

## Incorporation and Stabilization of Omega-3 Fatty Acids in Surimi Made from Cod, *Gadus morhua*

YOUNGJOON PARK, STEPHEN D. KELLEHER, D. JULIAN McCLEMENTS, AND  
 ERIC A. DECKER\*

Department of Food Science, University of Massachusetts, Amherst, Massachusetts 01003

Surimi containing  $\omega$ -3 fatty acids from algal oil was prepared by the addition of oil-in-water emulsions or bulk oil. Emulsion and bulk oil were added separately to surimi to provide ~500 mg of  $\omega$ -3 fatty acids per serving of surimi (85 g). Addition of the emulsion had no effect on surimi gel strength, whereas bulk oil decreased gel strength an average of 31%. All surimi treatments containing algal oil increased in Hunter  $b^*$  values due to the presence of carotenoids in the oil. Among cryoprotectants, sodium tripolyphosphate was the major surimi additive responsible for retarding the formation of lipid hydroperoxides and thiobarbituric acid reactive substances (TBARS). Lipid hydroperoxide and TBARS formation was lower in surimi containing bulk oil compared to surimi with emulsified oil. Both EDTA and lipid soluble antioxidants were able to decrease lipid oxidation in surimi fortified with  $\omega$ -3 fatty acids. This suggests that surimi containing nutritionally beneficial  $\omega$ -3 fatty acids could be developed with good oxidative stability and gel strength.

**KEYWORDS:** Lipid oxidation; surimi; emulsion; antioxidant; cryoprotectant; EDTA;  $\omega$ -3 fatty acid

### INTRODUCTION

There is increasing interest in the incorporation of  $\omega$ -3 polyunsaturated fatty acids, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), into foods because of their many health benefits (1). Most of the benefits of fish oils can be attributed to their high EPA and DHA contents. Commercial algal oils contain DHA as their major  $\omega$ -3 fatty acid (2). DHA is known to be crucial for proper nervous system and vision development (3). Both DHA and EPA are believed to have several health benefits for cardiovascular disease, immune disorders, inflammation, allergies, and diabetes (3, 4).

The susceptibility of lipids to oxidation is a major cause of quality deterioration in many types of natural and processed foods (5). In some foods, a limited amount of lipid oxidation is desirable with the generation of a desirable characteristic taste or smell (e.g., cheese; 5). On the other hand, lipid oxidation is undesirable in most foods because it leads to the development of off-flavors and potentially toxic reaction products (6). The highly unsaturated  $\omega$ -3 fatty acids are extremely susceptible to oxidation, resulting in potential alteration in nutritional composition as well as in the sensory quality of foods (7).

If  $\omega$ -3 polyunsaturated fatty acids are to be added to foods, it is likely to be in the form of lipid dispersions. The mechanism of lipid oxidation in bulk oils is different from that in oil-in-water emulsions (8). The interfacial membrane of an emulsion droplet is one of the main differences between oil-in-water emulsions and bulk oils that alter the chemistry of lipid oxidation

(9). Numerous research studies have shown that the characteristics of the emulsion droplet interfacial membrane can alter lipid oxidation rates by influencing interactions between aqueous phase prooxidants and oxidizable lipid substrates in relatively simple model systems (10, 11). However, little is known about how the interfacial properties of emulsion droplets will influence lipid oxidation rates in complex food systems.

Surimi contains little lipid, has a long shelf life, and is a highly functional myofibrillar protein ingredient of good nutritional quality (12). Surimi is obtained from fish skeletal muscle that is washed with water and blended with cryoprotectants. Cryoprotectants are compounds that extend the shelf life and quality of frozen foods. The presence of cryoprotectants is important to ensure maximum functionality of frozen surimi because freezing induces protein denaturation and aggregation (12). Typical cryoprotectants used in surimi include sucrose, sorbitol, and polyphosphates (13). Because many of the lipids and thus  $\omega$ -3 fatty acids can be removed by the surimi manufacturing process, the addition of  $\omega$ -3 fatty acids back to surimi could provide a useful functional food that mimics natural fish products. However, if this high  $\omega$ -3 fatty acid surimi is to be successful, oxidative deterioration must be controlled and the added lipids must not adversely impact texture. Because polyphosphates are excellent antioxidants (14), surimi may represent an excellent vehicle for oxidatively stable  $\omega$ -3 fatty acids.

