

## Ability of Lipid Hydroperoxides To Partition into Surfactant Micelles and Alter Lipid Oxidation Rates in Emulsions

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Lipid hydroperoxides are important factors in lipid oxidation due to their ability to decompose into free radicals. In oil-in-water emulsions, the physical location of lipid hydroperoxides could impact their ability to interact with prooxidants such as iron. Interfacial tension measurements show that linoleic acid, methyl linoleate, and trilinolein hydroperoxides are more surface-active than their non-peroxidized counterparts. In oil-in-water emulsion containing surfactant (Brij 76) micelles in the continuous phase, linoleic acid, methyl linoleate, and trilinolein hydroperoxides were solubilized out of the lipid droplets into the aqueous phase. Brij 76 solubilization of the different hydroperoxides was in the order of linoleic acid > trilinolein ≥ methyl linoleate. Brij 76 micelles inhibited lipid oxidation of corn oil-in-water emulsions with greater inhibition of oxidation occurring in emulsions containing linoleic acid hydroperoxides. Surfactant solubilization of lipid hydroperoxides could be responsible for the ability of surfactant micelles to inhibit lipid oxidation in oil-in-water emulsions.

**KEYWORDS:** Lipid oxidation; lipid hydroperoxides; surfactant; emulsion; micelles

### INTRODUCTION

Small molecule surfactants are commonly used in the food industry to stabilize oil-in-water emulsions (1). After a surfactant concentration is reached that saturates the droplet surface, excess surfactant molecules can form micelles in the continuous phase (2). It is well-known that micelles have the ability to solubilize certain compounds out of the lipid droplets into the continuous phase of the emulsion (3).

Several studies have shown that antioxidants can partition into different physical locations in emulsions, and this partitioning dramatically influences antioxidant effectiveness rates (4–7). Previous work has shown that in oil-in-water emulsions, surfactants, can influence the physical location of antioxidants in oil-in-water emulsions by causing solubilization of lipid-soluble antioxidants into the aqueous phase (8). Excess Brij micelles in an oil-in-water emulsion were found to increase the partitioning of phenolics into the continuous phase with polar antioxidants (propyl gallate) partitioning more than nonpolar antioxidants (butylated hydroxytoluene, BHT).

Little is known about the partitioning of lipid hydroperoxides into the aqueous phase and how lipid hydroperoxide location would influence the oxidative stability of food emulsions. Lipid hydroperoxides are more polar than the lipids from where they originated due to the presence of oxygen. It has been speculated

that the higher polarity of lipid hydroperoxides would cause them to diffuse toward the water–lipid interface of emulsions. The existence of the lipid hydroperoxides at the emulsion droplet interface suggests that surfactant micelles may solubilize these important lipid oxidation substrates out of the lipid droplets. This could prevent free radicals formed by hydroperoxide decomposition from attacking unsaturated lipids in the lipid droplet core, thus increasing the oxidative stability of emulsions. Alternatively, micelles could facilitate the transport of hydroperoxides from one emulsion droplet to the other, therefore accelerating oxidation rates.

The aim of this research was to study the influence of continuous phase surfactant micelles on the partitioning behavior of lipid hydroperoxides in oil-in-water emulsions. The effect of lipid hydroperoxide partitioning on the oxidative stability of food emulsions was also evaluated.

### MATERIALS AND METHODS

Polyoxyethylene 10 stearyl ether (Brij 76, average molecular weight = 711 g/mol), imidazole, sodium acetate, iminodiacetic acid (Chelex 100), ethylenediaminetetraacetic acid (EDTA), linoleic acid, methyl linoleate, trilinolein, Rose Bengal, and hexadecane were purchased from Sigma Chemical Co. (St. Louis, MO). Trilinolein produced only one spot on silica gel thin layer chromatography using hexane/ethyl ether/acetic acid (80:30:1 v/v) as the mobile phase, showing that there were no free fatty acids detected. Corn oil was purchased from a local store. All other reagents were of analytical grade or purer. Glassware was acid washed (2 N HCl), rinsed with double-distilled water, and dried overnight at 110 °C before use.

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**Buffer Treatment with Chelex-100.** Transition metals were removed from buffers (except in the oxidation studies) by gentle mixing of 2 g of Chelex 100/L of buffer for 24 h. Buffer was then separated from precipitated Chelex 100 by decantation (9).

