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Effect of Salt Concentration and Cryoprotectants on Gel-forming Ability of Surimi Prepared from Queen Fish (*Chorinemus lysan*) During Frozen Storage

M. Kamal, M. Ismail Hossain, M.N. Sakib, F.H. Shikha, M. Neazuddin, M.A.J. Bapary and M.N. Islam
Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh

Abstract: Studies were conducted to evaluate the effect of salt concentration on the gel forming ability of surimi prepared from Queen fish (*Chorinemus lysan*) using various concentration of salt (0, 1, 2, 3, 4, 5 and 6%) in meat paste. In both one step (50°C for 2 h) and two steps heating process (50°C for 2 h prior to heating at 80°C for 30 min), maximum gel-strength was obtained at the salt concentration of 3% NaCl. In order to investigate the effect of cryoprotectants on the gel-forming ability of surimi prepared from Queen fish (*Chorinemus lysan*) during 3 weeks frozen storage, different combination of sucrose, sorbitol and polyphosphate were used. In both one and two steps heating process, the highest gel-forming ability was obtained from combination of 4% sucrose + 4% sorbitol + 0.3% polyphosphate, indicating that above combination of cryoprotective agents during frozen storage is suitable for surimi prepared from Queen fish (*C. lysan*).

Key words: Surimi, gel forming ability, temperature, frozen storage

INTRODUCTION

Surimi is stabilized myofibrillar proteins obtained from mechanically deboned fish flesh that is washed with water and blended with cryoprotectants. For production of thermally irreversible hydrogel (Kamaboko type gels), surimi must be ground with NaCl to solubilize actomyosin (AM), following which inter molecular bonds are established among proteins which are further stabilized by heating to form thermally irreversible gels.

Surimi is an intermediate product used in a variety of products, ranging from the traditional kamaboko products of Japan to surimi seafoods. Before 1960, surimi was manufactured and used within a few days as a refrigerated raw material because freezing commonly deteriorated muscle proteins and induced protein denaturation, which resulted in poor functionality. However, with the discovery of cryoprotectants, the surimi industry was able to tap into previously unexploitable resources.

Nishiya *et al.*[1] at the Hokkaido Fisheries Research Station in Japan, discovered a technique that prevented freeze denaturation of proteins in Alaska pollack (*Theregra chalcogramma*) muscle. Addition of low-molecular-weight carbohydrates, such as sucrose and sorbitol are required in the myofibrillar proteins before freezing to prevent the denaturation of muscle proteins.

Cryoprotectants minimize protein denaturation during frozen storage. Sucrose (4%) and sorbitol (4-5%) are commonly used along with 0.3% sodium phosphate. Several studies have been conducted to investigate the changes of protein functionality during frozen storage by using different cryoprotectants[2-4]. The use of cryoprotectants to extend the shelf life of leached fish minces (i.e. the development of surimi) has been an important technological development in the seafood industry. Surimi has allowed the commercial exploitation from underutilized fish species. At present, the most commonly used cryoprotectants for cold-water species are sucrose and sorbitol, typically added in a blend of 4:4% with 0.2% phosphates, though the level of addition varies depending on species. In addition, sorbitol is often substituted for all or portion of the sucrose because of its lesser sweetness. These cryoprotectants have been chosen for their effectiveness, low cost, availability and low tendency to cause Maillard browning[5]. There is, however, interest to identify alternative cryoprotectants for specific applications with improved ability to stabilize myofibrillar proteins, as well as reduced sweetness. At present, very little is known on the potential of different cryoprotectants for protecting denaturation of myofibrillar proteins under various storage conditions.

Queen fish (*Chorinemus lysan*) is abundantly available round the year in Bangladesh marine water but it has limited value in the fresh fish market and not effectively utilized for human consumption. This species can be an attractive source of raw materials for production of surimi. Huge amounts of Queen fish (*Chorinemus lysan*) caught during peak season in inshore

Corresponding Author: Dr. Md. Ismail Hossain, Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
Tel: 880-91-55695/2201 Fax: 880-91-55810

and deep sea. To produce surimi from this species it is necessary to know the effect of cryoprotective agents and salt concentration on the gel-forming ability. The present study reports the effectiveness of cryoprotective agents independently or combination on the quality of frozen stored surimi of Queen fish (*C. lysan)*.

MATERIALS AND METHODS

Raw materials: Fresh queen fish (*Chorinemus lysan*) was collected from Cox's Bazar BFDC landing center, Bangladesh in several lots from July 2001 to December 2001. The fishes were transported in the laboratory of the Fisheries Technology, Bangladesh Agricultural University, Mymensingh in iced condition.