Due to the current interest in supplementing nutritional foods with  $\omega$ -3 fatty acids, the objective of this research was to determine the oxidative stability of  $\omega$ -3 fatty acids from bulk oil or oil-in-water emulsions in surimi. The role of exogenous

\* Author to whom correspondence should be addressed (e-mail edecker@foodsci.umass.edu).

and endogenous antioxidants on the oxidative stability of  $\omega$ -3 fatty acid was evaluated. The impact of bulk oil and emulsions on surimi gel strength and color was also determined.

## MATERIALS AND METHODS

**Materials.** Algal oil was donated by Martek Biosciences (Boulder, CO). DHA Gold oil with and without lipid soluble antioxidants was used in this study. Oil with lipid soluble antioxidants contained 1000 ppm of tocopherol isomers, 1000 ppm of rosemary extract, and 500 ppm of ascorbyl palmitate. Upon receipt of the oil, it was divided into capped vials (28 g each) and frozen at  $-80^{\circ}\text{C}$  until needed. Imidazole was obtained from Sigma Chemical Co. (St. Louis, MO). Whey protein isolates (whey protein isolate, 96.2% protein) were a gift from Davigo (Minneapolis, MN).  $\text{Na}_2\text{EDTA}$  was obtained from Curtin Matheson Scientific, Inc. (Houston, TX). Sodium acetate was purchased from Fisher Scientific (Fair Lawn, NJ). All other chemicals were of reagent grade or better. Cod was obtained from a local seafood processor in Gloucester, MA.

**Preparation of Emulsion.** A coarse emulsion was prepared by homogenizing 30% algal oil with 70% aqueous phase using a Brinkman Polytron (Westbury, NY), model PT 10/35. The aqueous phase consisted of a buffer solution containing 5 mM imidazole and 5 mM sodium acetate (pH 3) and 3% whey protein isolate. Whey protein isolate-stabilized emulsions have previously been shown to have excellent physical and oxidative stability at pH 3.0 (15). The coarse emulsion was passed through a high-pressure valve homogenizer (Rannie Pro APV-Gaulin, Copenhagen, Denmark) at 5000 psi, for a total of four times. Between each cycle, the emulsion was collected in a beaker submerged in an ice-water bath.

Particle size distributions of the oil-in-water emulsions were measured using a Coulter LS 230 light scattering instrument (Coulter Corp., Miami, FL). The measurement of droplets is based on the principles of the Mie scattering theory. Droplet sizes were checked immediately after the emulsions had been homogenized using the method of McClements and Dungan (16). The emulsion droplet particle size was  $0.77 \pm 0.02 \mu\text{m}$ .

**Preparation of Surimi.** Dark muscle was removed from cod filets. Filets were ground through a 6 mm plate using a KitchenAid grinder (St. Joseph, MI). The ground mince was placed into 3 times its weight of distilled, deionized water of  $6^{\circ}\text{C}$ , stirred for 2 min, then allowed to settle for 13 min. The mince was drained through a fiberglass screen (16  $\times$  18 mesh) for 5 min. This wash/dewater cycle was repeated two additional times. Final dewatering to  $\sim 75\%$  moisture of the drained mince was accomplished using an ultracentrifuge model L-65B (Beckman, Palo Alto, CA) and a no. 19 rotor at 14000 rpm for 30 min (29300g). When used, 4% sucrose, 5% sorbitol, 0.5% sodium triphosphate (STPP), and EDTA were blended into the dewatered mince using a model R301 chilled food processor (Robot Coupe, Inc., Ridgeland, MS) for 30 s. The control surimi mix contained 4% sucrose, 5% sorbitol, 0.5% STPP, and 0.5% NaCl. To 237.7 g of surimi was added 12.3 g of emulsions by mixing for 10 min in a food processor to provide  $\sim 500$  mg of  $\omega$ -3 fatty acids per 85 g of final product (a single serving size). For the bulk oil, 3.7 g of oil was added directly to 246.3 g of surimi using the same mixing method as above.

**Determination of Fatty Acids.** Fatty acid methyl esters were prepared from the extracted lipid materials (17). Nonadecanoic acid (C19:0) in hexane (400  $\mu\text{L}$  of stock solution containing 2 mM) was added to 0.2 g of surimi gel as an internal standard. Fatty acid methyl ester analyses were performed with a Shimadzu GC-17 gas chromatograph, equipped with a split/splitless capillary inlet system and a flame ionization detector interfaced to class-VP chromatography data system (Shimadzu Co., Kyoto, Japan). A DB-Wax fused-silica capillary column (J&W Scientific, Palo Alto, CA; dimensions of 30 m  $\times$  0.32 mm i.d. and 0.25  $\mu\text{m}$  film thickness) was used for separation. Samples (1  $\mu\text{L}$ ) were injected in the split injection mode. Split ratio was 12:1. The oven temperature was held at  $180^{\circ}\text{C}$  for 2 min and subsequently increased at a rate of  $5^{\circ}\text{C}/\text{min}$  to  $240^{\circ}\text{C}$ . The injector and detector temperatures were 220 and  $240^{\circ}\text{C}$ , respectively. Peaks were identified by comparison of retention time with standards, previously characterized using gas chromatography and mass spectroscopy.