**Methods. Peroxidation of Lipids.** Hydroperoxides were produced from different sources by mixing ~1 g of linoleic acid, methyl linoleate, or trilinolein and 0.05 g of Rose Bengal, a singlet oxygen generator. The mixture was incubated in a water bath at 6 °C and illuminated with a 90 W lamp for different periods of time (linoleic acid and methyl linoleate, 10 h; trilinolein, 32 h) to produce a hydroperoxide concentration of ~0.1 mmol/g of lipid (2.9, 2.8, and 8.8 mol % for linoleic acid, methyl linoleate, and trilinolein, respectively). Headspace hexanal (see Oxidation Studies for methodology) was not detected in any sample, indicating that minimal hydroperoxide breakdown occurred during hydroperoxide formation.

**Interfacial Tension Measurement.** Interfacial tension was determined using a digital tensiometer K 10 ST (Kruss USA, Charlotte, NC) equipped with a platinum–iridium Du Noüy ring. Oil phase (40 g with or without 0.5 mM hydroperoxides) and double-distilled water (40 g) were placed in the measuring vessel and allowed to equilibrate at 25 °C. Preliminary experiments showed that the interfacial tension decreased during the first 8 h and then it reached a plateau; therefore, measurements were taken after 10 h. Control samples contained all of the components except for the hydroperoxides.

**Emulsion Preparation.** Stock emulsions containing 10% oil phase and 90% aqueous phase were prepared using a hand-held Bio Homogenizer M133/1281-0 (Biospec Products Inc., Bartlesville, OK) on speed setting 1 for 30 s, followed by 60 s of sonication using a Braun-Sonic 2000 U ultrasonic generator (Braun Biotech, Allentown, PA) equipped with a 5T standard probe at a power setting of +250 and a 0.5 s repeating duty cycle. Peroxidized lipids (0.6 g) were added to corn oil (14.4 g) immediately prior to emulsion formation. The aqueous phase of the emulsion consisted of Brij 76 (34 mM) and EDTA (0.1 mM, except for the oxidation studies, in which no EDTA was added) dissolved in acetate–imidazole buffer (10 mM each, pH 7). EDTA was added to minimize lipid hydroperoxide decomposition. Stock emulsions were diluted 1:1 with Brij 76 solutions (0, 15, 45, 70, or 100 mM; final concentrations) containing 0.1 mM EDTA and acetate–imidazole buffer (10 mM each, pH 7) to give 5% oil-in-water emulsions.

Particle size distributions were measured using a Coulter LS 230 light scattering particle size analyzer (Coulter Corp., Miami, FL). The median droplet diameters ranged from 1.63 to 1.89  $\mu\text{m}$  and did not change during the course of the experiments.

**Partitioning Studies.** Emulsions (56 g each) were incubated at room temperature in a shaker. Every hour, 10 g of emulsion was centrifuged at 24000g for 35 min, and 0.2 mL of the continuous phase was removed to determine its hydroperoxide concentration. Additionally, 0.3 mL of unseparated emulsion was added to 1.5 mL of a mixture of isooctane/2-propanol (3:1), vortexed three times for 10 s, followed by centrifuging at 2000g for 2 min. Then 0.2 mL of the organic phase was used to measure total hydroperoxide concentration in the emulsions. Partitioning in a bilayer system was tested using hexadecane or corn oil (with or without 0.5 mM hydroperoxides) and double-distilled water at a ratio of 1:1. Preliminary experiments showed that lipid hydroperoxide concentrations in the two phases of the bilayer system reached equilibrium after 8 h; therefore, the bilayer systems were allowed to equilibrate at room temperature for 10 h, and hydroperoxide concentrations were determined in 0.2 mL of the oil phase.

Continuous phase and total lipid hydroperoxides or oil phase hydroperoxides (bilayers) were determined using a method adapted from that of Shanta and Decker (10). Sample (0.2 mL) was added to a mixture of methanol/butanol (2:1) followed by addition of 15  $\mu\text{L}$  of 3.94 M thiocyanate and 15  $\mu\text{L}$  of 0.072 M  $\text{Fe}^{2+}$ . The solution was vortexed, and after 20 min, the absorbance at 510 nm was measured using a spectrophotometer. The concentration of hydroperoxides was calculated from a cumene hydroperoxide standard curve.

