & Arai (1978) reported that the denaturation rate of Pacific mackerel myofibrils at pH 5.8 is twice that measured at pH 6.5.

Effect of washing solution. To study the effect of washing solution on the gel forming ability, both silver carp and pangas fish minces were washed by 0, 0.05, 0.1, 0.15 and 0.2% NaCl (Fig. 1). Absence of salt in washing solution showed value of 170 and 212 g.cm for silver carp and pangas, respectively. The severe loss of MHC occurred when the fish was washed by 0% NaCl solution particularly for four washing cycles reported by Morrissey *et al.* (2000). Increase of salt amount progress the gel forming ability and highest gel forming ability was observed when the mince was washed with 0.1% NaCl showing the value of 515 and 620 g.cm for Silver carp and Pangas, respectively. Washing with 0.15 and 0.2% NaCl reduced the gel forming ability of both fishes. These results suggested addition of more salt during washing might cause a partial unfolding of proteins and increase the sensitivity to denaturation causing weaker gel matrix. Lin and Park (1996) found that washing with higher amount of NaCl solution reduced the loss of myofibrillar proteins that ultimately results the inferior gel forming ability. However, Wu *et al.* (1991) showed that the least amount of myofibrillar proteins of red hake mince was solubilized at 0.5% NaCl as well as higher gel forming ability. In the studies of Stefansson and Hultin (1994), solubility at very low ionic strength (near zero) was always much greater than that at high ionic strength.

Fig. 1. Effect of washing solution (% NaCl) on the gel strength of silver carp and pangas meat mince



Effect of washing period. To study the suitable washing time for the gel forming ability of Silver carp and pangas, the mince of both fishes was washed for 5 minutes (3 min agitation + 2 min settling), 10 minutes (6 min agitation + 4 min settling and 15 minutes (9 min agitation + 6 min settling) with 0.1% NaCl (Table III). Highest gel forming ability was obtained when both fish minces was washed for 10 minutes (6 min agitation + 4 min settling). The gel texture severely deteriorated at 15 minutes or above. Lin and Park (1997) reported that wash time to 10 minutes gives the highest gel forming ability with sufficient removal of sarcoplasmic proteins. In this study we found that two times washing gave the best gel performance (data is not shown). Subsequent washing removes the residual sarcoplasmic proteins along with small amount of myofibrillar proteins. Consequently, after the sarcoplasmic proteins are

Table III. Effect of washing period on the gel strength of silver carp and pangas mince

Mince	5 min	10 min	15 min
Silver carp	480 ± 33	518 ± 29	270 ± 23
Pangas	556 ± 43	623 ± 48	208 ± 19

Results were the mean ± S.E. of three individual measurements completely removed, further washing causes a severe loss of myofibrillar proteins. Therefore, excessive washing not only increases the cost for water use and waste water treatment but also result in a loss of myofibrillar proteins. Nishioka (1984) indicated that the greater number of washing cycles applied to mince produced the stronger gel-forming ability of surimi. He also showed that the relative amount of MHC increased as the washing was repeated. However, this trend was only effective at lower salt concentrations. Consequently, three and four washing cycles negatively affected the relative amount of myosin heavy chain in the washed meat at 0.5% or higher salt concentration. The number of washing cycles varies with fish species, freshness of fish, type of washing unit, and the desired quality of the surimi (Lee, 1984).

Effect of heating. Several research works have been done to find out the optimum heating temperature for gel forming ability of fishes and it varies with species (Shimizu *et al*., 1981; Lee, 1984). To investigate the suitable heating temperature unwashed and washed meat of both Silver carp and Pangas fish were heated at various temperatures (40, 50, and 60°C) for 20 minutes and 2 hours (Fig. 2 & 3). At 40°C, unwashed meat of both fish mince did not give the good texture even after heating for 2 hr. The highest gel strength was obtained in washed meat heated at 50°C for 2 hrs. Nowsad *et al*. (1999) also found that silver carp paste did not set at low incubation temperatures till 40^{o}C in one-step heating and the highest setting ability was found at around 50^{o}C. The present results are in accordance with the previous results. The gelation ability of fish muscles has been extensively examined and their optimum gelation conditions for surimi have been established (Lee, 1984). However, the gelation phenomenon in tropical freshwater carp did not exactly follow the pattern as generally observed in case of most marine species (Nowsad *et al*., 1999). At 60°C, gel-forming ability of both fish mince deteriorated resulting an inferior gel matrix suggesting proteolysis or gel degradation occurred at this temperature. The gel strength drop heating at the range of temperatures 50-60°C (Saeki *et al*., 1995) is a characteristics behavior of myofibrillar proteins of some fish species (Shimizu *et al*., 1983) due to proteolysis. In all heating temperatures, in both fishes, washed mince showed the higher gel strength than unwashed meat as washing could upgrade the gel forming ability.