Table 1. Docosahexaenoic Acid Concentration of Surimi Treatments

surimi	C22:6 (mg/g of surimi) $\pm$ SD
control	$0.79 \pm 0.8^a$
+ emulsion	$6.98 \pm 1.1^b$
+ bulk oil	$6.18 \pm 0.9^c$

<sup>a, b, c</sup>Significantly different at  $p < 0.05$ .

**Formation of Surimi Gels.** Frozen surimi was tempered at refrigerated temperatures for 1 h, adjusted to between 79 and 80% moisture with distilled, deionized water, and chopped with 3% crystallized sodium chloride using a Robo-Coupe R301 Ultra chopper for 2 min. Equipment was prechilled, and chopping took place in a refrigerated room ( $6^{\circ}\text{C}$ ) to prevent the surimi from exceeding  $10^{\circ}\text{C}$  during chopping. Product was stuffed into stainless steel tubes (19 mm diameter  $\times$  175.1 mm) and sealed on both ends. The tubes were heated in water baths at  $90^{\circ}\text{C}$  for 20 min. Heated product was cooled in ice for  $\sim 15$ –30 min. Cooked gels were stored in polyethylene bags at  $6^{\circ}\text{C}$  for 48 h prior to analysis of gel strength and color and for up to 27 days for analysis of lipid oxidation products.

**Stress and Strain at Breakage.** Stress and strain at structural failure were determined on the gels using the procedure of Wu et al. (18). Sections of gels were machined to achieve a concave shape with a midsection diameter of 10 mm (19). The diameter was checked with a caliper, and adjustments to the shaper were performed if needed. Twisting of the gel was performed on a Haake rotovisco PK plate on a Brookfield DV-II viscometer. Rheological values as described by Lanier et al. (20) were obtained using a computer-aided software program linked to the computer and viscometer.

**Measurement of Color Values.** Color measurements were made with a Hunterlab Labscan II (Hunter Associates Laboratory, Inc., Reston, VA). A  $D_{65}$  illuminant was utilized, and reflected light was viewed at  $10^{\circ}$ . The port size was  $\approx 6$  mm. The instrument was standardized using a black tile and a white tile with the values  $L = 95.32$ ,  $a = -0.6$ , and  $b = 0.9$ . Whiteness index was determined as described in Lanier et al. (20), using the formula whiteness index =  $100 - [(100 - L)^2 + a^2 + b^2]^{0.5}$ .

**Determination of Lipid Oxidation Products.** Lipid hydroperoxides (LOOH) were extracted from 1 g of surimi gels with 10 mL of chloroform/methanol (2:1). After centrifugation (5 min at 2000g), an aliquot (2 mL) of the lower chloroform layer was mixed with an additional 1.3 mL of chloroform/methanol (2:1) and then reacted with 16.7  $\mu\text{L}$  each of 3.94 M ammonium thiocyanate and 0.072 M ferrous chloride (21). Absorbance was measured at 500 nm after 20 min of incubation, and LOOH were quantified on the basis of a standard curve prepared from cumene hydroperoxide. Thiobarbituric acid reactive substances (TBARS) in surimi were determined using a modified method of Srinivasan and Xiong (22) by mixing 1 g of surimi with 2.0 mL of 7.5% TCA, 0.1% propyl gallate, and 0.1% EDTA solution followed by centrifugation at 2000g for 5 min. A 1 mL aliquot of the supernatant was mixed with 1 mL of 0.02 M 2-thiobarbituric acid and then incubated for 20 min in a boiling water bath. Samples were centrifuged (5 min at 2000g), and the absorbance of the supernatant was read spectrophotometrically at 530 nm. TBARS were quantified on the basis of the molar extinction coefficient of  $1.56 \times 10^5 \text{ M}^{-1} \text{ cm}^{-1}$  malondialdehyde (23).

**Statistics.** All measurements were made on a minimum of three samples and reported as means  $\pm$  standard deviation. The general linear model procedure (24) was used to test for significance ( $p \leq 0.05$ ). Student's  $t$  test (24) was used to separate means.