**Oxidation Studies.** Emulsions consisting of 10% oil phase (14.4 g of corn oil + 0.6 g of linoleic acid or peroxidized linoleic acid, final hydroperoxide concentration in lipid of 4.5 mM) and 90% aqueous phase (34 mM Brij 76 in acetate imidazole buffer, pH 7.0) were prepared as described under Emulsion Preparation followed by dilution

**Table 1.** Interfacial Tension of Hexadecane/Water Bilayers (1:1 Ratio) Containing Different Forms of Linoleic Acid and Hydroperoxides

oil phase composition	control (mN/m)	with hydroperoxide (mN/m)	reduction %
hexadecane (H)	44.2		
H + Rose Bengal (RB)	34.6		
H + RB + linoleic acid	20.3	13.6	33.3
H + RB + methyl linoleate	36.5	21.0	42.6
H + RB + trilinolein	25.5	17.4	32.0

with buffer or 200 mM Brij 76 solution to a final concentration of 5% oil and 100 mM continuous phase Brij 76. The samples and the controls (1 mL) were immediately placed in 10 mL glass vials, sealed with polytetrafluoroethylene (PTFE)/butyl rubber septa using a crimper and aluminum seals, and incubated at 37 °C in the dark. Oxidation was followed by measuring hydroperoxides (as described above) and headspace hexanal.

Headspace hexanal was determined according to a method described by Mancuso et al. (11) using a Shimadzu GC-17A gas chromatograph (Columbia, MD) with a Hewlett-Packard 19395A headspace sampler (Avondale, PA) coupled to a computer. The chromatograms were integrated using Shimadzu CLASS-VP chromatography data system software. The headspace conditions were as follows: sample temperature, 55 °C; sample loop and transfer line temperature, 110 °C; pressurization, 10 s; venting, 10 s; injection, 1 min. The aldehydes were separated isothermally at 65 °C on an HP methyl silicone (DB-1) fused silica capillary column (50 m, 0.31 mm i.d., 1.03  $\mu\text{m}$  film thickness). The splitless injector temperature was 180 °C, and the flame ionization detector temperature was 250 °C. Concentrations were determined from peak areas using a standard curve made from authentic hexanal because it was determined that the surfactant micelles did not influence the amount of hexanal partitioning into the headspace.

**Statistics.** All of the experiments were done in triplicate. Statistical analyses were performed using Student's *t* test (12). Statistical differences were defined as  $p \leq 0.05$ .

## RESULTS

### Influence of Lipid Hydroperoxides on Interfacial Tension.

The ability of lipids and lipid hydroperoxides to alter interfacial tension was determined in a hexadecane/water bilayer to which unperoxidized and peroxidized lipids were added to the hexadecane (Table 1). Hexadecane was chosen because it is completely saturated; therefore, it cannot further oxidize and produce hydroperoxides. In addition, unlike vegetable oils, hexadecane would contain minimal concentrations of surface active materials (e.g., free fatty acids and mono- and diacylglycerols) that could compete with the lipid hydroperoxides for the oil–water interface. Because Rose Bengal was in the peroxidized lipids, its surface activity was also tested and found to decrease interfacial tension 22%, and because Rose Bengal was surface active, it was added to all other interfacial measurements. Among the nonperoxidized lipids, linoleic acid produced the lowest interfacial tension followed by trilinolein and methyl linoleate (Table 1). The observation that the free fatty acid had the most amphiphilic nature is not surprising considering its unesterified hydrophilic carboxylic headgroup. The fact that trilinolein had a lower interfacial tension than methyl linoleate is somewhat unexpected. This could be due to the presence of impurities in the trilinolein (e.g., free fatty acids and mono- or diacylglycerols). However, only one spot corresponding to triacylglycerols was observed by thin layer chromatography, meaning that no such compounds could be detected. However, it is possible that there were some contaminants present below the detection limit, because the method used to measure interfacial tension is very sensitive to the presence of impurities.

