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ORIGINAL ARTICLE

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Effect of polydextrose on physicochemical properties of threadfin bream (*Nemipterus* spp) surimi during frozen storage

Rodiana Nopianti · Nurul Huda · Noryati Ismail · Fazilah Ariffin · Azhar Mat Easa

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Abstract Physicochemical properties of threadfin bream surimi with different levels of polydextrose (3%, 6%, 9% and 12%), raw surimi, raw surimi with addition sodium tripolyphosphate and commercial surimi (sucrose) during 6 months of frozen storage were investigated. The analyses included the measurement of Ca^{2+}-ATPase, sulfhydryl contents, protein solubility, sodium dodecyl sulfate polyacrylamide gel electrophoresis, differential scanning calorimetry and scanning electron microscopy. The Ca^{2+}-ATPase, sulfhydryl content and protein solubility levels added with 3%, 6%, 9% and 12% polydextrose can be maintained until the 6 months of storage by 47.33%, 41.60% and 51.41%, respectively. Differential scanning calorimetry showed decreases in thermal stabilization of myosin with regard to transition termperature. Analysis by scanning electron microscopy demonstrated that the number of pores formed was increased after storage. This study suggested that surimi stored with the polydextrose as a cryoprotectant was able to maintain physicochemical of surimi better compared to raw surimi with no additives or raw surimi with sodium tripolyphosphate.

Keywords Cryoprotectant · Myofibrillar protein · Physicochemical properties · Polydextrose · Surimi

R. Nopianti · N. Huda (✉) · N. Ismail · F. Ariffin · A. M. Easa
Fish and Meat Processing Laboratory, Food Technology Programme, School of Industrial Technology, Universiti Sains Malaysia,
Minden 11800 Penang, Malaysia
e-mail: nrlhd@usm.my

R. Nopianti
Fish Product Technology Departement, Sriwijaya University, South Sumatera, Indonesia

Introduction

Surimi is stabilized with myofibrillar proteins and blended with cryoprotectant for long periods of frozen storage. Surimi is used as an intermediate foodstuff with long shelf-life and high potential for various texturized product (Pan et al. 2010). At 2006 survey found that a total of 80 surimi processing plants are located in the region; including 26 in Thailand, 15 in Vietnam, 3 in Myanmar, 8 in Indonesia and 15 in Malaysia (SEAFDEC 2009). In particular, myofibrillar protein is the main factor affecting meat quality deterioration during frozen storage (Zayas 1997). If myofibrillar proteins in surimi denatured, it will have an impact on the quality of surimi-based products produced. The denaturation of myosin can lead to decrease of surimi gel-forming ability (Pan et al. 2010). Zayas (1997) suggested that the degrees of protein denaturation and insolubilization during the freezing process and frozen storage are influenced by many factors, such as pretreatment before freezing, the degree of the autolytic process before freezing, the freezing rate, the freezing temperature, the storage temperature and time, as well as storage condition stability including temperature, thawing methods and thawing conditions.

Cryoprotectant protects myofibrillar proteins during frozen storage because it inhibits the denaturation of myofibrillar proteins that usually occurs during frozen storage. Surimi manufacturers commonly used cryoprotectant is the 1:1 incorporation of sucrose and sorbitol with polyphosphate at 0.3%, but they impart a taste to the meat that is very sweet. Low sweetness sugar can be used as an alternative to overcome this problem and is also beneficial due to its low energy content. Many studies have been carried out investigating low sweetness sugars, such as

lactitol (Sultanbawa and Li-Chan 1998; Sych et al. 1991a), litesse (Sultanbawa and Li-Chan 1998), trehalose (Zhou et al. 2006; Osako et al. 2005), palatinit and polydextrose (Sych et al. 1990, 1991b; Herrera and Mackie 2004).

Polydextrose is an odorless, white- to light cream-colored amorphous powder that has virtually no sweetness and an energy value of only 1 kcal/g. Polydextrose has a high average molecular weight, and it therefore has favorable effect on the freezing point depression compared with smaller molecules, such as sucrose or sorbitol (Roller and Jones 1996). The benefit effect is applied to frozen desserts because polydextrose has fat-sparing properties when used in frozen dessert. Solution polydextrose has higher viscocities than sucrose or sorbitol at equivalent concentrations. This higher viscocity contributes to desirable mountfeel (Marshall et al. 2003). Polydextrose has also shown some potential as a cryoprotectant in fish and beef surimi. In these products, polydextrose has some advantages over the traditionally used cryoprotectants, such as sorbitol because it does not add any unwanted sweet flavors (Roller and Jones 1996).