## RESULTS AND DISCUSSION

**Docosahexaenoic Acid Concentrations.** The major  $\omega$ -3 fatty acid in the algal oil used in this study is DHA. As expected, DHA concentrations in the surimi increased upon addition of the algal oil (Table 1). The no-added-algal-oil surimi control contained 0.79 mg of DHA/g of surimi, presumably due to the presence of membrane lipids in the washed cod. Surimi with

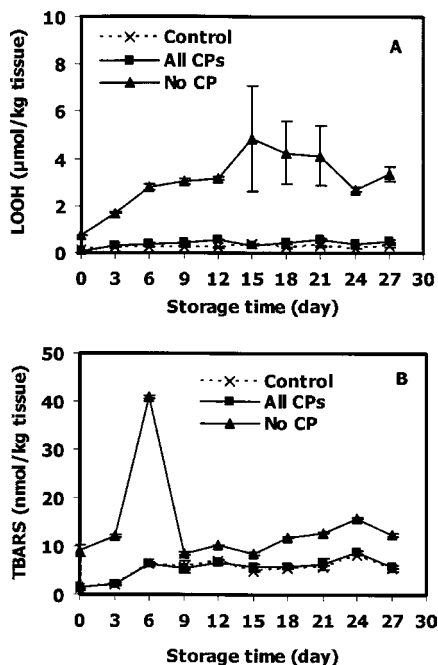


Figure 1. Lipid hydroperoxide (LOOH, A) and thiobarbituric acid reactive substances (TBARS, B) formation in control surimi, surimi + emulsion + all cryoprotectants (all CPs), and surimi + emulsion without cryoprotectants (no CPs). Data points represent means ( $n = 3$ )  $\pm$  standard deviation (some error bars are smaller than the data symbols).

oil-in-water emulsion contained significantly ( $p \leq 0.05$ ) more DHA than surimi with bulk oil. It is unclear why this difference occurred.

**Influence of Cryoprotectants on Lipid Oxidation in Surimi Containing Algal Oil Emulsions.** Polyphosphates are known to be excellent antioxidants, especially in muscle foods (14). Therefore, surimi may represent an excellent vehicle for oxidatively stable ω-3 fatty acids. A comparison of lipid oxidation rates of whey protein isolate-stabilized algal oil emulsions in surimi containing different cryoprotectants was made during the storage at 6 °C for 27 days. All samples contained 0.5% NaCl so that surimi gels could be produced.

Figure 1 shows the impact of cryoprotectants on the formation of lipid hydroperoxides and TBARS in surimi containing whey protein isolate-stabilized algal oil emulsions. Very little lipid oxidation was observed in surimi that did not contain the emulsified algal oil. Little lipid oxidation was also observed in emulsion-fortified surimi with all three cryoprotectants. However, when sucrose, sorbitol, and STPP were not added to the surimi, both lipid peroxides and TBARS were observed to increase after as little as 3 days of storage. This indicates that the cryoprotectants were able to protect the whey protein isolate-stabilized algal oil emulsion from oxidation. To determine which of the cryoprotectants was responsible for inhibiting oxidation, emulsion-containing surimi samples were prepared alternately, leaving one of the cryoprotectants out of the preparation. Of these treatments, the rapid formation of lipid hydroperoxides and TBARS was observed only in samples that did not contain STTP (Figure 2). This indicates that STTP was the main cryoprotectant exhibiting antioxidant activity.

The second study determined the influence of ω-3 fatty acid incorporation into surimi, either in the form of a whey protein isolate-stabilized oil-in-water emulsion or as bulk oil, on the physical properties and oxidative stability of the surimi. The ability of lipid soluble antioxidants to decrease lipid oxidation rates was also tested.

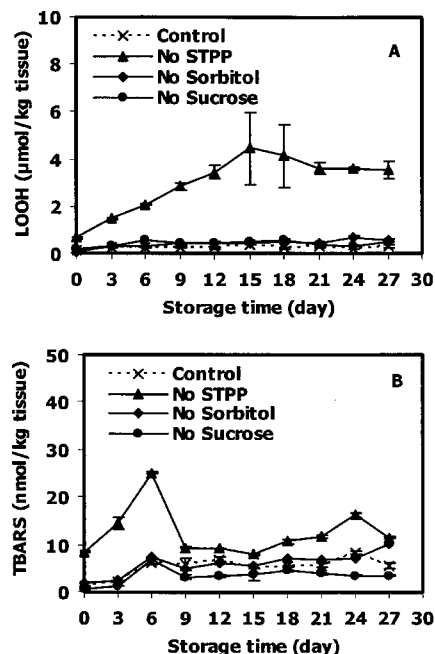


Figure 2. Lipid hydroperoxide (LOOH, A) and thiobarbituric acid reactive substances (TBARS, B) formation in control surimi, surimi + emulsion without STPP, surimi + emulsion without sorbitol, and surimi + emulsion without sucrose. Data points represent means ( $n = 3$ )  $\pm$  standard deviation (some error bars are smaller than the data symbols).

