PROPERTIES OF POLLOCK (THERAGRA CHALCOGRAMMA) 
SKIN HYDROLYSATES AND EFFECTS ON LIPID OXIDATION OF 
SKINLESS PINK SALMON (ONCORHYNCHUS GORBUSCHA) 
FILLETS DURING 4 MONTHS OF FROZEN STORAGE

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ABSTRACT

Fresh pollock skin was hydrolyzed for 10 min (PPH10), 30 min (PPH30) and 45 min (PPH45), and the chemical and functional properties of pollock skin protein hydrolysates were evaluated. PPH45 (79.6%) had significantly higher nitrogen solubility values than PPH30 (72.6%) and PPH10 (64.8%). Emulsifying stability values for PPH10 and PPH30 were significantly higher (P < 0.05) than PPH45. Fat adsorption values for PPH45 (4.7 mL oil/g protein) were greater (P < 0.05) than PPH10 (3.6 mL oil/g protein) and PPH30 (3.7 mL oil/g protein). Salmon fillets stored frozen for 4 months including those glazed with pollock skin protein hydrolysate, water and glycerin solutions had increased yield and thaw yield values when compared to the nonglazed (NG) control fillets. Thiobarbituric acid (mg malondialdehyde/kg sample) values from fillets coated with a solution containing PPH10 (0.8) and stored frozen for 4 months were significantly lower (P < 0.05) than other glazed and NG fillets.

PRACTICAL APPLICATIONS

Pollock skin is an abundant and underutilized resource that can be used as a unique protein source to make fish protein hydrolysates. Utilizing proteolytic

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enzymes, pollock skin protein hydrolysates (PPHs) can be prepared that have
new and/or improved chemical and functional properties. The natural antioxi-
dant and water-soluble properties and film-forming ability of PPHs make it
ideal for coating material to suppress lipid oxidation in fish fillets during
frozen storage. Edible coatings prepared from pollock skin hydrolysates have
potential applications for enhancing the storage stability and quality of frozen
fillets. The edible hydrolysate coating may also provide some degree of pro-
tection against damage during transportation and handling of fish fillets.

INTRODUCTION

Fish proteins from marine species are good sources of high-quality pro-
teins, and there is an opportunity to use more fish processing by-products
as protein ingredients for use in foods, feeds and industrial applications.
Pollock skin is an abundant and underutilized resource that can be used as a
unique protein source to make hydrolysates. In 2005, the estimated amount
of pollock skin produced from automated fish processing plants in Alaska
was 107,000 metric tons. Utilizing different proteolytic enzymes, pollock skin
protein hydrolysates (PPHs) can be prepared having different chemical and
functional properties. Previous studies have shown that PPHs have antioxidant
and other functional properties (Kim et al. 2001). Sathivel et al. (2003)
reported that protein powder hydrolysates from herring were good sources
of high-quality fish protein with many desirable functional and antioxidant
properties.

Raw fish is extremely perishable, and therefore, marketing often focused
on frozen and processed products (Bligh 1979). Freezing is a common pres-
ervation method used to control or decrease biochemical changes in fish that
occur during frozen storage. A chemical reaction such as lipid oxidation is not
completely inhibited during frozen storage, which can lead to quality deterio-
ration of fish. Preservatives such as phosphates are often used to improve the
shelf life, increase water-binding capacity, reduce oxidation and affect other
properties in seafood and meat products. Shahidi (1994) reported that phos-
phates are used in seafood to enhance water-holding capacity and to improve
cooking yield.

Butylated hydroxyanisole and butylated hydroxytoluene have been com-
monly used as antioxidants by the food industry to increase product shelf life
and quality. However, there is great consumers’ interest in natural ingredients
that enhance food quality and shelf life. Biodegradable natural ingredients
including polysaccharides and proteins can be used as fish and meat coatings
to control quality deterioration during frozen storage. Stuchell and Krochta
(1995) reported that king salmon coated with whey protein isolates delayed
lipid oxidation during frozen storage. Wanstedt et al. (1981) demonstrated that precooked frozen pork patties coated with alginate had better sensory qualities than control patties after being stored frozen. Jeon et al. (2002) reported that chitosan-coated Atlantic cod and herring had reduced moisture loss and lipid oxidation during storage, and Sathivel (2005) reported that chitosan and fish protein coatings affected increased yield, and reduced moisture loss and lipid oxidation of pink salmon fillets during frozen storage.

