Chitosan and Protein Coatings Affect Yield, Moisture Loss, and Lipid Oxidation of Pink Salmon (*Oncorhynchus gorbuscha*) Fillets During Frozen Storage

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**ABSTRACT:** The effects of chitosan (CH1 = 1% and CH2 = 2% solution), egg albumin (EA), soy protein concentrate (SPC), pink salmon protein powder (PSP), and arrowtooth flounder protein powder (AFP) as edible coatings on quality of skinless pink salmon fillets were evaluated during 3 mo frozen storage. Coating with 2% chitosan (CH2) resulted in significantly higher yield than coating with PSP and AFP. The thaw yield of salmon fillets coated with CH2 was higher than those of the control and fillets coated with AFP. The noncoated, CH1-, and CH2-coated fillets had similar drip loss (0.4% to 1.2%), which was lower than those observed for PSP- and AFP-coated fillets. All fillet samples had similar cook yield (84.2% to 88.8%). The fillet coated with CH1, CH2, SPC, and EA had significantly higher (P < 0.05) moisture content after thawing than the control noncoated fillets. Coating with CH1 and CH2 was effective in reducing about 50% relative moisture loss compared with the control noncoated fillets. Chitosan (CH1 and CH2) and SPC delayed lipid oxidation. There were no significant (P > 0.05) effects of coating on a*, b*, and whiteness values for cooked fillets after 3 mo frozen storage.

Keywords: edible coating, pink salmon fillets, chitosan, fish powders, lipid oxidation, relative moisture loss

**Introduction**

Fish is an extremely perishable food compared with other fresh commodities. Therefore, its marketing traditionally focused on frozen and processed products (Bligh 1979). Development of frozen and processed fish products from Alaska is hampered by the short shelf life of many seafoods. Freezing is a common preservation method used to control or decrease biochemical changes in fish that occur during storage. However, frozen storage does not completely inhibit chemical reactions (for example, lipid oxidation) that lead to quality deterioration of fish. Preservatives such as phosphates are often applied to seafood products to improve their shelf life, water-binding capacity, and frozen stability properties. Phosphates are used in seafood to enhance water-holding capacity, and to improve cooking yield (Shahidi 1994). Antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) have been commonly used by the food industry to improve product quality during storage, resulting in increased product shelf life. To meet consumer demands for safer foods, numerous studies are currently focused on using natural ingredients to enhance food quality and shelf life to avoid the use of synthetic preservatives. Biodegradable polysaccharides or proteins can be used to coat fish fillets to suppress quality changes during frozen storage. Some biodegradable edible coatings applied on frozen foods act as barriers to control moisture transfer and oxygen uptake (Debeaufort and others 1998).

A number of coating materials have been tested in attempt to maintain quality and prolong shelf life of meat products. Stuchell and Krochta (1995) reported that king salmon coated with whey protein isolates showed delayed lipid oxidation during frozen storage. Alginate-coated precooked, frozen stored pork patties had better sensory qualities than control patties (Wanstedt and others 1981). Jeon and others (2002) demonstrated that chitosan-coated Atlantic cod and herring had reduced moisture loss and lipid oxidation. Precooked, refrigerated pork chops and beef patties coated with starch-alginate, starch-alginate-tocopherol, and starch-alginate-rosemary exhibited reduced warmed-over flavor (Hargens-Madsen and others 1995; Handley and others 1996).

Research on shelf-life extension of fish by edible coating has so far been limited. Little has been published on fish protein powder edible coatings applied to foodstuff. The objective of this study was to evaluate the effects of coatings made with chitosan, egg albumin, soy protein, pink salmon protein powder, and arrowtooth flounder protein powder on quality of skinless pink salmon fillets during frozen storage.