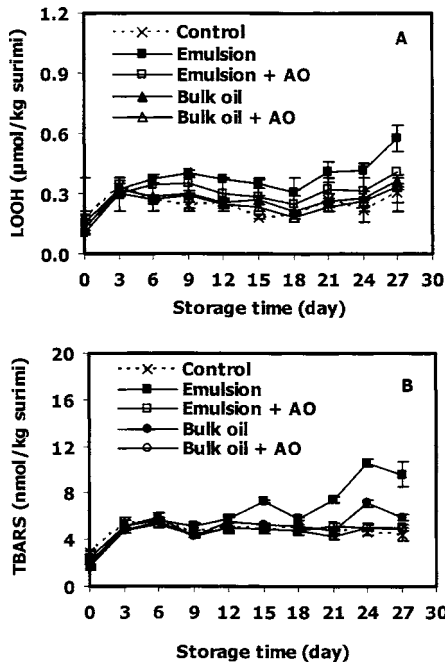
Table 2. Gel Strength and Hunter Colorimeter Values of Surimi with and without Emulsified or Bulk Algal Oil, Lipid Soluble Antioxidants (AO), and EDTA

surimi	strength (g·cm) $\pm$ SD	$b^*$ $\pm$ SD
control	175.9 <sup>a</sup> $\pm$ 8.8	4.8 <sup>a</sup> $\pm$ 0.2
+ emulsion	176.5 <sup>a</sup> $\pm$ 24.9	15.4 <sup>b</sup> $\pm$ 0.3
+ emulsion + EDTA (25 μM)	162.1 <sup>ab</sup> $\pm$ 24.7	14.8 <sup>cd</sup> $\pm$ 0.2
+ emulsion + AO	119.3 <sup>cd</sup> $\pm$ 8.2	15.2 <sup>bc</sup> $\pm$ 0.1
+ emulsion + AO + EDTA (25 μM)	148.8 <sup>abc</sup> $\pm$ 2.1	14.7 <sup>d</sup> $\pm$ 0.1
+ bulk oil	116.2 <sup>d</sup> $\pm$ 15.9	12.4 <sup>e</sup> $\pm$ 0.3
+ bulk oil + AO	132.5 <sup>bcd</sup> $\pm$ 16.6	12.6 <sup>e</sup> $\pm$ 0.1

<sup>a, b, c, d, e</sup>Significantly different at  $p < 0.05$ .

Gel strength, calculated on the basis of the force multiplied by deformation using a unit of g·cm, is used by the surimi industry as a symbol of quality (12). Addition of the emulsion to the surimi did not decrease gel strength compared to the control (no added lipid) surimi ( $p > 0.05$ , Table 2). This is in contrast to surimi with added bulk oil that had significantly lower gel strength than the control surimi and surimi with emulsion ( $p < 0.05$ ). The presence of lipid soluble antioxidants in the emulsion resulted in a significant decrease in gel strength compared to the control. This was not true in the surimi plus bulk oil, for which gel strength was statistically the same in both the presence and absence of lipid soluble antioxidants.

Hydrophobic interactions are important in the formation and stabilization of surimi gels (12). A decrease in gel strength in surimi with bulk oil compared to surimi with emulsified oil could be due to the bulk oil interfering with hydrophobic interaction between proteins during gelation. Because the emulsified oil is surrounded by whey proteins, this oil may not have access to the hydrophobic regions of fish myofibrillar proteins and thus would not interfere with gelation. The presence of lipid soluble antioxidants decreased gel strength in surimi with emulsion but had no influence on surimi with bulk oil.