PPHs contain natural antioxidants and have the ability to form films, and these properties make them candidates for use as coating material to suppress lipid oxidation in fish fillets during frozen storage (Kim et al. 2001). However, there is a lack of information on the application of effects of using hydrolysate coatings to enhance the quality of fish fillets during frozen storage. The objectives of this study were to determine the chemical and functional properties of PPHs, and to evaluate the effectiveness of coatings made with PPH on quality of skinless pink salmon fillets during frozen storage.

**MATERIALS AND METHODS**

**Preparation of Fish Protein Hydrolysates**

Fresh pollock skin was obtained from a commercial fish processing plant in Kodiak, AK; vacuum packaged; and stored at −40°C until further processed. The skin was thawed overnight at 4°C and ground in a Hobart grinder (K5SS, Hobart Corporation, Troy, OH) through a 7-cm-diameter plate having 12-mm-diameter openings, and subsequently ground through a plate with 6-mm-diameter openings. Hydrolysis conditions were similar to those documented by Hoyle and Merritt (1994) and Liceaga-Gesualdo and Li-Chan (1999). A 500 g portion of ground pollock skin was mixed with an equal volume of distilled water (DW) and homogenized in a Waring blender (New Hartford, CT) for about 2 min. The mixture was stirred at 50°C for optimal Alcalase activity (Novo Nordisk Technical Bulletin 1995). Alcalase (>0.24 U/g) (Novo Nordisk, Fralklinton, NC) was added to the mince protein at 0.5% w/w. The mixture was continuously stirred for 10, 30 and 45 min at 50°C, and the enzyme was inactivated at the end of each hydrolysis time by increasing the temperature to 85°C for 15 min. The soluble fraction of hydrolysate was recovered by centrifugation at 2,560 × g for 15 min, and the soluble aqueous fraction was decanted, freeze-dried, sealed in vacuum bags and stored at 4°C until used. The experiment was replicated three times.

**Degree of Hydrolysis (DH) Time Course**

Three separate experiments were performed to determine the effect of hydrolysis time on DH using the method of Hoyle and Merritt (1994). An
equal volume of water was added to 50 g portions of skin mince, and the mixture was brought to 50°C. The Alcalase enzyme was added to the fish mince at 0.5% w/w. At the end of each hydrolysis time of 0, 15, 30, 45, 60 and/or 75 min, an aliquot (50 mL) was removed and mixed with 50 mL of 20% trichloroacetic acid (TCA) and centrifuged at 2,560 × g for 15 min to obtain the 10% TCA-soluble nitrogen and 10% TCA-insoluble nitrogen fractions. The supernatant was decanted and analyzed for nitrogen by a combustion method using the Leco FP-2000 nitrogen analyzer (LECO Corporation, St. Joseph, MI).

The $DH$ was calculated as:

$$DH = \left(\frac{\text{10\% TCA \text{–} soluble N in the sample}}{\text{Total N in the sample}}\right) \times 100$$

**Proximate Composition**

Freeze-dried soluble fractions of pollock skin hydrolyzed for 10 min (PPH10), 30 min (PPH30) and 45 min (PPH45) were analyzed in triplicate for moisture and ash contents using the Association of Official Analytical Chemists standard methods 930.15 and 942.05, respectively (AOAC 1995). Fat content was determined using dichloromethyl ether in automated ASE-200 fat extractor (Dionex Corporation, Sunnyvale, CA). The nitrogen content was determined in triplicate using the Leco FP-2000 nitrogen analyzer (LECO Corporation). The protein content was calculated as percent nitrogen times 6.25. The yield was calculated by determining the dried protein powder weight as a percentage of the total raw material wet weight (Hoyle and Merritt 1994).

**Amino Acid and Mineral Analysis**

Amino acid profiles were determined by the AAA Service Laboratory Inc., Boring, OR. Samples were hydrolyzed with 6 N HCl and 2% phenol at 110°C for 22 h. Amino acids were quantified using a Beckman 6300 analyzer with post-column ninhydrin derivatization. Tryptophan and cysteine contents were not determined.