**Materials and Methods**

**Preparation of chitosan, egg albumin, and soy protein concentrate coating solutions**

Shrimp chitosan (CH) with a viscosity of 115 cps and 83.3% degree of deacetylation was purchased from Vanson Halosource Inc. (Redmond, Wa., U.S.A.). To prepare 1% and 2% w/v CH solutions in lactic acid, 10 and 20 g CH were, respectively, mixed with 970 and 960 g water, stirred for 10 min, then 20 g lactic acid was added and stirred for 2 h. Purified egg albumin (EA) containing 82.4% protein (J.T. Baker Inc., Phillipsburg, N.J., U.S.A.) and soy protein concentrate (SPC) containing 66% protein (Central Soya Inc., Fort Wayne, Ind., U.S.A.) were used to prepare edible coating solutions. Using the method of Rhim and others (1998), EA and SPC edible coating solutions were prepared by slowly dissolving 4.7% of EA or SPC in

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distilled water (93.5%) and glycerin (1.8%) with constant stirring. EA and SPC coating solutions were pH-adjusted to 10 ± 0.1 with 1 N NaOH and heated for 20 min in a temperature-controlled water bath at 70 °C. Coating solutions were filtered through 8 layers of cheesecloth to remove undissolved materials.

Preparation of fish protein coating solutions

Fresh skinless arrowtooth flounder (Atheresthes stomias) and pink salmon fillets (Oncorhynchus gorbuscha) were obtained from a commercial fish processing plant in Kodiak, Alaska. The arrowtooth flounder and salmon fish fillets were separately ground in a Hobart grinder (KSS, Hobart Corp., Troy, Ohio, U.S.A.) through a 7-cm dia plate having 12-mm dia openings, and subsequently ground through a plate with 6-mm dia openings. The ground samples were freeze-dried, and the resulting fish protein powders were vacuum packed and stored at 4 °C until analyzed. The fish protein coating solutions were prepared by slowly dissolving 4.7% protein powders in distilled water (93.5%) and glycerin (1.8%) with constant stirring, using the method of Rhim and others (1998). PSP and AFP coating solutions were pH-adjusted to 10 ± 0.1 with 1 N NaOH and heated for 20 min in a temperature-controlled water bath at 70 °C. Coating solutions were filtered through 8 layers of cheesecloth to remove undissolved materials.

Salmon fillets preparation and coating application

Fresh skinless medium-size pink salmon fillets were obtained from a commercial fish processing plant in Kodiak, Alaska. The average weight of salmon fillets was 200 to 250 g, and the fillets were cut into 85 to 100 g pieces. The fillet pieces were dipped in each freshly prepared coating solution for 5 min, drained for 2 min, weighed, packed in polyethylene freezer bags, and stored at –20 °C for 3 mo.

Yield of pink salmon fillets after coating, thawing and cooking, and drip loss

The fillet pieces (n = 3) were weighed before and after coating. The yield (wt gain, %) of coated fillets was calculated as (wt of coated fillet pieces/wt of noncoated raw fillet pieces) × 100. To calculate thaw yield (%), frozen fillet pieces were removed from the freezer, kept 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. Then the thawed fillet pieces were weighed. The thaw yield (%) was calculated as (wt of thawed coated fillet pieces/wt of noncoated raw fillet pieces) × 100. Drip loss (%) was calculated as [(wt of frozen coated fillet pieces – wt of thawed coated fillet pieces) × 100]/wt of frozen coated fillet pieces. The thawed fillet pieces were transferred to individual boiling bags, vacuumed, and cooked in a water bath at 95 °C for 5 min. The cooked samples were removed from the bag and placed on a rack for 1 min to release liquid drip, and the cooked fillet pieces were weighed. The cook yield (%) was calculated as (wt of cooked fillet pieces/wt of noncoated raw fillet pieces) × 100.

Determination of pH

The pH of the noncoated and coated salmon fillet pieces after 3 mo frozen storage was evaluated according to the method of Ingolfsdottir and others (1998) with some modifications. Twenty grams of minced muscle were placed in a 400-mL beaker and homogenized with 80 mL distilled water for 1 min by using a motorized stirrer of a homogenizer (model 6-105-AF; Virtis Co, Gardner, N.Y., U.S.A.). The pH of homogenized sample was measured with a Beckman pH meter.

Moisture content and relative moisture loss (RML)

The moisture content of fresh, frozen, thawed, and cooked fillet samples were measured. After 3 mo frozen storage, the relative moisture loss (RML, %) of pink salmon fillets was calculated as [(initial moisture content of fresh noncoated raw fillet – moisture content of thawed coated fillets)/initial moisture content of fresh noncoated raw fillet] × 100.