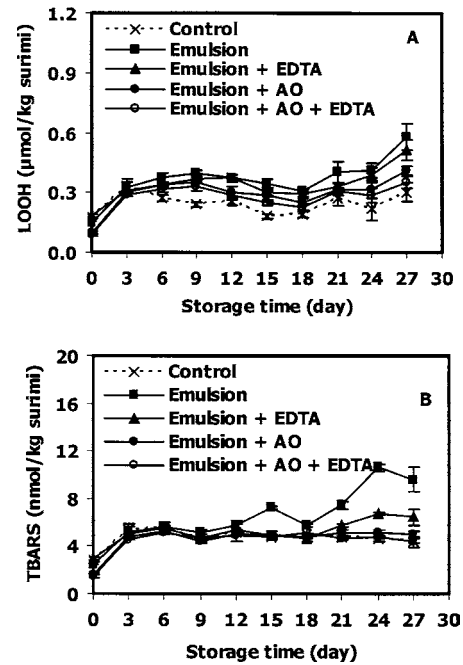


**Figure 3.** Lipid hydroperoxide (LOOH, A) and thiobarbituric acid reactive substances (TBARS, B) formation in control surimi, surimi + emulsion, surimi + emulsion + antioxidants (AO), surimi + bulk oil, and surimi + bulk oil + AO. Data points represent means ( $n = 3$ )  $\pm$  standard deviation (some error bars are smaller than the data symbols).

Antioxidants are surface active, so it is also possible that they could interfere with hydrophobic interaction in the protein gel. It is unclear why the lipid soluble antioxidants would affect gel strength in the presence of emulsion but not bulk oil unless the bulk oil's interference with hydrophobic interactions could not be further increased by the presence of antioxidants.

**Color.** All surimi treatments containing algal oil had an increase in  $L^*$ ,  $a^*$ , and  $b^*$  values. The largest change in color in surimi with algal oil was in the  $b^*$  values (yellowness); thus, the  $L^*$  and  $a^*$  values are not shown. The  $b^*$  values increased from 4.8 in the control to  $>14.0$  in emulsion-containing surimi and  $>12.0$  in surimi with bulk oil (Table 2). The change in  $b^*$  values was significantly greater in surimi with emulsion than in surimi with bulk oil. This larger increase in yellowness could be due to the more even distribution of the small emulsion droplets throughout the surimi. It was not surprising that the surimi with algal oil was yellower because microalgal oils typically contain carotenoids that produce yellow-orange colors (25).

**Lipid Oxidation in Surimi with Emulsified and Bulk Algal Oil.** Both LOOH and TBARS values were lower in surimi containing bulk oil compared to surimi with emulsified oil after 6 and 15 days, respectively (Figure 3). Algal oil emulsified with whey protein isolate could have lower oxidative stability due to its increased surface area. Homogenization of the whey protein isolate-stabilized algal oil-in-water emulsions by four passes at 5000 psi resulted in particle sizes of  $0.77 \pm 0.02 \mu\text{m}$ . It is unlikely that the bulk oil in the surimi would have such small particle sizes because it was introduced into the surimi during bowl chopping, conditions that result in much lower shear stress that would be less effective at decreasing lipid particle size compared to homogenization. The higher surface area of the emulsified lipid could lead to more interactions between aqueous phase prooxidants (e.g., iron) and lipid substrates. An additional factor would be that the surface charge of the whey protein isolate-stabilized emulsion droplets is negative at pH



**Figure 4.** Lipid hydroperoxide (LOOH, A) and thiobarbituric acid reactive substances (TBARS, B) formation in control surimi, surimi + emulsion, surimi + emulsion + EDTA, surimi + emulsion + antioxidants (AO), and surimi + emulsion + AO + EDTA. Data points represent means ( $n = 3$ ) plus standard deviation (some error bars are smaller than the data symbols).

values above the  $pI$  of the whey proteins (5.1; 26). Because the pH of the surimi was 6.9–7.1 (data not shown), the emulsion droplets would be anionic and thus could attract prooxidative metals, thus decreasing the oxidative stability of the emulsified lipid (26).

In an attempt to improve the oxidative stability of the algal oil in the surimi, oil containing lipid soluble antioxidants was tested (Figure 3). Addition of lipid soluble antioxidants to the bulk oil resulted in very little change in LOOH concentrations in the surimi during the 27 days of storage. Lipid soluble antioxidants in the bulk oil also did not have any effect on TBARS formation in the surimi until after 24 days of storage. The presence of lipid soluble antioxidants in the emulsified oil decreased LOOH concentrations in the surimi (7.4–34.1%) with significant differences ( $p < 0.05$ ) at 9, 12, 15, 21, 24, and 27 days of storage. Lipid soluble antioxidants in the emulsified oil also decreased TBARS concentrations in the surimi with significant ( $p < 0.05$ ) inhibition occurring after 9 days of storage. The apparent lack of effectiveness of the lipid soluble antioxidants in the surimi containing bulk oil is likely due to the fact that very little lipid oxidation had occurred during storage, thus making it difficult to observe an antioxidant effect.