Preparation of meat paste: The fishes were washed with chilled fresh water to remove blood and slime. The dorsal and lateral muscles of fish were excised as filleted form. The muscles were deboned by mechanical mincer. The mince was washed using chilled fresh water containing 0.1% common salt (NaCl) two times. The meat was stirred in 4-volume solution of its weight for 5 min and then allowed to stand for 10 min before dewatering. The water was removed through bag made of cotton cloth at the pressure of 5 kg cm^{-2} for 10 min and finally at 10 kg cm^{-2} for 15 min. All the operations were done in cold condition. To observe the effect of cryoprotectants, washed fish mince was mixed with various combinations of cryoprotective agents (sucrose, sorbitol and polyphosphate) during blending and was stored at -40°C for 3 weeks. To observe the effect of salt concentration, the surimi was ground with 0, 1, 2, 3, 4, 5 and 6% NaCl and 20% iced water by a mortar for 20 min.

Preparation of gel: The fish paste was stuffed into the polyethylene tube. The tube was heated at 40, 50, 60°C for 2 h (1-step) prior to heating at 80°C for 30 min (2-step) in the water bath. Then the gels were cooled immediately in ice water and were stored at 4°C until assessment of gel properties.

Measurement of gel strength: The gel-strength of the products was assessed by organoleptic methods. A five-person panel as described by Poon *et al.*[6] was constituted for the organoleptic assessment. The gel was removed from the tube and subjected to puncture test, folding test and teeth cutting test for physical measurement of the gel.

Puncture test: Puncture test was done by removing the gels from the tube and cut into equal pieces of 2 cm. The breaking force of the gel was measured against the penetration of a ball type spherical plunger (6 mm diameter) on the pan of an electronic balance. The force in gram required to break the gel by the plunger was recorded from the balance display window.

Table 1: Grade used in the folding test of the gel

Grade	Results on folding
AA	No crack visible when disc is folded into quarter
A	No crack when disc is folded into half but one or more cracks or breaks are visible when folded into quarter
B	One or more cracks are visible when disc is folded into half
C	Breaks, but does not split into halves
D	Splits into halves when folded into half
O	Sample to soft to evaluate

Table 2: Score used in the teeth-cutting test of the gel

Scores	Characteristics of the gel
0-1	Paste or mud like gel
2-3	Very frail gel
4-5	Frail
6	Medium gel strength
7-8	Strong gel strength
9-10	Very strong gel

Folding test: For folding test a spherical disc of 1 mm thick gel was cut off and placed on the index and middle finger of the right hand, the disc was folded first into halves and then quarter with the help of thumb and index finger. The gel was graded using scores presented in the following (Table 1) as suggested by Poon *et al.*[6].

Teeth cutting test: The disc gel of same size used in folding test was supplied to the panelist to recognize the test by cutting it through incisor for teeth cutting test. Gel-strength was evaluated by the following numerical scores as suggested by Shimizu *et al.*[7], which are presented in Table 2.

RESULTS AND DISCUSSION

Effect of salt concentration on gel-forming ability of surimi prepared from Queen fish (*Chorinemus lysan*): To find out the optimum temperature for gel-forming ability of Queen fish (*C. lysan*), both washed and unwashed fish paste in the polyethylene tube were heated in water bath at various temperatures of 40°, 50° and 60°C for 2 h (Fig. 1). The highest gel-forming ability of both washed and unwashed gels was found at 50°C. Poon *et al.*[6] reported that the product prepared from demersal fish heated for 20 min at 50°C in single-step heating treatment had the highest gel-strength. To observe the effect of salt concentration on the gel forming of surimi prepared from Queen fish (*C. lysan*) different amount of NaCl (0, 1, 2, 3, 4, 5 and 6%) was added to the surimi and both one and two-steps heating process were applied to investigate the effect of salt concentration. The maximum gel strength was obtained from surimi at 3% salt concentration in both heating process. The highest gel strength of 829 g in two-step and 746 g from one-step was

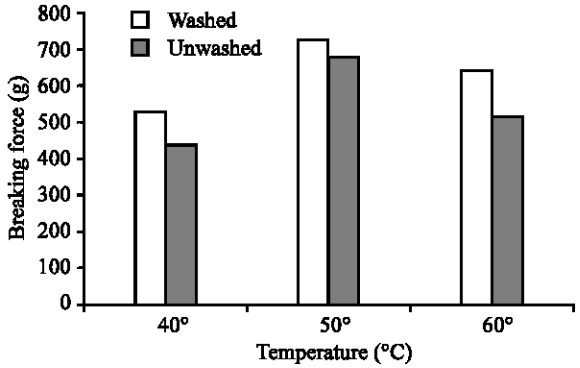

Fig. 1: Change in gel-forming ability of washed and unwashed meat of Queen fish (*C. lysan*) at different heating temperature