Thiobarbituric acid (TBA)

The TBA test was conducted on the salmon fillet samples after 3 mo frozen storage using the method of Lemon (1975). Malondialdehyde (MDA) in the samples was measured and reported as values of thiobarbituric acid (TBA) in units of mg MDA/kg samples.

Color

Color of raw and cooked coated salmon fillets was determined using a Minolta Chromameter (Model CR-300, Minolta Co., Ltd, Osaka, Japan) and reported as L*, a*, and b* values. L* describes the lightness of the sample, a* intensity in red (a* > 0), and b* intensity in yellow (b* > 0). Whiteness was calculated according to the following formula: Whiteness = 100 – [(100 – L*)2 + a*2 + b*2]1/2.

Statistical analysis

Mean values from the 3 separate experiments were reported. The statistical significance of observed differences among treatment means was evaluated by analysis of variance (ANOVA) (SAS Version 8.2, SAS Inst. Inc., Cary, N.C., U.S.A.), followed by the post hoc Tukey’s studentized range test (SAS 2002).

Results and Discussion

Chemical composition, TBA, color, and pH

The freeze-dried pink salmon and arrowtooth flounder protein powders, respectively, had 80.2% and 73.6% protein, 7.6% and 6.6% moisture, 6.4% and 15% fat, and 5.8% and 4.6% ash. The average (n = 3) protein, moisture, fat, and ash content of raw noncoated pink salmon fillets was 19.3%, 79.5%, 1.6%, and 1.2%, respectively. The initial TBA and pH value for the raw noncoated pink salmon fillets was 0.25 (mg MDA/kg fish) and 6.63, respectively. The L*, a*, b*, and whiteness values of raw noncoated pink salmon fillets were, respectively, 31.6, 8.6, 8.5, and 30.4.

Yield and drip loss

Chitosan and protein coatings increased the yield of raw pink salmon fillets, ranging from 2% to 3.8% (Table 1). Coating with 2% chitosan (CH2) resulted in significantly higher yield than coating with pink salmon protein powder (PSP) and arrowtooth flounder protein powder (AFP). The thaw yield of salmon fillets coated with CH2 was higher than those of the control and fillets coated with AFP. The noncoated, CH1-, and CH2-coated fillets had similar drip loss (0.4% to 1.2%), which was lower than those observed for PSP- and AFP-coated fillets. The higher drip loss observed with fillets coated with SA and AF may have been caused by dissolution or melting of PSP and
AFP protein coating from the surface of the fillets. Drip loss in frozen fish fillets is caused by myosin aggregation during frozen storage, thus leading to muscle toughening and drip loss during thawing (Mackie 1993). Slow thawing at 5 °C gives higher liquid loss than fast thawing at 25 °C in water (Bilinski and others 1977). All fillet samples had similar cook yield (84.2% to 88.8%), which implies that chitosan or protein coatings had no effect on the cook yield (Table 1). The pH of raw noncoated pink salmon fillets was 6.63. After 3 mo frozen storage, the pH of raw pink salmon fillets coated with chitosan or protein ranged from 6.4 to 6.7. It has been reported that freezing and thawing may cause changes in the pH of the fish muscle (Sigurgisladottir and others 2000). However, this was not observed in this study. Salmon fillets coated with chitosan at both 1% and 2% had a slightly lower pH (6.4) than that of AFP-coated fillets (pH 6.7).

### Moisture content and relative moisture loss (RML)

The fillet coated with CH1, CH2, SPC, and EA had significantly higher (P < 0.05) moisture content after thawing than the control noncoated fillets (Table 2). All cooked samples showed similar moisture content (P > 0.05) and cook yield (Table 1), which indicated that coating did not affect the moisture content during cooking.