Effect of salt concentration. To investigate the effect of salt concentration on the gel texture of Silver carp and Pangas various amount of salt (1, 2, 3 and 4%) were added in the washed fish mince (Fig 4 & 5). Both mince paste was heated at 50°C for 2 hours in one step heating. The salt ground pastes once heated at 50°C for 2 hours were further cooked at 80°C for 30 minutes in case of 2-step heating. Addition of 1% salt in the fish mince resulted the weaker gel showing value of 190 and 307 g.cm for 1-step and two step heating process, respectively, for silver carp (Fig. 4) and in case Pangas these values were 224 and 401 g.cm (Fig. 5). Gel strength increased with the increase of salt concentration and the highest gel strength was obtained at

Fig. 2. Effect of heating temperature on unwashed and washed meat gel of silver carp meat mince

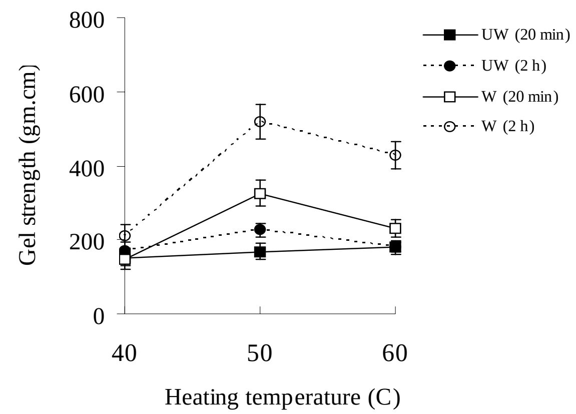


Fig. 3. Effect of heating temperature on unwashed and washed meat gel of pangas meat mince

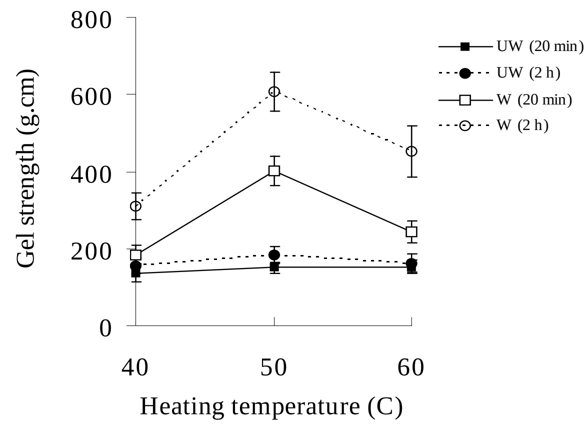


Fig. 4. Effect of salt concentration on the gel texture of washed meat mince of silver carp

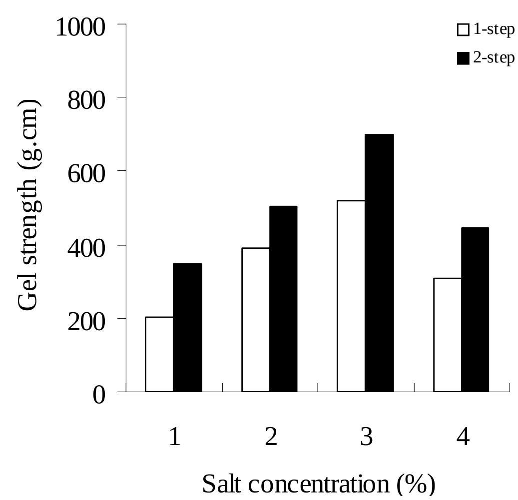
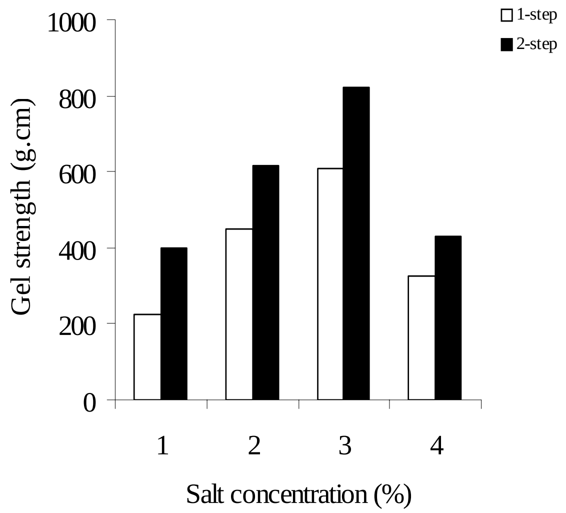


Fig. 5. Effect of salt concentration on the gel texture of washed meat mince of pangas



3% NaCl. Studies have shown that 1.7-3.5% NaCl is required for surimi to form an adequate gel (Roussel & Cheftel, 1990, Seki *et al*., 1998). Gel forming ability of both fish heated gel significantly decreased at 4% salt.

The overall study showed that surimi or surimi mince could be effectively produced from silver carp and pangas. Washed mince of both fish exhibited better textural properties than unwashed meat. To obtain good quality surimi, silver carp and pangas fish mince were required to be washed with 0.1% NaCl and the washing time should be limited within 10 min. Highest gel strength could be obtained from both mince ground with 3% NaCl and heating at 50°C in two step heating process.

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