



## Biochemical characterisation of Alaska pollock, Pacific whiting, and threadfin bream surimi as affected by comminution conditions

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### ABSTRACT

Salt-soluble protein, surface reactive sulfhydryl content, and surface hydrophobicity of Alaska pollock, Pacific whiting, and threadfin bream surimi were characterised, as affected by various comminution conditions. Chopping time/temperatures were explored in consideration of their habitat temperatures. Salt-soluble protein (SSP) significantly decreased when chopping time was extended. Corresponding to our follow-up study, no relationship between SSP and gel texture was found. Surface hydrophobicity was inversely proportional to SSP concentration, indicating the unfolding of protein upon comminution. Alaska pollock surimi demonstrated aggregation during chopping at 10 and 20 °C, based on the surface hydrophobicity. Surface reactive sulfhydryl (SRSH) contents of the three fish species behaved differently. The SH groups were oxidized to disulphide bonds when higher chopping temperature was applied. As a result, increased SRSH content was not observed in Alaska pollock (10 and 20 °C chopping) and threadfin bream paste (25 and 30 °C chopping).

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### 1. Introduction

Alaska pollock, Pacific whiting, and threadfin bream have been extensively utilized in the surimi industry (Guenneugues & Morrissey, 2005). Alaska pollock (AP) surimi is known as a premium grade product and had an annual production of nearly 250,000 MT during 1998–2003 (Guenneugues et al., 2005); however, the production declined to 100,000–125,000 MT in 2010, approximately (Halhjem, 2011). About 20,000 MT of Pacific whiting (PW) were produced, primarily on and off the northwest coast (Guenneugues et al., 2005). In tropical countries, threadfin bream (TB) is the major species for surimi and its production, only in Thailand, and reached 80,000 MT (Yongsawatdigul, Worratao, & Park, 2002) over a year while 180,000 MT of threadfin bream were caught worldwide (Guenneugues et al., 2005).

Commercial industries have tried various approaches to enhance gel qualities of surimi. Some examples are mixing PW surimi with enzyme inhibitors, such as egg white, or the application of fast cooking technology that can significantly reduce enzyme activity from endogenous proteolytic enzymes (Yongsawatdigul, Park, Kolbe, Abu Dagga, & Morrissey, 1995), as well as addition of other functional ingredients, including exogenous microbial transglutaminase. However, the effect of chopping conditions (time and temperature) has been overlooked. Esturk, Park, and Thawornchinsombut (2004) demonstrated the gel strength of surimi from

various fish species, as affected by different final chopping temperatures. They found that maximum gel strength could be obtained when the final chopping temperature corresponded to the habitat temperature of each fish species. Alaska pollock lives in cold water (0–5 °C) while the temperate fish, Pacific whiting, live in a cool temperature (5–15 °C). Threadfin bream is a tropical fish and lives in warm water (20–30 °C). Douglas-Schwarz and Lee (1988) also studied the impact of final chopping temperature on the gel strength of AP and observed a similar trend. According to our recent study (Poowakanjana, Mayer, & Park, 2012), surimi from temperate and warm water fish demonstrated maximum gel strength when chopped at higher temperature. Gel texture of AP provided maximum gel values when chopped at 0 °C. It was surprising that relatively strong AP gel could also be obtained when final chopping temperature approached 20 °C, but maintaining chopping at this high temperature for a longer time impaired gel texture. This conclusion seems to be in disagreement with Esturk et al. (2004) and Douglas-Schwarz et al. (1988). This was because, in these two studies, chopping was conducted using surimi started at a different temperature with fixed time to obtain a target final chopping temperature. Unlike the study of Poowakanjana et al. (2012), chopping started with partially thawed surimi at –5 °C for all samples. We believed that, using partially thawed sample, at the beginning of the chopping process, made AP fish actomyosin somewhat tolerable to 15–20 °C. This was because salt was added at the beginning when the surimi was still cold. Adding salt to the surimi, when the temperature was above freezing point, might destabilize the myofibrillar protein structure. Poowakanjana et al. (2012) also analysed the change in secondary structure of protein, as affected by various

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chopping conditions, using Raman spectroscopy. Raman spectroscopy revealed a significant change in disulphide linkage and the reduction of secondary structure upon extended chopping.