After 3 mo frozen storage, the control noncoated fillets exhibited 4.1% moisture loss (Figure 1). Compared with the noncoated fillet, significantly lower RML in fillets was observed with CH1 (1.5%), CH2 (2.1%), SPC (1.8%), and EA (1.9%). Coating with CH1 and CH2 was effective in reducing about 50% relative moisture loss compared with the control noncoated fillets. The drip loss of fillets coated with CH1 and CH2 was less than that of SPC and AFP coatings (Table 1). Drip loss has been linked to partial denaturation of proteins that may function as moisture-sacrificing agents instead of moisture capacity (Mackie 1993). By using high quality raw materials and good control of storage conditions, drip loss after thawing and moisture loss during frozen storage could be minimized (Cormier and Leger 1987).

Both CH1 and CH2 seem to be good candidates for minimizing drip and moisture losses. Chitosan coating was effective in reducing water loss and in prolonging the storage life of cod fillets (Jeon and others 2002). It has been reported that water permeability is dependent on the relative polarity of the carbohydrate polymers. Kester and Fennema (1986) have reported that chitosan coatings may function as moisture-sacrificing agents instead of moisture barriers, thus moisture loss from the product could be delayed until the moisture contained within the chitosan coating had evaporated. Cuq and others (1995) reported that water vapor transfers through hydrophilic coating/film by sorption and diffusion is affected by a number of factors. Therefore, the mechanism involved in the water permeability through the chitosan coatings merits further investigations. A similar RML was observed for the control noncoated fillet and fillets coated with PSP and AFP.

### Lipid oxidation

The TBA value of fillets coated with CH2 1 mg MDA/kg sample, CH1 (1.1), SPC (1.4), and AFP (1.4) was significantly lower than the

### Table 2—Moisture content of thawed and cooked salmon fillets coated with chitosan and proteins after 3 mo frozen storage

<table>
<thead>
<tr>
<th>Samples</th>
<th>Moisture content after thawing (%)</th>
<th>Moisture content after cooking (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NC</td>
<td>76.3 ± 0.5a</td>
<td>74.7 ± 0.5a</td>
</tr>
<tr>
<td>CH1</td>
<td>78.3 ± 0.5a</td>
<td>74.4 ± 0.3a</td>
</tr>
<tr>
<td>CH2</td>
<td>77.9 ± 0.6a</td>
<td>75.6 ± 0.1a</td>
</tr>
<tr>
<td>SPC</td>
<td>78.1 ± 0.5a</td>
<td>75.0 ± 0.2a</td>
</tr>
<tr>
<td>EA</td>
<td>78.0 ± 0.5a</td>
<td>74.9 ± 1.1a</td>
</tr>
<tr>
<td>PSP</td>
<td>77.3 ± 0.9ab</td>
<td>75.2 ± 0.9a</td>
</tr>
<tr>
<td>AFP</td>
<td>77.1 ± 0.1ab</td>
<td>74.6 ± 0.7a</td>
</tr>
</tbody>
</table>

*aValues are means ± S.D. of 3 determinations. Means with the same letters in each column are not significantly different (P > 0.05).

*bNC = control; CH1 = salmon fillets coated with 1% chitosan; CH2 = salmon fillets coated with 2% chitosan; SPC = salmon fillets coated with soy protein concentrate; EA = salmon fillets coated with egg albumin; PSP = salmon fillets coated with pink salmon protein powder; AFP = salmon fillets coated with arrowtooth flounder protein powder.

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**Figure 1**—Relative moisture loss of pink salmon fillets during a 3-mo frozen storage. "ab"Means with the same letters are not significantly different (P > 0.05). NC = control; CH1 = salmon fillets coated with 1% chitosan; CH2 = salmon fillets coated with 2% chitosan, SPC = salmon fillets coated with soy protein concentrate; EA = salmon fillets coated with egg albumin; PSP = salmon fillets coated with pink salmon protein powder; AFP = salmon fillets coated with arrowtooth flounder protein powder.
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Table 3—Color L* a* b* values of chitosan- and protein-coated raw and cooked pink salmon fillets after 3 mo frozen storage