Table 3: Effect of salt concentration on gel-forming ability of surimi prepared from Queen fish (*C. lysan*) influenced by one-step heating process

	Salt concentration (%)						
	0	1	2	3	4	5	6
Breaking force (g)	412	557	564	746	640	559	506
Teeth cutting test	6	7	8	8	8	7	7
Folding test	A	AA	AA	AA	AA	AA	AA

Table 4: Effect of salt concentration on gel-forming ability of surimi prepared from Queen fish (*C. lysan*) influenced by two-steps heating process

	Salt concentration (%)						
	0	1	2	3	4	5	6
Breaking force (g)	353	579	725	829	697	657	524
Teeth cutting test	4	7	8	8	8	8	7
Folding test	B	A	AA	AA	AA	AA	A

obtained from gel treated with 3% salt concentration (Table 3 and 4). Further increase of salt concentration decreased the gel strength. Several studies have suggested that 1.7-3.5% NaCl is required for surimi to form adequate gel[8,9]. Schwarz and Lee[10] reported for walleye pollack that gel strength gradually decreased as a salting out effect occurred on further increases in salt concentration. The salt concentration required for gel formation of Kamaboko or surimi like materials ranges from 2 - 3% of the weight of surimi[11] depending on the process and saltiness requirements. The highest TCT was 8 for both one-step and two-step heating process and the lowest were 4 for two-steps and 6 for one-step heating process. The highest FT scores was AA grade for both one-step and two-steps and the lowest was B grade for two-steps and A for one-step heating process. The result of the present study indicates that the addition of 3% salt in the gel is optimum concentration for obtaining highest gel-forming ability of surimi prepared from Queen fish (*C. lysan*). The result obtained from the present study is also in agreement with Lanier *et al.*[12] who reported that 2.5-3.0% NaCl produced an optimum gelling effect in terms of gel strength compliance. Guillen *et al.*[13] observed that in case of gel made from minced flesh of sardine (*Sardina pikhardus*) the highest values for gel-strength, breaking force and breaking deformation were obtained from surimi with 2.5% salt content, which produced a more homogenous gel matrix.

Effect of cryoprotectants on gel-forming ability of surimi prepared from Queen fish (*C. lysan*) during frozen storage: Different combination of sucrose, sorbitol and polyphosphate were used in surimi as cryoprotective agents. The results of gel-forming ability of surimi treated by cryoprotective agents are shown in Table 5 and 6. In both one-step and two-steps heating process, the highest gel-forming ability was found in the surimi blended with 4% sucrose, 4% sorbitol and 0.3% polyphosphate after 1 week of frozen storage. The gel strength of surimi decreased gradually at the end of 3 weeks frozen storage. The highest breaking force of 988 and 1086 g was obtained from one-step and two-steps heating process, respectively. The highest TCT score of 8 was obtained from 1 week frozen stored surimi. The score, which was more or less remained unchanged during 3 weeks frozen storage for both the one-step and two-steps heating process. The highest folding test grade AA also found with the gel treated with 4% sucrose, 4% sorbitol and 0.3% polyphosphate in both one-step and two-steps heating process. The result indicates that these combinations of cryoprotectants suitable for surimi prepared from Queen fish (*C. lysan*). Ryu *et al.*[14] obtained the best cryoprotective effect from sucrose/sorbitol 1:1 (w/w) mixture at 8% with 0.2% sodium tripolyphosphate. Poor gel-forming ability was obtained from surimi without adding any cryoprotectant in both one-step and two-steps heating process. The initial breaking force was 829 g in one-step and 902 g in two-steps, which decreased very sharply from 829 to 523 g in one-step and 902 to 589 g in two-steps during 3 weeks frozen storage (Table 5 and 6). This result suggested that a severe denaturation occurred in the myofibrillar protein during frozen storage. Several studies reported that the myofibrillar proteins of fish muscle aggregated into high molecular weight polymers during frozen storage[15,19]. During frozen storage, several changes occur in fish muscle proteins. These include denaturation, ice crystallization, dehydration and changes in intermolecular conformation, such as salt-soluble protein, pH, ionic-strength[17]. Many proteins have exhibited instability as measured by the partial loss of functionality at subfreezing temperatures. The deterioration of proteins during frozen storage as reflected by their sharp decrease in gel forming ability, water-holding capacity and fat-emulsifying capacity[18,19].