A number of analyses, to elucidate the effect of various chopping conditions on structural changes of fish actomyosin, were extensively studied, based on biochemical measurement of protein solubility, degree of protein unfolding, and oxidation of sulfhydryl group. Protein solubility in salt solution can reveal the structural changes of protein. Salt solubilizes the actomyosin by binding to the myofibrillar protein (through the electrostatic repulsive force resulting in unwinding protein structure) and increases water binding sites (Rosco & Bledsoe, 2006). Since protein unfolding is a prerequisite for gelation, high salt-soluble protein is believed to well correspond with the strong gel texture. This relationship was somehow uncertain in the case of fish protein-based surimi and will be substantially discussed later. Due to limited studies on the specific comminution conditions relating to SSP, surface reactive sulfhydryl (SRSH) content, and surface hydrophobicity ( $S_0$ ), understanding biochemical characteristics of fish proteins, as affected by comminution conditions, would provide a way to maximize gel values based on the thermal stability of the respective species. The objective of this study was to investigate the effect of comminution variables (both chopping time and temperatures) on biochemical characteristics of AP, PW, and TB surimi paste.

## 2. Materials and methods

### 2.1. Materials

Alaska pollock (AP) (*Theragra chalcogramma*) surimi ("A" grade: approximately 3 frozen months old with 5% sorbital, 4% sucrose, and 0.3% phosphate as cryoprotectants) was obtained from American Seafoods (Seattle, WA, USA). Pacific whiting (PW) (*Merluccius productus*) surimi ("A" grade: approximately 4 frozen months old with 5% sorbital, 4% sucrose, and 0.3% phosphate as cryoprotectants) was obtained from Trident Seafoods (Seattle, WA, USA). Threadfin bream (TB) (*Nemipterus* spp.) surimi ("SA" grade: approximately 2 frozen months old with 6% sucrose and 0.2% sodium polyphosphate as cryoprotectants) was obtained from Mana Frozen Foods, Bangkok, Thailand. Potassium chloride (KCl) was purchased from VWR International (West Chester, PA, USA). Tris-HCl was purchased from J.T. Baker Chemical Company (Phillipsburg, NJ, USA). Bradford reagent was obtained from Bio-Rad Laboratory (Hercules, CA, USA). Ethylenediaminetetraacetic acid (EDTA), 5-5'-dithiobis-(2-nitrobenzoic acid) (DTNB), and anilino-naphthalene-8-sulfonic acid (ANS) were purchased from Sigma Chemical Company (St. Louis, MO, USA).

### 2.2. Preparation of surimi paste

Paste preparation was conducted according to the method of Poowakanjana et al. (2012). Upon arrival, frozen surimi was cut

into smaller blocks (approximately 1000 g), sealed in a vacuum bag and kept at  $-18^{\circ}\text{C}$  for approximately 5 months to complete all experiments. Frozen surimi was partially thawed at room temperature, for 1 h, to allow the core temperature to reach approximately  $-5^{\circ}\text{C}$ . Blocks were then cut into cubes (2 cm) and placed in the chopping bowl. A sample was chopped at 1800 rpm for 1 min, using a silent cutter (UM 5 universal, Stephan Machinery Corp, Columbus, OH, USA). Two percent salt was added and chopping continued at 1800 rpm for 1 min. Moisture content was adjusted to 78%, using ice before continuing to chop at 1800 rpm for 1 min. The chopping bowl was then connected to a vacuum pump (40–60 kPa) and chopping continued at 3600 rpm until total chopping time was achieved for 6, 9, 12, 15, 18, and 21 min, respectively (Table 1). Chopping (AP surimi at 0 and  $10^{\circ}\text{C}$ , and PW surimi at  $5^{\circ}\text{C}$ ) was performed in a walk-in freezer ( $-30^{\circ}\text{C}$ ). Chopping (AP surimi at  $20^{\circ}\text{C}$ , PW surimi at 15 and  $20^{\circ}\text{C}$ , and TB surimi at 15, 25, and  $30^{\circ}\text{C}$ ) was conducted at room temperature ( $20$ – $25^{\circ}\text{C}$ ), using a chopping bowl equipped with circulating coolant ( $-5^{\circ}\text{C}$ ). Chopping was started with the partially thawed surimi at around  $-5^{\circ}\text{C}$ .