<table>
<thead>
<tr>
<th>Samples</th>
<th>Coatingsb</th>
<th>L*</th>
<th>a*</th>
<th>b*</th>
<th>Whiteness</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw</td>
<td>NC</td>
<td>38.0 ± 2.1ab</td>
<td>8.1 ± 0.9a</td>
<td>11.4 ± 2.0a</td>
<td>36.5 ± 2.1ab</td>
</tr>
<tr>
<td></td>
<td>CH1</td>
<td>35.9 ± 4.0abc</td>
<td>8.0 ± 1.0a</td>
<td>9.6 ± 1.4a</td>
<td>34.6 ± 3.6ab</td>
</tr>
<tr>
<td></td>
<td>CH2</td>
<td>37.6 ± 2.1ab</td>
<td>7.1 ± 1.0a</td>
<td>9.4 ± 1.3a</td>
<td>36.5 ± 1.9ab</td>
</tr>
<tr>
<td></td>
<td>SPC</td>
<td>33.4 ± 1.3a</td>
<td>7.3 ± 0.5a</td>
<td>9.0 ± 0.6a</td>
<td>32.4 ± 3.4b</td>
</tr>
<tr>
<td></td>
<td>EA</td>
<td>35.1 ± 1.2ab</td>
<td>6.9 ± 0.6a</td>
<td>9.7 ± 1.0a</td>
<td>34.0 ± 2.2ab</td>
</tr>
<tr>
<td></td>
<td>PSP</td>
<td>41.4 ± 2.3a</td>
<td>8.0 ± 0.7a</td>
<td>12.5 ± 0.9a</td>
<td>39.5 ± 2.0a</td>
</tr>
<tr>
<td></td>
<td>AFP</td>
<td>36.8 ± 0.6abc</td>
<td>7.4 ± 0.4a</td>
<td>9.7 ± 1.7a</td>
<td>35.6 ± 0.5ab</td>
</tr>
<tr>
<td>Cooked</td>
<td>NCC</td>
<td>69.6 ± 2.7abc</td>
<td>8.2 ± 1.3a</td>
<td>15.7 ± 0.9a</td>
<td>64.8 ± 2.8a</td>
</tr>
<tr>
<td></td>
<td>CHC1</td>
<td>68.4 ± 4.3abc</td>
<td>9.8 ± 1.4a</td>
<td>17.4 ± 2.2a</td>
<td>62.5 ± 3.0a</td>
</tr>
<tr>
<td></td>
<td>CHC2</td>
<td>70.8 ± 0.3abc</td>
<td>10.2 ± 0.4a</td>
<td>18.2 ± 1.4a</td>
<td>64.1 ± 0.4a</td>
</tr>
<tr>
<td></td>
<td>SPCC</td>
<td>72.2 ± 3.5a</td>
<td>9.3 ± 3.5a</td>
<td>15.7 ± 5.1a</td>
<td>66.7 ± 6.1a</td>
</tr>
<tr>
<td></td>
<td>EAC</td>
<td>65.0 ± 2.0a</td>
<td>9.1 ± 1.0a</td>
<td>15.5 ± 0.5a</td>
<td>60.6 ± 1.7a</td>
</tr>
<tr>
<td></td>
<td>PSPC</td>
<td>69.1 ± 1.5abc</td>
<td>9.6 ± 1.5a</td>
<td>15.5 ± 0.5a</td>
<td>64.1 ± 1.1a</td>
</tr>
<tr>
<td></td>
<td>AFPC</td>
<td>72.4 ± 1.7a</td>
<td>9.7 ± 0.8a</td>
<td>16.5 ± 1.0a</td>
<td>66.4 ± 1.9a</td>
</tr>
</tbody>
</table>

*aValues are means ± S.D. of 3 determinations. Means with the same letter in each column are not significantly different (P > 0.05).

control sample (3.3), indicating that the coatings were effective in reducing lipid oxidation (Figure 2). In this study, chitosan (CH1 and CH2) lowered the lipid oxidation. The antioxidant properties of chitosan in foods have been reported (Shahidi and others 1999). Jeon and others (2002) reported that chitosan coatings reduced the lipid oxidation in herring and Atlantic cod. The primary amino groups of chitosan would form a stable fluorosphere with volatile aldehydes such as malondialdehyde, which is derived from breakdown of fats during the oxidation (Weist and Karel 1992). Chitosan coating and film have been reported to be good barriers to oxygen permeability (Butler and others 1996). Therefore, chitosan (CH1 and CH2) coating applied on the surface of the pink salmon fillets may have acted as a barrier between the fillet and its surrounding, thus slowing down the diffusion of oxygen from the surrounding to the surface of the fillet.