Herrera and Mackie (2004) and Sych et al. (1990) reported that polydextrose performed excellent cryoprotective effect in the cryoprotection of actomyosin on rainbow trout (*Onchorhynchus mykiss*) and Cod (*Gadus morhua*), respectively. To date, polydextrose cryoprotection of surimi is only applied to cold water fish, and less study regarding the physicochemical properties of surimi produced from tropical fish with polydextrose as a cryoprotectant has been reported. Therefore, the objective of this study was to determine the effect of different levels of polydextrose on the physicochemical properties of threadfin bream surimi during frozen storage.

Materials and methods

Surimi preparation

Threadfin bream (*Nemipterus* spp) surimi preparation was done at QL Food Sdn. Bhd. (Hutan Melintang, Perak). Different levels of polydextrose (3%, 6%, 9% and 12%) and 0.3% sodium tripolyphosphate were added to surimi. Raw surimi (washed minced fish), raw surimi with 0.3% sodium tripolyphosphate (STPP) and commercial surimi were used as controls. The samples were identified as follows:

A. 3% polydextrose+0.3% STPP
B. 6% polydextrose+0.3% STPP
C. 9% polydextrose+0.3% STPP
D. 12% polydextrose+0.3% STPP (commercial surimi)
E. Raw surimi (washed minced fish)
F. Raw surimi+0.3% STPP
G. 6% sucrose+0.3% STPP

Storage study

Surimi samples were transported to the Universiti Sains Malaysia by a refrigerated truck. Surimi samples were stored in a freezer at −18 °C during the storage study. During the storage study, the following analyses were conducted: Ca^{2+}-ATPase activity, sulfhydryl content (SH contents), salt extractable protein (SEP), sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE), scanning electron microscopy (SEM) and differential scanning calorimetry (DSC).

Preparation of actomyosin

The following analyses and techniques required actomyosin: Ca^{2+}-ATPase, sulfhydryl content, protein solubility and SDS-PAGE. Actomyosin was prepared according to the method of Benjakul et al. (1997). Surimi (3 g) was homogenized in 30 ml of chilled 0.6 M KCl (pH 7.0) for 4 min with an IKA®T25 digital ultra-turrax homogenizer, and the container with the sample was placed on ice. Each 20 s of homogenization was followed by a 20 s rest interval to avoid overheating during extraction. The homogenate was centrifuged at 8,370 g for 30 min at 4 °C in a Kubota 6500 centrifuge. Three volumes of chilled distilled water were added to precipitate actomyosin. Actomyosin was collected by centrifugation at 8,370 g for 20 min at 4 °C, and the pellet was dissolved by stirring in an equal volume of chilled 0.6 M KCl (pH 7.0). Undissolved debris was removed by centrifugation at 8,370 g for 30 min at 4 °C. Actomyosin was kept on ice during all analyses.

Ca^{2+}-ATPase

The Ca^{2+}-ATPase measurement was described by Wang et al. (1997), in which 0.2 ml actomyosin was diluted in a reaction solution (7.6 mM ATP; 15 mM $CaCl_2$; 150 mM KCl; and 180 mM Tris-HCl, pH 7.4). The reaction was carried out at 25 °C for 10 min, and 1.0 mL of 10% trichloroacetic acid was added to stop the reaction. The mixture was subsequently centrifuged at 2,500 g for 5 min, and 1.0 mL of the supernatant was reacted with 3.0 mL of 0.66% ammonium molybdate in 0.75 N sulfuric acid. A 0.5-mL aliquot of freshly prepared 10% $FeSO_4$ in 0.15 N sulfuric acid was then added, and the mixture was allowed to react for 2 min for color development. The absorbance was read at 700 nm to determine the ATPase activity. NaH_2PO_4 was used to prepare the standard curve for phosphate calculation.

Sulfhydryl content (SH)

The procedure to measure the sulfhydryl content was previously described by Wang et al. (1997). A 0.5 mL

aliquot of actomyosin was mixed with 1.0 mL of a urea and sodium dodecyl sulfate (SDS) solution (8.0 M urea, 3% SDS and 100 mM phosphate buffer with a final pH of 7.4). The titration of sulfhydryl was initiated by the addition of 0.5 mL of 5, 5′-dithio-bis (2-nitrobenzoic-acid) (DTNB) reagent (10 mM DTNB in 0.1 M phosphate buffer with a final pH of 7.4). A sample blank was run with 0.5 mL of phosphate buffer without DTNB. A reagent blank was run with only water. The absorbance was read at 420 nm after 15 min at room temperature. The concentration of sulfhydryl was calculated using a molar extinction coefficient of 11,400 $M^{-1}cm^{-1}$.