**Table 2.** Partitioning Coefficients in Hexadecane or Corn Oil/Water Bilayers (1:1 Ratio) Containing Different Types of Hydroperoxides

oil phase composition	% hydroperoxide in water
hexadecane + linoleic acid hydroperoxides	8.5a ± 0.3
hexadecane + methyl linoleate hydroperoxides	4.9b ± 0.9
hexadecane + trilinolein hydroperoxides	2.1c ± 0.9
corn oil + linoleic acid hydroperoxides	1.3c ± 0.1
corn oil + methyl linoleate hydroperoxides	0.9c ± 0.2
corn oil + trilinolein hydroperoxides	1.9c ± 1.1

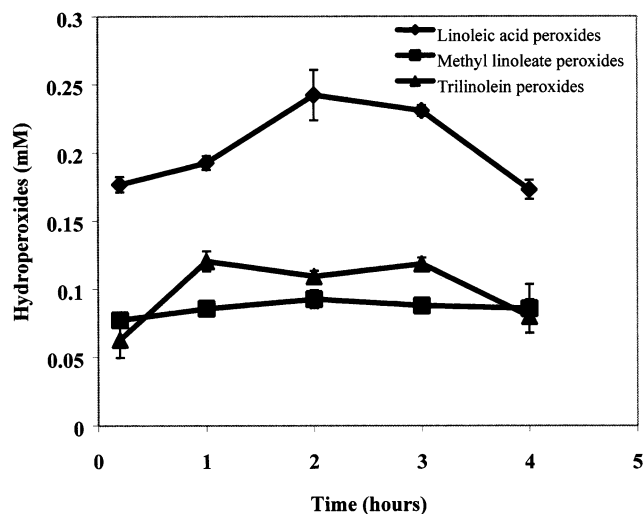
<sup>a</sup> All samples contained Rose Bengal (0.2 mM) to match conditions used in the interfacial tension measurements (Table 1). Reported values are average ± SD ( $n = 3$ ). Samples with different letters are significantly different ( $\alpha = 0.05$ ).

In general, all of the different types of peroxidized lipids (free fatty acid, methyl ester, and triacylglycerol) tested were able to reduce interfacial tension more than their nonperoxidized counterparts (Table 1). A comparison among the different hydroperoxides showed that linoleic acid hydroperoxides resulted in the lowest absolute interfacial tension, followed by trilinolein and methyl linoleate hydroperoxides. Methyl linoleate hydroperoxides resulted in the highest reduction of interfacial tension compared to its nonoxidized counterpart (42.6% compared with the nonoxidized methyl linoleate control) followed by linoleic acid and trilinolein hydroperoxides, which decreased interfacial tension 33.3 and 32.0%, respectively, compared to their nonoxidized counterparts.

In addition to the surface activity of the lipids, another factor that could affect the ability of lipid hydroperoxides to decrease interfacial tension would be their ability to partition into the water phase (e.g., high water solubility would decrease the amount of hydroperoxide in the lipid and at the lipid–water interface). To test the ability of hydroperoxides to partition out of the lipid to the aqueous phase in the absence of surfactants, the amount of aqueous phase hydroperoxides in hexadecane or corn oil and water (1:1 ratio) bilayer systems was determined. Overall, the amount of lipid hydroperoxides that partitioned into the aqueous phase in the absence of surfactant micelles was very low (0.9–8.5%, 0.025–0.062 mM). Partitioning of different lipid hydroperoxides into the water layer of the hexadecane/water bilayer system was significantly different and was in the following order: trilinolein < methyl linoleate < linoleic acid (Table 2). Linoleic acid hydroperoxides decreased the interfacial tension more than the other hydroperoxides (Table 1) despite having the highest concentration in the aqueous phase (Table 2).

The physical location of the lipid hydroperoxides might also be altered by the polarity of the lipid. In a corn oil/water bilayer significant differences in water partitioning among trilinolein, linoleic acid, and methyl linoleate hydroperoxides were not observed (Table 2). In addition, partitioning of linoleic acid and methyl linoleate hydroperoxides into the water phase in the corn oil system was lower than in the hexadecane/water bilayer. Trilinolein hydroperoxide concentration in the water layer was not significantly different between the two biphasic systems tested (hexadecane/water and corn oil/water). The lower partitioning of the linoleic and methyl linoleate hydroperoxide into the water phase of the corn oil system could be due to the fact that corn oil has a higher polarity than hexadecane. The higher polarity of corn oil could increase the solubility of the linoleic acid and methyl linoleate hydroperoxides in the oil phase, thereby decreasing partitioning into the water phase.