The salmon fillets coated with SPC have also been found to be very effective in controlling lipid oxidation by serving as a barrier to O2 permeability (Brandenburg and others 1993). Kunte (1996) reported that chicken breast tumbled with 7S soy protein had less lipid oxidation, while soy protein decreased the TBA values in the cooked beef (Romijn and others 1991). The AFPC-coated salmon fillets showed reduced lipid oxidation activity similar to that of fillets coated with CH1, CH2, and SPC. This is likely due to the presence of antioxidant activity of peptide fractions in the arrowtooth flounder proteins. Sathivel and others (2003) reported that the fish protein hydrolysates had antioxidant properties. The extracted AFP from the skinless arrowtooth fillets had a smear of low molecular weight protein material due to partial hydrolysis (Sathivel and others 2004).

Color

Quality parameters, including flesh coloration, determine acceptability and price of salmon (Skrede and Storebakkan 1986). Chitosan and protein coatings had minimal effect on color lightness and whiteness of raw fillets (Table 3). However, the fillets coated with PSP (lighter and whiter) had significantly higher (P < 0.05) L* and whiteness values than fillets coated with SPC (darker). Coatings did not affect color yellowness (b*) and redness (a*) of raw salmon fillets. All stored samples had higher L* values than that of fresh raw salmon fillet (L* = 31.6). However, all stored samples had lower a* values compared with fresh raw salmon fillets (a* = 8.6). This indicates that reduction in red pigment may have occurred during the frozen storage. The coloration of salmonid fish flesh is due to carotenoid pigments, notably astaxanthin and canthaxanthin (Skrede and Storebakken 1986). Carotenoids are degraded in salmon during frozen storage (Sheehan and others 1998). Christiansen and others (1995) reported a minor decrease in pigment concentration of salmon during frozen storage, whereas Scott and others (1994) reported that the pigments were stable during frozen storage.

There were no significant (P > 0.05) effects of coating on a*, b*, and whiteness values for cooked fillets after 3 mo frozen storage. However, all cooked samples showed higher L* values compared with raw samples. Sikorski and others (1984) reported that increased lightness of the fish muscle during cooking may be due to leaching of collagen, which is located in between segments of muscles known as myotomes. Redness contributes significantly to the overall enjoyment of cooked salmon fish flesh (Sylvia and others 1995). The a* and b* values for all cooked samples were higher than that of raw samples. Carotenoids are bleached when exposed to...
adverse conditions including direct sunlight, excessive or prolonged heat, strong acids, or peroxides (Schiedt and Liaaen-Jensen 1995). Therefore the increased $a^*$ and $b^*$ values of cooked sample were unexpected. The moisture content of all cooked salmon fillets ranged from 74.4 to 75.6 (Table 2), whereas the moisture content of frozen sample ranged from 76.3 to 78.3. This indicates that moisture was lost during the cooking. The moisture loss might explain the increase in the yellow and red color pigment concentration (Choubert and others 1992).

Conclusions

This study shows potential of chitosan (both 1% and 2% solutions) as an edible coating for pink salmon fillets. Chitosan-coated fillets had a higher thaw yield than that of the control non-coated salmon fillets. The noncoated and chitosan (both 1% and 2% solution) coated fillets had lower drip loss than that of salmon and arrowtooth protein powders coated fillets. Chitosan (both 1% and 2% solutions) were effective in reducing moisture loss. Chitosan (1% and 2%), soy protein concentrate—coated fillets delayed lipid oxidation in pink salmon fillets during 3 mo frozen storage. There were no significant effects of coating on $L^*$, $a^*$, $b^*$, and whiteness values for cooked fillets.

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References

Kunte LA. 1996. Effectiveness of 75 soy protein and edible film in controlling lipid oxidation in chicken (MSc thesis). Available from Univ. of Nebraska Library. Lincoln, Neb.: Univ. of Nebraska. p 114.

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