Protein solubility

Protein solubility (actomyosin) was determined using the Kjeldahl method according to AOAC (2000). Briefly, the sample was digested, distilled and titrated using 0.02 N HCl. Protein levels were calculated using a conversion factor of 6.25.

Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE)

The SDS-PAGE procedure followed the instructions provided with the wealtec V-Ges vertical gel electrophoresis version 1.0 manufacturer and the method previously described by Laemmli (1970). A 12% resolving gel and a 5% stacking gel were used. The sample buffer consisted 1.5 M Tris-HCl pH 6.8, 2 M Dithiothreitol (DTT), SDS, bromophenol blue, glycerol in deionized water. The sample preparation was conducted by mixing protein samples with loading buffer in a ratio 1:1 and heating the mixture to 100 °C for 5 min to denature the protein samples. An SDS-PAGE broad range molucelar weight standard (6.5 to 200 kDa) (Bio-Rad, Alfred Nobel Drive, Hercules, CA, U.S.A) was used. The running buffer consisted of 12 mM Tris base, 192 mM glycine and 0.1% SDS (w/v) in deionized water. The voltage that was used for pre-running the gel was 80 V for 20 min and 120 V for 80 min to run the gel. Gels were then immersed in deionized water for 5 min and stained with a coomassie blue stain buffer for 20 min, and the gels were destained with a destaining buffer for 1 min. The gels were then immersed in 1% acetic acid and incubated for 1 h on a shaker. The acetic acid was replenished every hour until the gel was completely destained, and the gels were visualized using an UV Tec gel imaging system (Cambridge, UK) that used UVI band version 12.11 software (serial number 08.630445).

Scanning electron microscopy (SEM)

The microstructure of surimi gels was determined with SEM. The SEM procedure has been previously described by Rawdkuen and Benjakul (2008). Briefly, the surimi gels were prepared by cutting them into slices with a 2 mm to 3 mm thickness. The slices were then fixed with 2.5% (v/v) glutaraldehyde in 0.2 M phosphate buffer (pH 7.2). The samples were rinsed for 1 h in distilled water. The samples were then dehydrated with the following serial concentrations in ethanol: 50%, 70%, 80%, 90% and 100% (v/v). The dried samples were cut (0.5 cm × 0.5 cm) and put on the aluminum stub using carbon tape, and the samples were coated with gold for approximately 60 s. The specimens were observed with a SEM (LEO supra 50VP field emission SEM Carl-Ziess SMT; Oberkochen, Germany).

Differential scanning calorimetry (DSC)

The changes in thermal stability of actin and myosin of threadfin bream surimi were monitored with DSC as previously described (Nasab et al. 2005). Briefly, freeze-dried samples were subjected to thermal analysis using a differential scanning calorimeter Q100 equipped with a DSC refrigerated cooling system and monitored by universal analysis 2000 software (ver.3.7A). Nitrogen was used as the purge gas (50 mL/min) for all scans. The heat flow and melting points were calibrated from the melting endotherm of indium. The heating rate used for calibration was the same rate that was used for scanning samples to be analyzed. Samples were weighed (2 mg to 3 mg) into aluminum pans with the addition of phosphate buffer (pH 7.4) until the total weight of 13 mg to 15 mg was reached. The pan was sealed hermetically and placed on the calorimetric cell of Q100. The cell was allowed to equilibrate at 25 °C and was then heated to 100 °C at the rate of 5 °C/min.

Statistical analysis

Data were subjected to analysis of variance (ANOVA). Comparison of means was carried out by Duncan's test was used to determine the significant difference between treatments at $P<(0.05)$ (Cochran and Cox 1992).

Results and discussion

Changes in Ca^{2+}-ATPase activity

The level of polydextrose affected the Ca^{2+}-ATPase of actomyosin of threadfin bream surimi during 6 months of frozen storage, and the changes are shown in Fig. 1. Generally, the Ca^{2+}-ATPase activity of all the samples decreased throughout the 6 months of frozen storage. Surimi with polydextrose had higher values and significantly different when compared to the values of raw surimi