Table 5: Effect of cryoprotectants on gel-forming ability of surimi prepared from Queenfish (*C. lysan*) influenced by 1st-step heating process during frozen storage

		Cryoprotectants combination					
	Storage period (week)	Sucrose-0% Sorbitol-0% P. phosphate-0%	Sucrose-8% Sorbitol-0% P. phosphate-0.3%	Sucrose-0% Sorbitol-8% P. phosphate-0.3%	Sucrose-4% Sorbitol-4% P. phosphate-0.3%	Sucrose-4% Sorbitol-4% P. phosphate-0%	Sucrose-0% Sorbitol-0% P. phosphate-0.3%
Breaking force (g)	1	829	973	926	988	951	893
	2	646	927	887	966	863	669
	3	523	923	839	946	834	532
Teeth cutting test	1	7	8	8	8	8	8
	2	7	8	8	8	8	8
	3	6	8	8	8	8	7
Folding test	1	A	AA	AA	AA	AA	AA
	2	A	AA	AA	AA	A	AA
	3	C	AA	AA	AA	AA	A

Table 6: Effect of cryoprotectants on gel-forming ability of surimi prepared from Queen fish (*C. lysan*) influenced by 2nd-step heating process during frozen storage

		Cryoprotectants combination					
	Storage period (week)	Sucrose-0% Sorbitol-0% P. phosphate-0%	Sucrose-8% Sorbitol-0% P. phosphate-0.3%	Sucrose-0% Sorbitol-8% P. phosphate-0.3%	Sucrose-4% Sorbitol-4% P. phosphate-0.3%	Sucrose-4% Sorbitol-4% P. phosphate-0%	Sucrose-0% Sorbitol-0% P. phosphate-0.3%
Breaking force (g)	1	902	1072	1017	1086	1011	975
	2	776	1061	995	1071	994	720
	3	589	1056	933	1066	915	621
Teeth cutting test	1	8	8	8	8	8	8
	2	8	8	8	8	8	8
	3	7	8	8	8	8	7
Folding test	1	A	AA	AA	AA	AA	AA
	2	A	AA	AA	AA	AA	AA
	3	B	AA	AA	AA	AA	A

The TCT scores decreased from 7 to 6 in one-step and from 8 to 7 in two-steps at the end of 3 weeks of storage period. The FT grade also decreased from A to C in one-step and from A to B in two-steps heating process during frozen storage. The above results suggest that cryoprotectants are needed to prevent the denaturation of muscle protein during frozen storage. Studies conducted with surimi treated with 0.3% polyphosphate showed more or less similar results with that obtained from surimi without cryoprotectant. Breaking forces of 893 g in one-step and 975 g in two-steps heating process were obtained from surimi treated with 0.3% polyphosphate which sharply declined with the lapse of 3 week frozen storage. Surimi treated with 4% sucrose, 4% sorbitol and 0.3% polyphosphate showed better gel-forming ability than surimi treated with 4% sucrose, 4% sorbitol and 0% polyphosphate (Table 5 and 6). These results indicate that 0.3% polyphosphate can be used only as synergists to the cryoprotective effects of carbohydrate additives. Park and Morrissey[5] also reported that a mixture (1:1) of sodium tripolyphosphate and tetrasodium pyrophosphate at 0.2-0.3% is commonly used as a synergist to the cryoprotective effects of carbohydrate additives. In order to elucidate the effect of cryoprotectants on the gel-forming ability, studies were conducted with surimi treated with 8% sucrose and 0.3% polyphosphate and, 8% sorbitol and 0.3% polyphosphate. The results showed that surimi with 8% sucrose showed better result than surimi treated with 8% sorbitol. The highest breaking force of 973 g in one-step heating process and 1072 g in two-steps heating process were obtained from the gel treated with a combination of 8% sucrose and 0.3% polyphosphate. On the other hand maximum breaking force of 926 g in one-step and 1017 g in two-steps heating process was obtained from the gel treated with the combination 8% sorbitol and 0.3% polyphosphate. This result is in agreement with Ryu *et al.*[14] who reported sucrose as the most effective cryoprotectants.

From the present study, it may be concluded that good quality surimi can be successfully prepared from frozen stored Queen fish (*Chorinemus lysan*) using 4% sucrose, 4% sorbitol and 0.3% polyphosphate. The gel prepared from queen fish surimi is temperature dependent and shows better performance with 3% salt in two-step heating process.

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