Samples for mineral analysis were ashed overnight at 550°C. Ashing residues were digested overnight in an aqueous solution containing 10% (v/v) hydrochloric acid and 10% (v/v) nitric acid. Digested solutions were diluted as needed and analyzed for Ag, Ca, Cd, Cu, Fe, K, Hg, Mg, Mn, Ni, P, Pb, Sr and Zn by inductively coupled plasma optical emission spectroscopy on a Perkin Elmer Optima 3000 Radial ICP-OES (PerkinElmer Life and Analytical Sciences, Inc., Boston, MA).
Physical Properties

Bulk density was measured in triplicate using a 25 mL glass measuring cylinder. The dried pollock skin hydrolysate sample was slowly filled to the appropriate level in the cylinder without shaking, and a level surface was obtained. The cylinder weight and weight-to-volume ratio were taken, and the pollock skin hydrolysate bulk density was reported as gram per milliliter. The AquaLab water activity meter (model Series 3TE, Decagon Devices, Inc., Pullman, WA) was used to measure in triplicate the water activity of samples at 22°C.

Functional Properties

Three separate experiments were conducted for all functional properties, and results are reported on a protein content basis. Nitrogen solubility was determined following the procedure of Morr et al. (1985). Five hundred milligrams of PPH10, PPH30 and PPH45 samples was dispersed in 50 mL of 0.1 M NaCl at pH 7.0, and the solution was stirred for 1 h at 25°C, and then centrifuged at 2,560 × g for 30 min. The supernatant was analyzed for nitrogen content using a Leco FP-2000 nitrogen analyzer. Percent nitrogen solubility of PPH samples was calculated as follows: nitrogen solubility (%) = (supernatant nitrogen content/total sample nitrogen content) × 100.

Emulsifying stability (ES) was evaluated according to the method of Yatsumatsu et al. (1972). Five hundred milligrams of PPH10, PPH30 and PPH45 samples was transferred into a 250 mL beaker and dissolved in 50 mL of 0.1 M NaCl, and then 50 mL of soybean oil was added. The homogenizer equipped with a motorized stirrer driven by the rheostat was immersed to 75% of the total depth of the mixture, and operated for 2 min at 100% output at 120 V to make an emulsion. Three 25 mL aliquots were immediately taken from the emulsion and transferred into three 25 mL graduated cylinders. The emulsions were allowed to stand for 15 min at 25°C, and then the aqueous volume was read and ES percent was calculated as follows: ES (%) = [(total volume – aqueous volume)/total volume] × 100.

The fat adsorption capacity of the PPH10, PPH30 and PPH45 samples was determined by placing 500 mg of PPH10, PPH30 and PPH45 samples into a 50 mL centrifugal tube and adding 10 mL of soybean oil (Shahidi et al. 1995). The samples were thoroughly mixed with a small steel spatula, kept for 30 min at 25°C with intermittent mixing every 10 min and then centrifuged at 2,560 × g for 25 min. Free oil was decanted, and the fat absorption of the sample was determined from the weight difference. The fat adsorption capacity was expressed in terms of milliliters of fat adsorbed by 1 g protein.
Sodium Dodecyl Sulfate-Polyacrylamide Gel Electrophoresis (SDS-PAGE) Electrophoresis

The SDS tricine/PAGE system was used with a Photodyne Foto/Force 300 power supply (Hartland, WI) and a single-sided vertical gel electrophoresis system (Owl Separation Systems, Portsmouth, NH) under reducing conditions according to Schagger and Von Jagow (1987). Novex Precast 10–20% Tricine gels (Invitrogen Life Technologies, Carlsbad, CA) were used, and ColorBurst molecular mass standards were purchased from Sigma-Aldrich (number C 4105, St. Louis, MO). Protein bands were visualized by staining the gels with Coomassie blue.