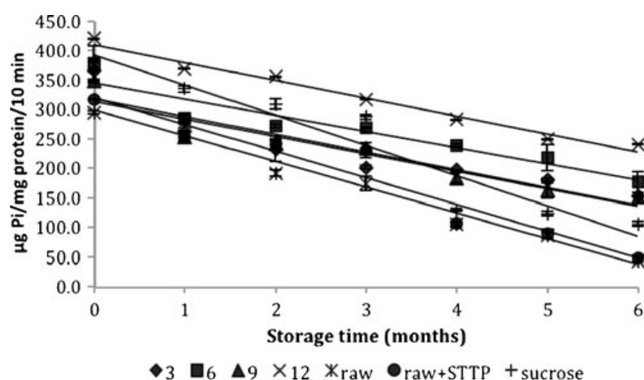

Fig. 1 Changes in Ca^{2+}-ATPase activity of natural actomyosin with different levels of polydextrose during six months of frozen storage. Data are shown as the mean ± SD. Values shown are averages of triplicate analysis on duplicate surimi blocks

and raw surimi with STPP ($P < 0.05$). Surimi that used 12% polydextrose as a cryoprotectant maintained the highest Ca^{2+}-ATPase (421.1 μg Pi/mg proteins/10 min), with a 42.9% decrease until 6 month of frozen storage and significantly different with the other samples ($P<0.05$), and followed by 6%, 9% and 3%. The Ca^{2+}-ATPase values of raw surimi, raw surimi with STPP and commercial surimi were 295.0 μg Pi/mg proteins/10 min, 317.2 μg Pi/mg proteins/10 min and 384.1 μg Pi/mg proteins/10 min, respectively. The results demonstrated that the addition of polydextrose maintained Ca^{2+}-ATPase activity since the freezing process. The percentage decrease of Ca^{2+}-ATPase from surimi that used sucrose as a cryoprotectant was 72.3%. The highest rate of decrease was found in the raw surimi and raw surimi with STPP (decreasing more than 80%). Benjakul et al. (2005) also reported that the Ca^{2+}-ATPase of threadfin bream surimi and surimi made from several other species with sucrose and sorbitol in Thailand also decreased after 6 months of storage, and the decrease was 53.4%.

The globular heads of myosin are responsible for Ca^{2+}-ATPase activity. Changes in actomyosin were reflected by a considerable loss in ATPase activity of actomyosin (Zayas 1997). The great sharp decrease in Ca^{2+}-ATPase activity of raw surimi and surimi with STPP indicated the great denaturation of myosin. Benjakul and Bauer (2000) suggested that the loss of Ca^{2+}-ATPase activity is due to the tertiary structural changes caused by ice crystals and the increase in ionic strength of the system. The rearrangement of proteins via protein-protein interactions was also presumed to contribute to the loss of activity. The higher level of Ca^{2+}-ATPase activity of surimi with polydextrose, indicated that polydextrose can maintain Ca^{2+}-ATPase activity. The conclusion has also been confirmed in study of rainbow trout during storage. Together, these data suggest that 12% polydextrose prevents the decrease in Ca^{2+}-ATPase activity most successfully, followed by 6% polydextrose. This indicates that this compound has cryoprotective effects on the threadfin bream surimi during 6 months of frozen storage. However, the Ca^{2+}-ATPase activity of surimi added with 9% polydextrose was lowered than that of surimi treated with 6% polydextrose even though it was expected to be higher. It was presumed that the myofibrillar protein in surimi treated with 9% polydextrose had undergone certain extent of denaturation before freezing process and frozen storage.

Changes in total sulfhydryl contents (SH contents)

Generally, the sulfhydryl content of all samples decreased during the 6 months of frozen storage (Fig. 2). Surimi stored with polydextrose had higher sulfhydryl contents at the beginning of the storage period (0 month) when compared to the sulfhydryl contents of raw surimi, raw surimi with STPP and commercial surimi after storage ($P<0.05$). Actomyosin from the surimi with polydextrose had the value of 13.34 mol SH/10^5 g protein at the beginning (0 months). Raw surimi, raw surimi with STPP and commercial surimi had the values of 13.01 mol SH/10^5 g protein, 12.51 mol SH/10^5 g protein and 12.64 mol SH/10^5 g protein, respectively. These data suggest that surimi stored with polydextrose used as a cryoprotectant preserved the sulfhydryl content present at the beginning of the freezing process. Among all of the samples, 12% polydextrose had the greatest sulfhydryl content after 6 months of storage and showed a 54.4% decrease in comparison with its initial value. Actomyosin from raw surimi had the lowest sulfhydryl content throughout the storage period of 6 months, being reduced by 83.8% in comparison with its initial value. The sulfhydryl content of sucrose decreased by 51.9% after 6 months of storage.