EDTA has previously been shown to be very effective at inhibiting lipid oxidation in oil-in-water emulsions (27–29). Therefore, its ability to protect the emulsified algal oil in the surimi was tested (Figure 4). Compared to surimi containing only emulsion, EDTA (25  $\mu\text{M}$ ) caused small decreases in LOOH concentrations ranging from 1.1 to 19.3% that were significantly different at 9, 15, and 21 days of storage. EDTA also significantly decreased TBARS in the surimi with emulsified oil after 15 days of storage. EDTA had no impact on the gel strength of the surimi containing emulsified algal oil (Table 2). The combination of EDTA and lipid soluble antioxidants in the emulsified oil was better than EDTA alone, with LOOH being lower in the surimi at 6, 12, 15, 18, and 27 days of storage

and TBARS being lower in the surimi after 21 days of storage. Neither surimi prepared with emulsion plus EDTA and lipid soluble antioxidants nor surimi prepared with emulsion with lipid soluble antioxidant alone had any differences in lipid hydroperoxide or TBARS formation during the entire storage period; therefore, it was impossible to determine if the presence of EDTA was able to improve the activity of the lipid soluble antioxidants.

**Conclusions.** Incorporation of  $\omega$ -3 fatty acids from algal oil into surimi products could produce a functional food product that mimics natural fish products. STPP used as a cryoprotectant in surimi was found to be an effective inhibitor of the oxidation of  $\omega$ -3 fatty acids. Addition of  $\omega$ -3 fatty acids into the surimi in the form of an oil-in-water emulsion did not impact gel strength compared to bulk oil that caused significant gel weakening. However, emulsified  $\omega$ -3 fatty acids in the surimi were more susceptible to oxidation than bulk oil. Lipid soluble antioxidants were more effective than EDTA at decreasing  $\omega$ -3 fatty acid oxidation. These results suggest that surimi containing nutritionally beneficial  $\omega$ -3 fatty acids could be developed with good oxidative stability and gel strength.