Glazing Salmon Fillets with PPH Solutions

PPH-coating solutions were prepared from PPH10, PPH30 and/or PPH45 by slowly dissolving 4.7% PPH in 93.5% DW and 1.8% glycerin (GC). Fresh skinless pink salmon fillets (Oncorhynchus gorbuscha) were obtained from a seafood processing plant in Kodiak, AK. Before glazing, triplicate samples of the initial pink salmon fillets were analyzed for proximate composition, thiobarbituric acid (TBA) and pH.

The weight of salmon fillets was 200–250 g, and the fillets were cut into 85–100 g pieces and frozen at –20°C for 1 h using a Dole plate freezer (Freeze-Cel Plate Freezer, Dole Refrigerating Company, Lewisburg, TN). The frozen fillets were dipped in pollock skin hydrolysate solutions at 4°C for 30 s, drained for 30 s, frozen again at –20°C in the plate freezer for 1 h, individually packed in zip lock freezer bags and stored in a box at –35°C for 4 months. Nonglazed (NG) fillets, fillets coated with DW and/or GC were used as controls.

Yield of Pink Salmon Fillets After Glazing, Thaw Loss and Drip Loss

The fillet pieces (n = 3) were weighed before and after glazing. The yield (wt gain, %) of glazed fillets was calculated as follows: yield % = (wt of glazed fillet pieces/wt of nonglazed raw fillet pieces) × 100.

To calculate thaw yield (%), the frozen fillet pieces were removed from the freezer, thawed for 22 h at refrigerated temperature (5°C), removed from the zip-lock polyethylene bag and placed on a rack for 2 min to release liquid drip, and then the fillet pieces were weighed again. Calculations for thaw % and drip % are as follows:

\[
\text{Thaw yield \%} = \left(\frac{\text{wt of thawed glazed fillet pieces}}{\text{wt of nonglazed raw fillet pieces}}\right) \times 100
\]
Drip loss % = [(wt of frozen glazed fillet pieces – wt of thawed glazed fillet pieces) × 100]/wt of frozen glazed fillet pieces

**Determination of pH and TBA**

The pH of the nonglazed and glazed salmon fillet pieces after 8 months frozen storage was evaluated according to the method of Ingolfsdottir *et al.* (1998) with some modifications. Twenty grams of minced muscle was placed in a 400 mL beaker and homogenized (model 6-105-AF, Virtis Co., Gardner, NY) with 80 mL DW for 1 min, and the pH was measured with a Beckman pH meter.

The TBA test was conducted on the salmon fillet samples after 4 months of frozen storage using the method of Lemon (1975). Malondialdehyde (MDA) in the samples was measured and reported as values of TBA in units of mg MDA/kg samples.

**Statistical Analysis**

Mean values from the three separate experiments or replicate analyses were reported. The statistical significance of observed differences among treatment means was evaluated by analysis of variance (SAS version 8.2, SAS Institute, Inc., Cary, NC), followed by the post hoc Tukey’s studentized range test (SAS 2002).

**RESULTS AND DISCUSSION**

**Properties of Raw Pink Salmon Fillets and PPH**

The freeze-dried PPH10, PPH30 and PPH45 had protein values of 93.7, 94.5 and 93.8%, respectively. All the PPH had less than 1% fat content (Table 1). Bulk density values of the pollock skin hydrolysate samples were 0.12–0.14 g/cm³ (Table 1). Water activities of all fish meals were similar, although statistical differences were found, with a range of 0.06–0.08 (Table 1). The low water activity values indicated that molds, yeasts and bacteria would not grow on the dried hydrolysates (Rockland and Stewart 1981). During storage and transport, cycled dehydration and humidification of fish protein meal can cause major changes in the water activity, which may lead to severe problems including microbial deterioration and caking (Bligh 1992). A steady increase in $DH\%$ was observed with increased hydrolysis time (Fig. 1); however, after an initial rapid onset phase, the rates of hydrolysis tended to slightly decrease, an observation consistent with that of Mackie.
The general shape of the hydrolysis curves (Fig. 1) was similar to those published for other fish protein hydrolysates (Hoyle and Merritt 1994; Liceaga-Gesualdo and Li-Chan 1999; Sathivel et al. 2003). The DH% values after 10, 30 and 45 min of hydrolysis were 13.2, 28 and 36%, respectively.