Many factors affected the sulfhydryl content of surimi. The decrease in sulfhydryl contents was considered to be due to the formation of disulfide bonds through the oxidation of sulfhydryl groups or disulfide interchanges (Hayakawa and Nakai 1985). Benjakul and Bauer (2000)

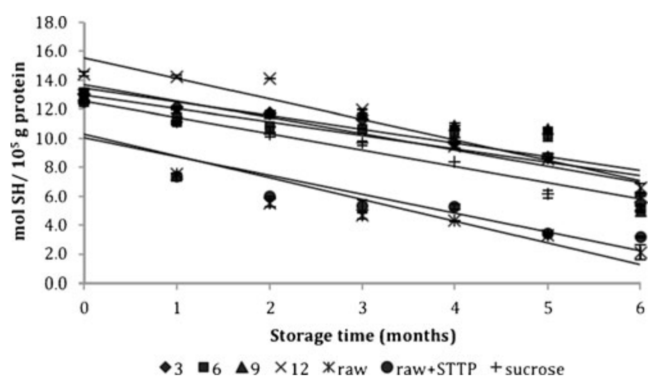

Fig. 2 Changes in sulfhydryl contents of natural actomyosin with different levels of polydextrose during six months of frozen storage. Data are shown as the mean ± SD. Values shown are averages of triplicate analysis on duplicate surimi blocks

reported that the decrease in Ca^{2+}-ATPase in cod muscle protein was in agreement with the decrease in sulfhydryl contents. Duangmal and Taluengphol (2010) reported, SH content on tilapia surimi also decreased as the concentration in the presence of sodium ascorbate. The similar trend was also reported by Zhou et al. (2006) that the sulfhydryl content of tilapia surimi with 8% trehalose and 8% sucrose/sorbitol decreased by after 6 months of storage.

These data demonstrate that surimi with polydextrose as a cryoprotectant maintains sulfhydryl content from the beginning of the frozen storage whereas raw surimi and raw surimi with STPP were unable to maintain sulfhydryl contents. The sharp decrease in sulfhydryl content of raw surimi revealed the denaturation of threadfin bream protein. Furthermore, 12% polydextrose maintains sulfhydryl contents for the 6 months of frozen storage.

Changes in protein solubility

The actomyosin of proteins is soluble only in salt solutions of high ionic strength. The percentage of protein solubility in all of the samples continuously decreased during the 6 months of frozen storage (Fig. 3). Surimi with polydextrose had the highest percentage of protein solubility at the beginning of the storage period (0 month) when compared to the protein solubility percentage found in raw surimi, surimi with STPP and commercial surimi ($P<0.05$). The percentage of protein solubility in surimi with polydextrose was 56.09%, whereas raw surimi, raw surimi with STPP and commercial surimi had protein solubility values of 24.72%, 24.88% and 45.93%, respectively. Polydextrose was able to maintain protein solubility levels from the beginning of the freezing process. The lowest protein solubility levels were observed in raw surimi and raw surimi with STPP with a decrease of 60.8% and 65% after 6 months of storage in comparison to its initial levels, respectively. Surimi with 12% polydextrose had the highest value among the samples but also had the largest loss of protein solubility (55%) in comparison to 3%, 6% and 9% polydextrose. This loss was potentially due to the dilution effect of cryoprotectants on the protein content in the surimi. The smallest loss of protein solubility was with 9% polydextrose (41.8%) after 6 months of frozen storage. Commercial surimi had a 51.8% reduction in protein solubility levels after 6 months of frozen storage. All surimi that used polydextrose as a cryoprotectant had higher protein solubility values when compared to commercial surimi, raw surimi, and raw surimi with STPP. Herrera and Mackie (2004) reported that polydextrose is more effective in maintaining solubility levels than glucose syrup and the mixture of sucrose/sorbitol in the cryoprotection of actomyosin in frozen stored farmed rainbow trout (*Onchorhynchus mykiss*). The use of polydextrose as a cryoprotectant prevented the drastic changes of proteins associated with freezing, which resulted in protein aggregation that occurred more slowly.

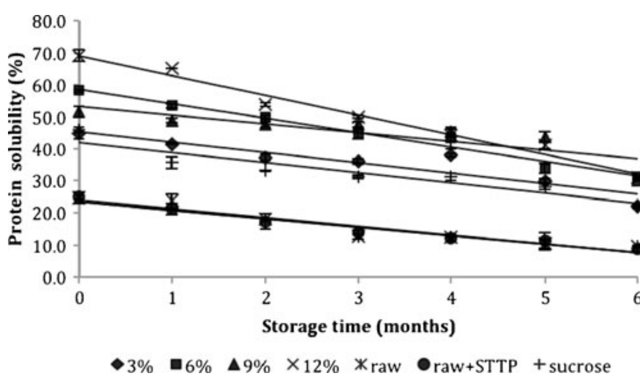

Fig. 3 Changes in protein solubility with different levels of polydextrose during six months of frozen storage. Data are shown as the mean ± SD. Values shown are averages of triplicate analysis on duplicate surimi blocks

The decrease in protein solubility is a primary indicator of protein denaturation during the frozen storage period. Fish muscle proteins are more sensitive to freezing than meat proteins. The insolubilization of myofibrillar proteins was the main factor affecting the functional properties of fish proteins after frozen storage. Protein solubility in fish muscle stored in a frozen state has been used as a criterion for the alterations of proteins. The change of protein solubility during rigor mortis is potentially due to the decrease in pH and the strong association of myosin and actin (Zayas 1997). Together, these data demonstrate that the addition of 6% polydextrose to surimi causes a slower decrease in protein solubility levels when compared to commercial surimi resulting in a good cryoprotective effect on threadfin bream surimi proteins.

Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE)

The protein pattern of surimi with polydextrose at various levels as a cryoprotectant, raw surimi, raw surimi with STPP and commercial surimi are shown in Fig. 4. The myosin band intensity of all the samples decreased after 6 months of frozen storage, and the actin band intensity also decreased throughout the storage period. Surimi that used 6% and 12% polydextrose as cryoprotectants had the highest intensities of myosin and actin according to the three-dimensional image of each band throughout the 6 month storage period. The lowest intensities of actin and myosin were observed in commercial surimi, raw surimi, and raw surimi with STPP after 6 months of storage. The intensities of actin and myosin decreased during frozen storage because denaturation of myofibrillar proteins occurred during the frozen storage. Tseng et al.

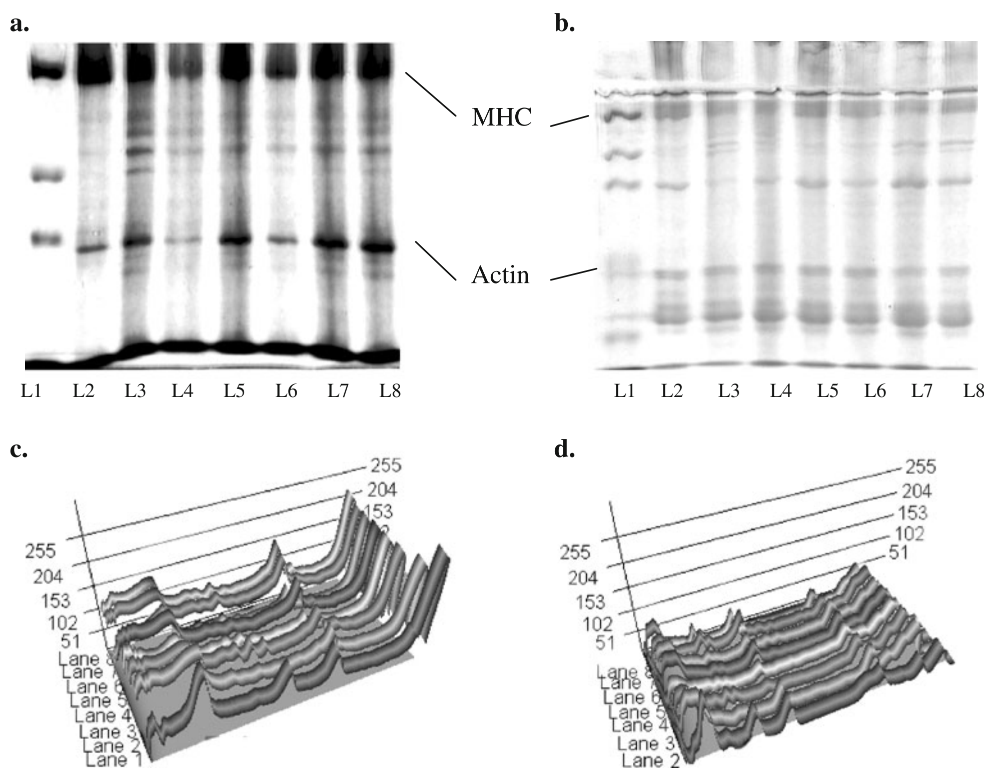

Fig. 4 SDS-PAGE during the six months and a one-dimensional image of treadfin bream surimi on 0 month (**a**, **c**), six month (**b**, **d**). The numbers represent the conditions as follows: L1 = standard, L2 = sucrose/commercial, L3 = raw + STPP, L4 = raw L5 = 12%, L6 = 9%, L7 = 6% and L8 = 3%; MHC, myosin heavy chain

(2002) suggested that the reduction in protein ostensibly resulted from the proteolytic degradation and the formation of insoluble protein aggregates. Together, our data suggest that 6% and 12% polydextrose protects protein from denaturation for until 6 months of frozen storage.