**Influence of Surfactant Micelles on Lipid Hydroperoxide Solubilization.** The concentrations of linoleic acid, methyl

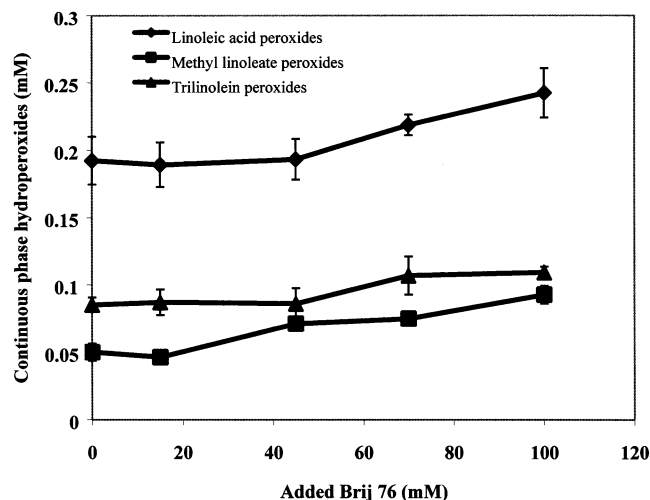


**Figure 1.** Hydroperoxide (linoleic acid, methyl linoleate, or trilinolein; 0.45 mM hydroperoxide) solubilization into the continuous phase of 5% corn oil-in-water emulsions containing 100 mM Brij 76. Data markers represent average ± SD ( $n = 3$ ).

linoleate, and trilinolein hydroperoxides in the continuous phase were measured as a function of time in corn oil-in-water emulsions to which 100 mM Brij 76 was added after emulsion preparation. In general, all of the hydroperoxides solubilized rapidly into the aqueous phase (Figure 1) with the majority of partitioning occurring by the first sampling time (10 min). After the initial increase, linoleic acid hydroperoxide concentration in the continuous phase increased further during the first 2 h (1.4-fold compared to the 10 min sample) and then decreased to reach a concentration at 4 h not significantly different from the 10 min value. The trilinolein hydroperoxides showed a similar behavior, with their concentration initially increasing during the first hour (1.9-fold compared to the 10 min sample) but then decreasing so that there was no significant difference between the 10 min and 4 h samples. In the case of methyl linoleate hydroperoxides, concentration in the continuous phase remained fairly constant during the course of the experiment (Figure 1). Although it is unclear why continuous phase linoleic acid and trilinolein hydroperoxides decreased during the latter stages of incubation, this was not due to hydroperoxide breakdown or formation because total hydroperoxide concentrations did not change and headspace hexanal was not detected (e.g., hydroperoxides were not breaking down) during the course of the experiment (data not shown).

The different types of hydroperoxides (methyl ester, triacylglycerol, or free fatty acid) showed different degrees of Brij 76 solubilization into the continuous phase of the water-in-oil emulsion (Figure 1). Linoleic acid hydroperoxides were solubilized the most, with continuous phase concentrations being 2.8-, 2.2-, and 2.2-fold greater than the trilinolein hydroperoxides after 0.2, 2, and 4 h, respectively. Continuous phase trilinolein and methyl linoleate hydroperoxides were not significantly different from each other at 0.2, 2, and 4 h.

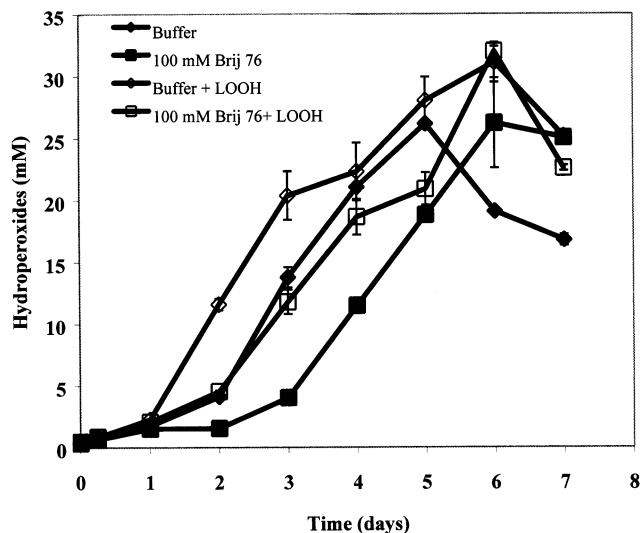
The influence of different concentrations of added micelles on the solubilization of hydroperoxides into the continuous phase of a corn oil-in-water emulsion was also studied (Figure 2). Stock emulsions (10% oil) were diluted with various concentrations of Brij 76 to give final concentrations of 5% oil and 0–100 mM Brij 76. Again, different types of hydroperoxides were tested: a free fatty acid (linoleic acid) and two esters (methyl linoleate and trilinolein). Emulsions containing no excess surfactant had significant levels of lipid hydroperoxides in the