### Amino Acid and Mineral Contents

The amino acid contents of the PPH samples are listed in Table 2. The essential amino acid contents (mg of amino acid/g protein) of all the samples were higher than the recommended values for human adults (FAO/WHO 1990). The lysine content of PPH samples ranged from 44.9 to 46.1 (mg of amino acid per g protein) and was lower than that reported for pollock

### Table 1

PROXIMATE COMPOSITION OF RAW SALMON FILLETS AND PROXIMATE COMPOSITION AND PHYSICAL PROPERTIES OF POLLOCK SKIN HYDROLYSATES*

<table>
<thead>
<tr>
<th></th>
<th>Raw</th>
<th>PPH10</th>
<th>PPH30</th>
<th>PPH45</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (%)</td>
<td>24.1 ± 1.8</td>
<td>93.7 ± 1.0</td>
<td>94.5 ± 1.5</td>
<td>93.8 ± 1.5</td>
</tr>
<tr>
<td>Moisture (%)</td>
<td>77.3 ± 1.0</td>
<td>4.8 ± 0.1</td>
<td>4.1 ± 0.1</td>
<td>4.7 ± 0.1</td>
</tr>
<tr>
<td>Lipid (%)</td>
<td>0.3 ± 0.1</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>1.4 ± 0.1</td>
<td>1.5 ± 0.2</td>
<td>1.4 ± 0.1</td>
<td>1.5 ± 0.2</td>
</tr>
<tr>
<td>Density (g/cm³)</td>
<td>0.12 ± 0.00ª</td>
<td>0.12 ± 0.01ª</td>
<td>0.14 ± 0.01ª</td>
<td></td>
</tr>
<tr>
<td>Water activity</td>
<td>0.08 ± 0.00ª</td>
<td>0.06 ± 0.00ª</td>
<td>0.07 ± 0.00ª</td>
<td></td>
</tr>
</tbody>
</table>

* Values are means ± standard deviation of three determinations. Means with the same letter in each row are not significantly different (P > 0.05).

PPH10, 10 min hydrolyzed pollock skin; PPH30, 30 min hydrolyzed pollock skin; PPH45, 45 min hydrolyzed pollock skin.

FIG. 1. EFFECT OF DIGESTION TIME ON DEGREE OF HYDROLYSIS (%DH) OF POLLOCK SKIN

(1982). The general shape of the hydrolysis curves (Fig. 1) was similar to those published for other fish protein hydrolysates (Hoyle and Merritt 1994; Liceaga-Gesualdo and Li-Chan 1999; Sathivel et al. 2003). The DH% values after 10, 30 and 45 min of hydrolysis were 13.2, 28 and 36%, respectively.

### Amino Acid and Mineral Contents

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by-products including heads, liver and viscera protein powders (Sathivel and Bechtel 2006). The high values for hydroxyproline (53.4–58.6 mg amino acid/g protein) and proline (94.5–96.1 mg amino acid/g protein) in the PPH samples indicated higher concentrations of connective tissue proteins in these samples. The hydroxyproline contents of the PPH samples were much higher than those reported for pollock heads, pollock viscera and pollock liver (Sathivel and Bechtel 2006). The total essential amino acid content constituted (17.7–18.3%) of the total amino acids.

The PPH samples were rich in K and P, and had relatively low levels of Ca (0.05% on a dry weight basis) (Table 3). The mineral contents of the skin hydrolysate powders were generally lower than those reported for soluble protein powders made from pollock heads, pollock viscera and pollock liver (Sathivel and Bechtel 2006). Hydrolyzed skin samples had low calcium-to-phosphorus ratios, as expected for tissues lacking bone, and also low values for Mn, Fe, Cd and Pb.