Since comminution variables (time and temperature) were dependent on each other, chopping was switched on and off to fulfil two variables while controlling temperature. Until the total chopping time was achieved, if the surimi paste sample reached the target temperature, chopping was halted until the temperature decreased by  $5^{\circ}\text{C}$  from the target temperature. Chopping continued on and off until the accumulated chopping time reached the final target (6, 9, 12, 15, 18, or 21 min). Therefore, total preparation time included total chopping time and the waiting time needed to maintain the final target temperature. Comminution variables are described in Table 1.

In our preliminary study, a linear relationship between chopping time and paste temperature was observed. With the use of a circulating coolant at  $-5^{\circ}\text{C}$ , the temperature rose linearly from  $-5$  to  $30^{\circ}\text{C}$  by continuous chopping at 1800 rpm for 3 min and at 3600 rpm for the next 18 min, resulting in a heating rate of  $1.67^{\circ}\text{C}/\text{min}$ .

### 2.3. Salt-soluble proteins (SSP)

SSP was measured as outlined by Thawornchinsombut and Park (2006) with slight modification. The sample paste was taken immediately from the silent cutter after chopping. Three grammes of paste were homogenized at speed 1 with 27 ml of 0.6 M KCl in 20 mM Tris-HCl buffer (pH 7) for 1 min, using a homogenizer (model GLH-115, PG 700, Fisher Scientific, Pittsburgh, PA, USA). The homogenized samples were then centrifuged at 10,000g (Sorvall RC-5B, Newtown, CT, USA) at  $4^{\circ}\text{C}$  for 30 min. After centrifugation, the supernatant was diluted to approximately 1 mg protein/ml with 0.6 M KCl in 20 mM Tris-HCl buffer (pH 7) before measuring salt-soluble protein. Bradford's dye reagent was diluted 5 times and then diluted reagent (5 ml) was brought to 100  $\mu\text{l}$  of sample solution. The sample was allowed to stand at room temperature

**Table 1**  
Various comminution conditions for Alaska pollock, Pacific whiting, and threadfin bream surimi.

| Sample          | Target Temp, $^{\circ}\text{C}$ | Final Temp ( $^{\circ}\text{C}$ )/Chopping time (min)/Total Preparation time (min) |         |          |          |          |          |
|-----------------|---------------------------------|--|---------|----------|----------|----------|----------|
| Alaska pollock  | 0                               | 0/6/12   | 0/9/25  | 0/12/43  | 0/15/62  | 0/18/82  | 0/21/103 |
|                 | 10                              | 5/6/8  | 10/9/12 | 10/12/25 | 10/15/37 | 10/18/50 | 10/21/64 |
|                 | 20                              | 5/6/8  | 12/9/12 | 18/12/18 | 20/15/31 | 20/18/44 | 20/21/59 |
| Pacific whiting | 5                               | 5/6/8  | 5/9/19  | 5/12/32  | 5/15/44  | 5/18/56  | 5/21/69  |
|                 | 15                              | 5/6/8  | 12/9/12 | 15/12/20 | 15/15/33 | 15/18/47 | 15/21/62 |
|                 | 20                              | 5/6/8  | 12/9/12 | 18/12/16 | 20/15/25 | 20/18/36 | 20/21/48 |
| Threadfin bream | 15                              | 5/6/8  | 12/9/12 | 15/12/20 | 15/15/33 | 15/18/47 | 15/21/62 |
|                 | 25                              | 5/6/8  | 12/9/12 | 18/12/15 | 23/15/20 | 25/18/28 | 25/21/39 |
|                 | 30                              | 5/6/8  | 12/9/12 | 18/12/15 | 23/15/20 | 27/18/24 | 30/21/29 |

Note: Salting time was 1 min less than total preparation time. Vacuum time was 3 min less than total chopping time.

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