Differential scanning calorimetry (DSC)

The changes in thermal stability of threadfin bream surimi during frozen storage are presented in Tables 1 and 2. Generally, the surimi thermograms contained two endothermic transitions. The first peak was a transition temperature for myosin, and the second peak was a transition temperature for actin (Nasab et al. 2005; Tseng et al. 2002; Sych et al. 1991a, b; Sych et al. 1990). The threadfin bream surimi using 12% polydextrose as a cryoprotectant had a maximal transition temperature for myosin at 45.13 °C and dropped to 43.89 °C after 6 months of storage. The percentage decrease was 2.75% in comparison with its initial value. The highest percentage loss among all of the samples was surimi with STPP (6.94%). However, all of the temperature values in the surimi with 12% polydextrose were higher until 3 months of storage when compared to the other samples. The lowest percentage decrease during the 6 months of storage was 1.6% and occurred in the surimi with 9% polydextrose. The decrease of actin transition temperature was less than the myosin transition temperature, as it remained unchanged for the 6 months of frozen storage. There were no significant differences between cryoprotectant levels of the samples.

Tseng et al. (2002) reported that the transition temperatures for myosin (50.2 °C) and actin (72.6 °C) in red claw muscle proteins had a significant decrease after 14 days of storage at 0 °C, dropping to 39.4 °C and 60.3 °C, respectively. The decreases found in our study were less than this, and this difference was probably due to different raw materials and storage conditions. Tseng et al. (2002) suggested that the loss

Table 1 Temperature (°C) denaturation of threadfin bream surimi myosin at different polydextrose treatment levels during a six month frozen period

Storage (months)	3%	6%	9%	12%	Raw	Raw + STTP	Sucrose
0	43.2^{bA}±1.05	43.3^{abA}±1.36	43.4^{abA}±0.35	45.1^{aA}±0.63	41.3^{cA}±0.01	41.1^{cA}±0.01	44.9^{abA}±0.88
3	42.1^{cdA}±0.09	43.2^{bcA}±0.96	42.5^{cdA}±1.00	45.2^{aA}±0.44	40.3^{eB}±0.06	41.0^{deA}±0.02	44.2^{abA}±0.88
6	42.2^{abA}±0.29	42.5^{abA}±0.13	42.7^{abA}±0.15	43.9^{aA}±2.02	40.2^{bcB}±0.15	38.2^{cA}±2.86	44.6^{aA}±0.01

Mean with different superscript letters within the same row indicate a significant difference ($P<0.05$). Mean with the same capitalized letters in the same column is not significantly different ($P<0.05$). Values shown are averages of triplicate analysis on duplicate surimi blocks

Table 2 Temperature (°C) denaturation of threadfin bream surimi actin at different polydextrose treatment levels during a six month frozen period

Storage (months)	3%	6%	9%	12%	Raw	Raw + STTP	Sucrose
0	$64.0^{aA}\pm0.09$	$63.0^{aA}\pm0.72$	$62.8^{aA}\pm1.66$	$63.8^{aA}\pm0.04$	$64.5^{aA}\pm0.25$	$63.6^{aA}\pm0.19$	$63.9^{aA}\pm0.55$
3	$63.1^{aA}\pm0.57$	$64.8^{aA}\pm0.96$	$64.0^{aA}\pm0.52$	$64.3^{aA}\pm0.32$	$64.3^{aA}\pm0.23$	$63.4^{aA}\pm0.19$	$64.7^{aA}\pm1.66$
6	$65.0^{aA}\pm3.20$	$64.2^{aA}\pm1.30$	$65.3^{aA}\pm3.10$	$64.2^{aA}\pm0.62$	$64.5^{aA}\pm0.20$	$64.1^{aA}\pm1.21$	$62.8^{aA}\pm0.71$

Mean with the same superscript letters in the same row is not significantly different ($P<0.05$). Mean with the same capitalized letters in the same column is not significantly different ($P<0.05$). Values shown are averages of triplicate analysis on duplicate surimi blocks

in thermal stability of proteins results from an interaction with oxidized lipids, including lipid free radicals and secondary products such as reactive aldehydes, and from conformation alterations due to enzyme induced hydrolysis. Together, these results indicate that 9% polydextrose maintains the thermal transition of surimi throughout frozen storage.