### TABLE 2.
**AMINO ACID COMPOSITION OF POLLOCK SKIN HYDROLYSATES**

<table>
<thead>
<tr>
<th>Amino acid</th>
<th>PPH10</th>
<th>PPH30</th>
<th>PPH45</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydroxyproline</td>
<td>58.6</td>
<td>53.8</td>
<td>53.4</td>
</tr>
<tr>
<td>Aspartic acid</td>
<td>75.4</td>
<td>74.8</td>
<td>74.7</td>
</tr>
<tr>
<td>Threonine†</td>
<td>31.3</td>
<td>31.0</td>
<td>31.0</td>
</tr>
<tr>
<td>Serine</td>
<td>60.5</td>
<td>59.0</td>
<td>58.8</td>
</tr>
<tr>
<td>Glutamic acid</td>
<td>112.9</td>
<td>111.0</td>
<td>111.0</td>
</tr>
<tr>
<td>Proline</td>
<td>96.1</td>
<td>94.7</td>
<td>94.5</td>
</tr>
<tr>
<td>Glycine</td>
<td>202.8</td>
<td>203.3</td>
<td>203.6</td>
</tr>
<tr>
<td>Alanine</td>
<td>78.7</td>
<td>79.2</td>
<td>79.6</td>
</tr>
<tr>
<td>Valine†</td>
<td>27.2</td>
<td>26.8</td>
<td>27.1</td>
</tr>
<tr>
<td>Methionine†</td>
<td>20.4</td>
<td>24.6</td>
<td>24.7</td>
</tr>
<tr>
<td>Isoleucine†</td>
<td>17.9</td>
<td>18.2</td>
<td>18.4</td>
</tr>
<tr>
<td>Leucine†</td>
<td>35.0</td>
<td>35.5</td>
<td>35.8</td>
</tr>
<tr>
<td>Tyrosine</td>
<td>9.9</td>
<td>10.7</td>
<td>10.6</td>
</tr>
<tr>
<td>Phenylalanine†</td>
<td>22.3</td>
<td>23.7</td>
<td>23.0</td>
</tr>
<tr>
<td>Histidine</td>
<td>15.5</td>
<td>15.9</td>
<td>16.1</td>
</tr>
<tr>
<td>Lysine†</td>
<td>44.9</td>
<td>45.7</td>
<td>46.1</td>
</tr>
<tr>
<td>Arginine</td>
<td>90.8</td>
<td>91.8</td>
<td>91.6</td>
</tr>
<tr>
<td>TEAA</td>
<td>176.7</td>
<td>181.9</td>
<td>183.1</td>
</tr>
<tr>
<td>TAA</td>
<td>1,000.0</td>
<td>1,000.0</td>
<td>1,000.0</td>
</tr>
<tr>
<td>TEAA/TAA (%)</td>
<td>17.7</td>
<td>18.2</td>
<td>18.3</td>
</tr>
</tbody>
</table>

* Data expressed as milligrams of amino acid per gram of protein.
† Essential amino acids for adult humans.

PPH10, 10 min hydrolyzed pollock skin; PPH30, 30 min hydrolyzed pollock skin; PPH45, 45 min hydrolyzed pollock skin; TEAA, total essential amino acid; TAA, total amino acid.
Functional Properties

Protein solubility is one of the most important physicochemical and functional properties (Kinsella 1976; Mahmoud et al. 1992). Nitrogen solubility (%) values for the pollock hydrolysate protein powders are shown in Table 4. PPH45 (79.6%) had significantly higher solubility than PPH30 (72.6%) and PPH10 (64.8%). These solubility values were lower than those reported for egg albumin (89.8%) and higher than soy protein concentrate (9.8%) (Sathivel et al. 2004). Hoyle and Merritt (1994) reported higher solubility values of 89.7–93.1% for herring muscle protein hydrolysates; however,
Sathivel et al. (2004) reported values of 63.4–87.2% for soluble herring protein powders. High nitrogen solubility values often indicate potential applications of protein ingredients in formulated food systems for enhancement of product appearance and mouthfeel (Petersen 1981).

Chobert et al. (1988) reported that smaller peptide units often had greater solubility than intact proteins. PPH10 and PPH30 had similar banding patterns with a predominance of smaller proteins and peptides with molecular weights less than 25 kDa (Fig. 2). The PPH45 samples had a greater abundance of proteins and fragments with molecular weights below 13 kDa. All three pollock skin hydrolysates had common protein bands between 8 and 25 kDa. It was interesting to note that PPH10 and PPH30 had different percent nitrogen solubility values (Table 4) and different DH% (Fig. 1), but some similarity in electrophoresis protein banding patterns (Fig. 2).