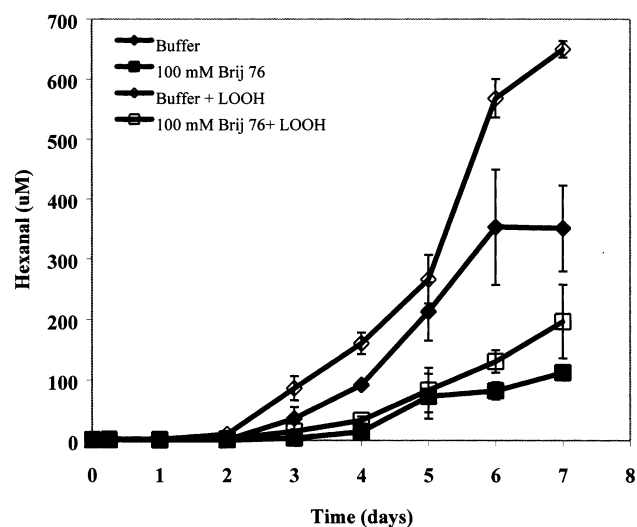
**Figure 2.** Influence of Brij 76 concentration on solubilization of linoleic acid, methyl linoleate, or trilinolein hydroperoxides (final concentration = 0.45 mM) into the continuous phase of a 5% corn oil-in-water emulsion. Continuous phase hydroperoxide concentrations were measured after 2 h of incubation. Data markers represent average  $\pm$  SD ( $n = 3$ ).

continuous phase. This suggests that the hydroperoxides could partition into the continuous phase without the aid of surfactant micelles. However, previous experiments in which the solubility of the lipid hydroperoxides was evaluated in a corn oil/water bilayer showed that the water solubility of the hydroperoxides was very low (Table 2). In this bilayer system the surface area of the lipid-water interface would be much smaller than in an emulsion. In an attempt to mimic what would happen to the water solubility of the linoleic acid hydroperoxides during the formation of oil-in-water emulsions, 5% corn oil containing linoleic acid hydroperoxides was sonicated in buffer without Brij 76 and then linoleic acid hydroperoxide concentrations were determined in the continuous phase. In this system, 0.05 mM linoleic acid hydroperoxides were found to partition into the continuous phase (data not shown). This compares to 0.19 mM linoleic acid hydroperoxides in the continuous phase of the corn oil-in-water emulsion prepared with 17 mM Brij 76 (Figure 2). The difference in the amount of continuous phase linoleic acid hydroperoxides in the emulsions prepared in the presence and absence of Brij 76 is likely to be due to Brij 76 micelles. The critical micelle concentration (CMC) of Brij 76 is very low ( $< 1$  mM; 13), which means that the surfactant in the emulsion that is not associated with the emulsion droplet interface would likely form surfactant micelles in the continuous phase. Previous work in similar oil-in-water emulsion systems has found that 40–60% of the surfactant used to prepare the emulsion remains in the continuous phase (8). Therefore, it is possible that this aqueous phase surfactant could form micelles and be responsible for the observed partitioning of lipid hydroperoxides into the continuous phase in the absence of added Brij 76 (Figure 2).

Increasing Brij 76 micelle concentrations increased lipid hydroperoxide solubilization into the continuous phase (Figure 2). Significant increases in continuous phase lipid hydroperoxides compared to samples without added Brij 76 were observed at 70, 45, and 100 mM Brij 76 for linoleic acid, methyl linoleate, and trilinolein, respectively (Figure 2). At a Brij 76 micelle concentration of 100 mM, continuous phase linoleic acid, methyl linoleate, and trilinolein hydroperoxide were 1.3-, 1.9-, and 1.3-fold greater than samples with no added surfactant (Figure 2). After 2 h, the 100 mM Brij 76 samples contained 52% linoleic



**Figure 3.** Hydroperoxide formation in 5% corn oil-in-water emulsions with or without peroxidized linoleic acid (final hydroperoxide concentration = 0.45 mM) and Brij 76 micelles. Data markers represent average  $\pm$  SD ( $n = 3$ ).