#### LITERATURE CITED

- (1) Nair, S. S. D.; Leitch, J. W.; Falconer, J.; Garg, M. L. Prevention of cardiac arrhythmia by dietary ( $n$ -3) polyunsaturated fatty acids and their mechanism of action. *J. Nutr.* **1997**, *127*, 383–393.
- (2) Cheung, P. C. K.; Leung, A. Y. H.; Ang, P. O., Jr. Comparison of supercritical carbon dioxide and Soxhlet extraction of lipids from a brown seaseed, *Sargassum hemiphyllum* (Turn.) C. Ag. *J. Agric. Food Chem.* **1998**, *46*, 4228–4232.
- (3) Simopoulos, A. P. Omega-3 fatty acids in health and disease. In *Nutrition and Aging*; Liss, A. R., Ed.; Liss: New York, 1990; pp 129–156.
- (4) Kinsella, J. E. Food components with potential therapeutic benefits: the  $n$ -3 polyunsaturated fatty acids of fish oils. *Food Technol.* **1986**, *40*, 89–146.
- (5) Nawar, W. W. Lipids. In *Food Chemistry*, 3rd ed.; Fennema, O. R., Ed.; Dekker: New York, 1996; pp 225–319.
- (6) Kubow, S. Lipid oxidation products in foods and atherogenesis. *Nutr. Rev.* **1993**, *51*, 33–40.
- (7) Decker, E. A.; Xu, Z. Minimizing rancidity in muscle foods. *Food Technol.* **1998**, *52*, 54–59.
- (8) Fritsch, C. W. Lipid oxidation—the other dimensions. *Int. News Fats, Oils Relat. Mater.* **1994**, *5*, 423–436.
- (9) McClements, D. J.; Decker, E. A. Lipid oxidation in oil-in-water emulsions: impact of molecular environment on chemical reactions in heterogeneous food system. *J. Food Sci.* **2000**, *65*, 1270–1282.
- (10) Mei, L. Y.; McClements, D. J.; Wu, J. N.; Decker, E. A. Iron-catalyzed lipid oxidation in emulsion as affected by surfactant, pH and NaCl. *Food Chem.* **1998**, *61*, 307–312.
- (11) Mancuso, J. R.; McClements, D. J.; Decker, E. A. The effect of surfactant type, pH, and chelators on the oxidation of salmon oil-in-water emulsions. *J. Agric. Food Chem.* **1999**, *47*, 4112–4116.
- (12) MacDonald, G. A.; Lanier, T. C.; Carvajal, P. A. Stabilization of proteins in surimi. In *Surimi and Surimi Seafood*; Park, J. W., Ed.; Dekker: New York, 2000; pp 91–125.
- (13) Sultanbawa, Y.; Li-Chan, E. C. Y. Structural changes in natural actomyosin and surimi from ling cod (*Ophiodon elongates*) during frozen storage in the absence or presence of cryoprotectants. *J. Agric. Food Chem.* **2001**, *49*, 4716–4725.
- (14) Erickson, M. C. Antioxidants and their application to frozen foods. In *Quality in Frozen Food*; Erickson, M. C., Hung, Y. C., Eds.; Chapman and Hall: New York, 1997; pp 233–263.
- (15) Hu, M.; McClements, D. J.; Decker, E. A. Impact of whey protein emulsifiers on the oxidative stability of salmon oil-in-water emulsions. *J. Agric. Food Chem.* **2003**, *51*, 1435–1439.
- (16) McClements, D. J.; Dungan, S. R. Effect of colloidal interactions on the rate of interdroplet heterogeneous nucleation in oil-in-water emulsions. *J. Colloid Interface Sci.* **1997**, *186*, 17–28.
- (17) AOCS. *Official Methods and Recommended Practices of the American Oil Chemists' Society*, 5th ed.; AOCS: Champaign, IL, 1998.
- (18) Wu, M. C.; Hamann, D. D.; Lanier, T. C. Rheological and calorimetric investigations of starch–fish protein systems during thermal processing. *J. Texture Stud.* **1985**, *16*, 53–74.
- (19) Montejano, J. C.; Hamann, D. D.; Lanier, T. C. Final strengths and rheological changes during processing of thermally induced fish muscle gels. *J. Rheol.* **1983**, *27*, 557–579.
- (20) Lanier, T. C. Measurement of surimi composition and functional properties. In *Surimi Technology*; Lanier, T. C., Lee, C. H., Eds.; Dekker: New York, 1992; pp 123–163.
- (21) Shantha, N. C.; Decker, E. A. Rapid, sensitive, iron-based spectrophotometric methods for determination of peroxide values of food lipids. *J. AOAC Int.* **1994**, *77*, 421–424.
- (22) Srinivasan, S.; Xiong, Y. L. Sodium chloride-mediated lipid oxidation in beef heart surimi-like material. *J. Agric. Food Chem.* **1996**, *44*, 1697–1703.
- (23) McDonald, R. E.; Hultin, H. O. Some characteristics of the enzymic lipid peroxidation system in the microsomal fraction of flounder skeletal muscle. *J. Food Sci.* **1987**, *52*, 15–21.
- (24) Snedecor, G. W.; Cochran, G. W. *Statistical Methods*, 8th ed.; Iowa State University Press: Ames, IA, 1989.
- (25) Tonegawa, I.; Okada, S.; Murakami, M.; Yamaguchi, K. Pigment composition of the green microalga. *Fish. Sci.* **1998**, *64*, 305–308.
- (26) Hu, M.; McClements, D. J.; Decker, E. A. Lipid oxidation in corn oil-in-water emulsions stabilized by casein, whey protein isolate, and soy protein isolate. *J. Agric. Food Chem.* **2003**, *51*, 1696–1700.
- (27) Mancuso, J. R.; McClements, D. J.; Decker, E. A. Ability of iron to promote surfactant peroxide decomposition and oxidize  $\alpha$ -tocopherol. *J. Agric. Food Chem.* **1999**, *47*, 4146–4149.
- (28) Mei, L.; Decker, E. A.; McClements, D. J. Evidence of iron association with emulsion droplets and its impact on lipid oxidation. *J. Agric. Food Chem.* **1998**, *46*, 5072–5077.
- (29) Jacobsen, C.; Hartvigsen, K.; Thomsen, M. K.; Hansen, L. F.; Lund, P.; Skibsted, L. H.; Holmer, G.; Adler-Nissen, J.; Meyer, A. S. Lipid oxidation in fish oil enriched mayonnaise: calcium disodium ethylenediaminetetraacetate, but not gallic acid, strongly inhibited oxidative deterioration. *J. Agric. Food Chem.* **2001**, *49*, 1009–1019.

Received for review June 30, 2003. Revised manuscript received December 2, 2003. Accepted December 4, 2003.

JF034703M