ES has been used to evaluate whether protein powders are able to form a stable protein emulsions containing a high level of oil. The ES value for PPH10 and PPH30 was significantly higher (P > 0.05) than for the PPH45 (Table 4). The ES values of 64.5–66.4% were reported for herring protein powders (Sathivel et al. 2004), values of 52–61% for hydrolyzed Atlantic

FIG. 2. SODIUM DODECYL SULFATE (SDS)-TRICINE/POLYACRYLAMIDE GEL ELECTROPHORESIS PROFILES OF THE POLLOCK SKIN HYDROLYSATES AND SDS MARKERS
Lane 1, SDS marker (8–210 kDa); lane 2, lower molecular marker (8–38 kDa); lane 3, pollock skin protein hydrolysate (PPH) 30 min; lane 4, PPH 10 min; lane 5, PPH 45 min.
salmon muscle protein (Kristinsson and Rasco 2000) and 48.6–54.2% for hydrolyzed herring products (Sathivel et al. 2003). Lee et al. (1987) reported that a peptide should have a minimum length of 20 residues to possess good emulsifying and interfacial properties. Jost et al. (1977) stated that smaller proteins and peptides may result in lower emulsifying values, such as those observed in this study for PPH45 (34.7%). PPH10 and PPH30 with similar electrophoretic profiles (Fig. 2) had slightly higher ES values than PPH45; however, a relationship between the protein and peptide size and emulsifying properties was evident. Gauthier et al. (1993) stated that other factors such as protein solubility and hydrophobicity also play major role in emulsifying properties.

As shown in Table 4, the fat adsorption values for PPH45 (4.7 mL oil/g protein) were greater \((P < 0.05)\) than PPH10 (3.6 mL oil/g protein) and PPH30 (3.8 mL oil/g protein). Reported fat adsorption capacity values ranged from 3.9 to 11.5 mL oil/g protein for herring protein powders (Sathivel et al. 2004), from 3.7 to 7.3 mL oil/g protein for hydrolyzed herring by-product proteins (Sathivel et al. 2003) and from 2.9 to 7.1 mL oil/g protein for Atlantic salmon protein hydrolysates (Kristinsson and Rasco 2000).

**Fillet Yield, Drip Loss, pH and Color**

Glazing solutions containing PPH, DW and GC significantly increased the yield of fillets, with increases from 11.1 to 13.5% over fillets receiving no

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**FIG. 3. THIOBARBITURIC ACID (TBA)-REACTIVE-SUBSTANCES OF GLAZED PINK SALMON FILLETS DURING 4 MONTHS OF FROZEN STORAGE**

NG, nonglazed; PPH10, glazed with 10 min hydrolyzed pollock skin; PPH30, glazed with 30 min hydrolyzed pollock skin; PPH45, glazed with 45 min hydrolyzed pollock skin; DW, glazed with distilled water; GC, glazed with 1.8% glycerin. RAW = pink salmon fillet at day 0 of storage study.
glazing (NG) (Table 5). The thaw yield of glazed fillets was higher \( (P < 0.05) \) than NG; however, the glazed fillets had similar large drip losses (8.0–10.4%), which were higher \( (P < 0.05) \) than NG (0.7%). Drip loss in frozen fish fillets can be caused by myosin aggregation during frozen storage, which leads to muscle toughening and drip loss during thawing (Mackie 1982). Thawing temperature has been shown to be important with slow thawing at 5°C resulting in higher liquid losses than fast thawing at 25°C in water (Bilinski et al. 1977). The initial pH of raw nonglazed pink salmon fillets was 6.63 and after 4 months of frozen storage, the pH of glazed pink salmon fillets remained relatively unchanged with values ranging from 6.5 to 6.8. Sigurgisladottir et al. (2000) reported that freezing and thawing may cause changes in the pH of the fish muscle; however, this was not observed in this study.