**Figure 4.** Hexanal formation in 5% corn oil-in-water emulsions with or without peroxidized linoleic acid (final hydroperoxide concentration = 0.45 mM) and Brij 76 micelles. Data markers represent average  $\pm$  SD ( $n = 3$ ).

acid, 21% methyl linoleate, and 22% trilinolein of the total emulsion hydroperoxides in the continuous phase.

**Oxidation Studies.** To test the ability of surfactant micelles to prevent lipid oxidation, corn oil-in-water emulsions were prepared with either linoleic acid or linoleic acid hydroperoxides (0.22 mM hydroperoxides in total emulsion) and surfactant micelles (100 mM Brij 76). As expected, corn oil-in-water emulsions prepared with added linoleic acid hydroperoxides oxidized more rapidly than emulsions without hydroperoxides added. In emulsions without added Brij 76 micelles and with added linoleic acid hydroperoxides, hydroperoxide formation was 2.8-, 1.5-, and 1.1-fold higher (Figure 3) and hexanal formation was 7.3-, 2.4-, and 1.7-fold greater (Figure 4) than in emulsions with no added hydroperoxides and no added Brij 76 micelles after 2, 3, and 4 days, respectively.

Surfactant micelles were able to decrease oxidation in both systems. In the emulsions with added linoleic acid hydroperoxides, the absence of Brij 76 micelles resulted in hydroperoxide concentration being 2.6-, 1.7-, and 1.2-fold higher than in

emulsion with added Brij 76 micelles after 2, 3, and 4 days, respectively (Figure 3). Hexanal formation was 3.9-, 5.6-, and 4.8-fold higher in than the samples without added micelles after 2, 3, and 4 days, respectively (Figure 4). Similar results were obtained for the no added linoleic acid hydroperoxide samples, with emulsions without added Brij 76 micelles having lipid hydroperoxide concentrations 2.6-, 3.4-, and 1.8- times higher and hexanal concentrations 2.2-, 9.0-, and 6.3-fold greater than the added micelles samples after 2, 3, and 4 days, respectively (Figures 3 and 4). The corn oil used in these experiments contained 2.2 mM lipid hydroperoxides, which would contribute 0.11 mM hydroperoxides to the emulsions. It is likely that the Brij micelles added to the emulsion without added linoleic acid hydroperoxides would be able to solubilize the endogenous hydroperoxides in the corn oil, thus decreasing their ability to promote lipid oxidation.

## DISCUSSION

The fact that the hydroperoxide-containing samples had lower interfacial tension than their nonoxidized counterparts shows that lipid hydroperoxides have surface activity. This suggests that lipid hydroperoxides would migrate to and concentrate at the surface of an emulsion droplet. If lipid hydroperoxides concentrate at the emulsion droplet surface, they could be susceptible to interactions with aqueous phase oxidation catalysts such as iron. In emulsions produced with surfactants of various head and tail group sizes, larger surfactants are found to decrease both lipid hydroperoxide-iron interactions and lipid oxidation (14, 15). The ability of large surfactants to protect lipid hydroperoxides is likely due to the ability of these surfactants to alter the surface activity of lipid hydroperoxides or to provide a protective barrier around the emulsion droplet that would decrease lipid hydroperoxide-continuous phase prooxidant (e.g., iron) interactions.

The surface activity of the lipid hydroperoxides also seems to be related to their ability to be solubilized by Brij 76 micelles because linoleic acid (most surface active) was solubilized to a much greater extent than methyl linoleate and trilinolein. This is similar to the Brij micelle solubilization behavior of antioxidants in oil-in-water emulsions, with polar (propyl gallate) antioxidants being solubilized more than nonpolar (butylated hydroxytoluene) antioxidants (8). An additional factor that could contribute to the greater micellar solubilization of linoleic acid hydroperoxide is its smaller size, which could allow it to fit into micelles more easily than the large bulky trilinolein (16).

The ability of surfactant micelles to solubilize lipid hydroperoxides could be responsible for the ability of continuous phase surfactants to increase the oxidative stability of oil-in-water emulsions. The observed inhibition of lipid oxidation by Brij 76 micelles could be due to the ability of the surfactant micelles to remove hydroperoxides from the oil droplets, thus preventing radicals resulting from hydroperoxide breakdown from attacking unsaturated lipids in the emulsion droplets and thus propagating the chain reaction of lipid oxidation. However, it is also possible that the surfactant micelles could be inhibiting lipid oxidation by additional mechanisms such as causing alterations in the physical location of iron, the primary catalyst responsible for hydroperoxide decomposition in oil-in-water emulsions (11).