Scanning electron microscopy (SEM)

The microstructures of threadfin bream surimi with different levels of polydextrose, raw surimi, raw surimi with STPP and commercial surimi were visualized by SEM, as shown in Fig. 5. In general, there were few pores formed in all of the samples at the beginning of storage (0 month), but the number of pores increased after 6 months of storage. However, the surimi with 12% polydextrose did not have many pores formed and the structure was still compact. The structure in the raw surimi was not compact and was delicately impressed because the pores were larger than the pores found in the other surimi samples. Huda et al. (2011) reported, water holding capacity of surimi that treated with different level of polydextrose decreased during 16 weeks frozen storage. Decreased in water holding capacity, is a consequence of changes in the muscle structure, for

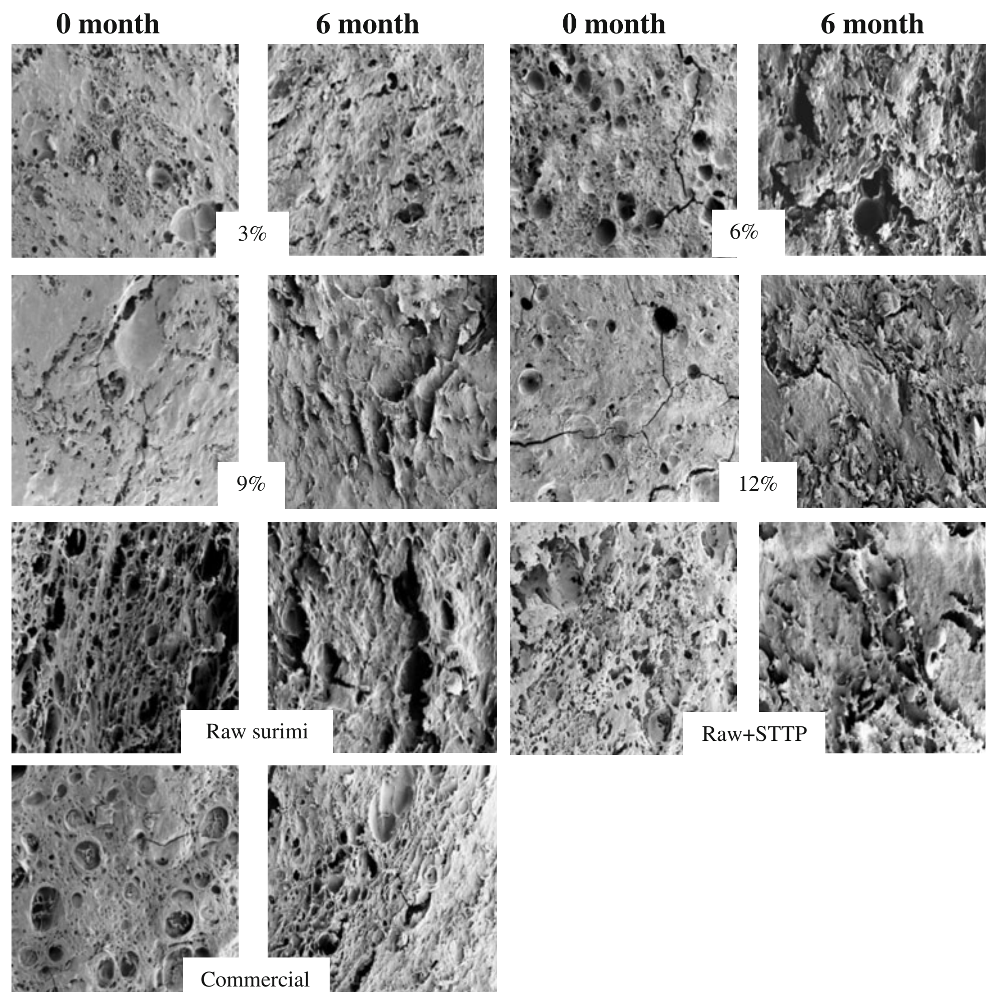

Fig. 5 SEM of threadfin bream surimi with different levels of polydextrose during six months of frozen storage (Magnification 50X)

example pores sized. Benjakul et al. (2005) reported that the decrease in water holding capacity due to formaldehyde was an effective cross linker of protein molecules to form protein aggregates. In addition, polyphosphate affects the microstructures in surimi. Julavittayanukul et al. (2006) reported that the addition of polyphosphate in surimi made from bigeye snapper resulted in the formation of an ordered structure with finer strands. In general, surimi without cryoprotectant had a more poorly organized network that was confirmed with less gel forming ability. Surimi that used 12% polydextrose as a cryoprotectant exhibited a fine and compact structure with smaller pores in the matrix.

Conclusion

Generally, surimi with polydextrose as a cryoprotectant had better physicochemical properties than raw surimi, surimi with STPP and sucrose in maintaining myofibrillar protein from denaturation. Polydextrose can take the place of sucrose and be used as a new kind of non sweet cryoprotectant for surimi. Surimi with 12% or 6% polydextrose with 0.3% STPP as cryoprotectants had the best physicochemical properties when compared to the other surimi samples and an optimal level of polydextrose was recommended to be 6%.

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