### Lipid Oxidation

The average \( (n = 3) \) protein, moisture, fat and ash contents of raw nonglazed pink salmon fillet were 18.6 ± 0.6, 79.1 ± 0.2, 1.7 ± 0.5 and 1.4 ± 0.01%, respectively. After 4 months of frozen storage, the TBA of NG was approximately 16 times higher that of fresh raw pink salmon fillets (Fig. 3). TBA (mg MDA/kg sample) values of fillets glazed with PPH, DW and GC were significantly lower than NG (3.9). Fillets glazed with PPH10 had a significantly lower TBA value than PPH30, PPH45, DW and GC treatments. Kim et al. (2001) reported that low-molecular-weight proteins and peptides generated during hydrolysis coupled with free amino acids could be responsible for antioxidant activity. Chen and Decker (1994) postulated that peptides

<table>
<thead>
<tr>
<th>Sample</th>
<th>Yield (%)</th>
<th>Thaw yield (%)</th>
<th>Drip loss (%)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>NG</td>
<td>100.0 ± 0.0b</td>
<td>98.8 ± 0.3b</td>
<td>0.7 ± 0.4b</td>
<td>6.6 ± 0.1a</td>
</tr>
<tr>
<td>PPH10</td>
<td>112.0 ± 1.7a</td>
<td>100.7 ± 0.3a</td>
<td>10.4 ± 1.0a</td>
<td>6.6 ± 0.1a</td>
</tr>
<tr>
<td>PPH30</td>
<td>113.0 ± 1.8a</td>
<td>101.1 ± 0.3a</td>
<td>10.3 ± 1.0a</td>
<td>6.5 ± 0.1a</td>
</tr>
<tr>
<td>PPH45</td>
<td>113.5 ± 1.8a</td>
<td>101.3 ± 0.9a</td>
<td>9.8 ± 2.1a</td>
<td>6.5 ± 0.1a</td>
</tr>
<tr>
<td>DW</td>
<td>111.7 ± 1.7a</td>
<td>100.8 ± 0.5a</td>
<td>8.6 ± 0.1a</td>
<td>6.8 ± 0.1a</td>
</tr>
<tr>
<td>GC</td>
<td>111.1 ± 1.6a</td>
<td>101.0 ± 0.5a</td>
<td>8.0 ± 1.0a</td>
<td>6.6 ± 0.1a</td>
</tr>
</tbody>
</table>

* Values are means ± standard deviation of three determinations. Means with the same letter in each column are not significantly different \( (P > 0.05) \). NG, nonglazed; PPH10, salmon fillets glazed with 10 min hydrolyzed pollock skin; PPH30, salmon fillets glazed with 30 min hydrolyzed pollock skin; PPH45, salmon fillets glazed with 45 min hydrolyzed pollock skin; DW, salmon fillets glazed with distilled water; GC, salmon fillets glazed with 1.8% glycerin.
containing basic amino acids could be electron acceptors from radicals formed during the oxidation of unsaturated fatty acids. Although PPH45 had lower molecular weight proteins and peptides than PPH30 and PPH10, a clear relationship between peptide size in the glazing solution and lipid oxidation in the frozen fillets was not found. In this study, the PPH45 was less effective than that of PPH10 in controlling lipid oxidation. Cisneros-Zevallos *et al.* (1994) reported that effectiveness of food surface coatings depended on a number of factors such as coating permeability, thickness and surface coverage by the coating materials. Additional knowledge of the physical and chemical properties of the PPH glazing material, such as permeability properties of the PPH films, and the glazing interactions with the surface of the salmon fillets will result in a better understanding of factors affecting lipid oxidation of the glazed salmon fillets.

**CONCLUSION**

The study demonstrated that DH% significantly affected the functional properties of pollock skin hydrolysates. PPH45 had significantly higher solubility and fat adsorption values but lower ES values than that of PPH30 and PPH10 hydrolysate powders. Glazed fillets had a higher yield and thaw yield than control nonglazed salmon fillets. After 4 months of frozen storage, the TBA value of NG was approximately 16 times higher than that of fresh raw pink salmon fillets, and the TBA values of glazed fillets were significantly lower (*P* < 0.05) than NG. The pollock skin hydrolysates have many desirable properties, including ES and fat adsorption capacity, which support the use as natural food ingredients.

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powders from arrowtooth flounder (Atheresthes stomias) and herring (Clupea harengus) byproduct. J. Agric. Food Chem. 52, 5040–5046.