These results show that lipid hydroperoxides are surface active and can be removed from oil-in-water emulsion droplets by surfactant micelle solubilization. The ability of surfactant micelles to increase the oxidative stability of oil-in-water

emulsions could be utilized by the food industry to improve product quality. However, to utilize surfactant solubilization as an antioxidant technology, a better understanding of how surfactant micelles affect antioxidants and prooxidants such as iron is needed.

## LITERATURE CITED

- (1) Dickinson, E. *An Introduction to Food Colloids*; Oxford Science Publishers: Oxford, U.K., 1992.
- (2) McClements, D. J. *Food Emulsions: Principles, Practice, and Techniques*; CRC Press: Boca Raton, FL, 1999.
- (3) McClements, D. J.; Dungan, S. R. Light-scattering study of solubilization of emulsion droplets by nonionic surfactant solutions. *Colloids Surf. A-Physicochem. Eng. Aspects* **1995**, *104*, 127-135.
- (4) Huang, S. W.; Frankel, E. N.; Aeschbach, R.; German, J. B. Partition of selected antioxidants in corn oil-water model systems. *J. Agric. Food Chem.* **1997**, *45*, 1991-1994.
- (5) Jacobsen, C.; Schwarz, K.; Stockmann, H.; Meyer, A. S.; Adler-Nissen, J. Partitioning of selected antioxidants in mayonnaise. *J. Agric. Food Chem.* **1999**, *47*, 3601-3610.
- (6) Pekkarinen, S. S.; Stockmann, H.; Schwarz, K.; Heinonen, I. M.; Hopia, A. I. Antioxidant activity and partitioning of phenolic acids in bulk and emulsified methyl linoleate. *J. Agric. Food Chem.* **1999**, *47*, 3036-3043.
- (7) Schwarz, K.; Huang, S. W.; German, J. B.; Tiersch, B.; Hartmann, J.; Frankel, E. N. Activities of antioxidants are affected by colloidal properties of oil-in-water and water-in-oil emulsions and bulk oils. *J. Agric. Food Chem.* **2000**, *48*, 4874-4882.
- (8) Richards, M. P.; Chaiyasit, W.; McClements, D. J.; Decker, E. A. Ability of surfactant micelles to alter the partitioning of phenolic antioxidants in oil-in-water emulsions. *J. Agric. Food Chem.* **2002**, *50*, 1254-1259.
- (9) Decker, E. A.; Ivanov, V.; Zhu, B. Z.; Frei, B. Inhibition of low-density lipoprotein oxidation by carnosine and histidine. *J. Agric. Food Chem.* **2001**, *49*, 511-516.
- (10) Shanta, N. C.; Decker, E. A. Rapid, sensitive, iron-based spectrophotometric methods for determination of peroxides values of food lipids. *J. Assoc. Off. Anal. Chem.* **1994**, *77*, 421-424.
- (11) Mancuso, J. R.; McClements, D. J.; Decker, E. A. The effects of surfactant type, pH, and chelators on the oxidation of salmon oil-in-water emulsions. *J. Agric. Food Chem.* **1999**, *47*, 4112-4116.
- (12) SAS. *The SAS Program for Windows*; SAS Institute: Cary, NC, 2001.
- (13) Kreit, J.; Lefebvre, G.; Germain, P. Membrane-bound cholesterol oxidase from *Rhodococcus* sp. cells. Production and extraction. *J. Biotechnol.* **1994**, *33*, 271-282.
- (14) Silvestre, M. P. C.; Chaiyasit, W.; Brannan, R. G.; McClements, D. J.; Decker, E. A. Ability of surfactant head group size to alter lipid and antioxidant oxidation in oil-in-water emulsions. *J. Agric. Food Chem.* **2000**, *48*, 2057-2061.
- (15) Chaiyasit, W.; Silvestre, M. P. C.; McClements, D. J.; Decker, E. A. Ability of surfactant tail group size to alter lipid oxidation in oil-in-water emulsions. *J. Agric. Food Chem.* **2000**, *48*, 3077-3080.
- (16) Coupland, J. N.; Weiss, J.; Lovy, A.; McClements, D. J. Comparison of the solubilization kinetics of triacylglycerol and hydrocarbon emulsion droplets in a micellar solution. *J. Food Sci.* **1996**, *61*, 1114-1117.

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