Food Chemistry 252 (2018) 28–32

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Short communication

# The prooxidant activity of salts on the lipid oxidation of lecithin-stabilized oil-in-water emulsions

Leqi Cui[a], Jing Fan[b], Yifei Sun[c], Zhenbao Zhu[c], Jianhua Yi[c,*]

[a] School of Food Equipment and Engineering Science, Xi'an Jiaotong University, Xi'an 710049, China
[b] Xi'an Center for Disease Control and Prevention, Xi'an 710054, China
[c] School of Food and Biological Engineering, Shaanxi University of Science and Technology, Xuefu Road, Xi'an, Shaanxi 710021, China

ARTICLE INFO

*Keywords:*
Salts
Emulsion
Lecithin
Lipid oxidation
Chelating activity

ABSTRACT

Salts reduction/substitution have gained a lot interest from food industry since the US Food and Drug Administration (FDA) has issued a draft guidance for salt reduction. However how changes of salts in food formulation could influence lipid oxidation is still not fully understood. Using oil-in-water emulsions stabilized by a natural emulsifier – lecithin at pH 7.0 as a model system, this study evaluated how salts affect the physical parameters of the emulsion, the chelating activity of lecithin and thus the lipid oxidation of these emulsions. Results showed that salts increased the particle size, the negative charge of the oil droplets, and the amount of iron chelated by lecithin. Lipid oxidation lag phases were shortened by addition of salts, by 1 day and 2 days for lipid hydroperoxides and thiobarbituric acid reactive substances measurements respectively. These results provide some new insights on the mechanisms of how salts could affect the lipid oxidation of food emulsions.

## 1. Introduction

Food emulsions represent a large of group of foods including but not limiting to salad dressings, ready-to-eat soups, sauces, dips and condiments. One of the main factors influencing the shelf life of these emulsions is lipid oxidation. As lipids oxidize, a series of oxidation products are formed so that the quality and flavor of the food are adversely influenced. For this reason, lipid oxidation of emulsions has been a major concern of researchers for decades (Berton-Carabin, Ropers, & Genot, 2014; Decker et al., 2017; McClements & Decker, 2000).

One effective strategy to control lipid oxidation is using antioxidants. Known as one of the most important natural emulsifier, lecithin, which refers to a mixture of phospholipids extracted from animal or vegetable source, has also drawn a lot of attention due to its antioxidant activity. For example, Choe, Oh, and Choe (2014) reported that addition of soybean lecithin to canola oil emulsions decelerated iron-catalyzed oil oxidation and they suggested that addition of soybean lecithin as an emulsifier could also improve the oxidative stability of emulsions. The possible mechanisms behind lecithin's antioxidant activity might be: 1) lecithin regenerates primary antioxidants such as tocopherols, 2) lecithin reacts with lipid oxidation products to form antioxidative nonenzymatic browning reaction products, and 3) lecithin chelates prooxidative transition metals (Cui & Decker, 2016). However, it should be noted that when used as emulsifier, lecithin-stabilized droplets are negatively charged so that metal ions can be attracted to the water/oil interface to accelerate lipid oxidation (McClements & Decker, 2000).

Another factor that could influence lipid oxidation of emulsions is salt. The term "salt" typically refers to sodium chloride. It is of vital importance in terms of a food product's taste and microbial growth inhibition (Taormina, 2010). Almost all food emulsions contain salts. For example, ketchup, mayonnaise and barbecue/chili/hot sauces contain around 0.6–1.2, 0.4–1.9 and 0.6–3.6 wt% (ranging from 68 to 616 mmol/kg) sodium respectively (U.S. Department of Agriculture, 2013). However, it is still unclear how these salts influence lipid oxidation and thus the shelf life of food emulsions as different effects of salts were reported. For example, NaCl was found to inhibit lipid oxidation of soy protein isolate (SPI)-stabilized soybean oil-in-water emulsions (Shao & Tang, 2014) and sodium dodecyl sulfate (SDS)-stabilized salmon oil-in-water emulsions at pH 3.0 (Mei, Decker, & McClements, 1998). Nevertheless, 500 mM NaCl was reported to be pro-oxidative when added to a whey protein isolate (WPI)-stabilized canola oil/caprylic acid structured lipid-in-water emulsion at pH 3.0 (Osborn-Barnes & Akoh, 2003). Similar pro-oxidant activity has also been reported in Tween-20-stabilized corn oil-in-water emulsions at pH 7.0 (Cui, Cho, McClements, Decker, & Park, 2016). In addition, NaCl was also found to have minimal impact on emulsion oxidation (Mei,

* Corresponding author.
*E-mail address:* yijianhua@sust.edu.cn (J. Yi).

https://doi.org/10.1016/j.foodchem.2018.01.094
Received 8 September 2017; Received in revised form 10 January 2018; Accepted 12 January 2018
Available online 12 January 2018

McClements, Wu, & Decker, 1998). Due to direct association with elevated blood pressure, salt consumption has been an issue in Europe for more than ten years (European Food Safety, 2005). In 2016, the US FDA also issued a draft guidance for voluntary salt reduction. The salt reduction trend is urging the industry to find a solution to properly reformulate their products that hold a desirable shelf life and meet consumer's taste preference as well. Nevertheless, since the mechanism of salts' impact on the lipid oxidation of emulsion is still not fully understood, even less is known on how the reduction and/or substitution of salts can possibly change the oxidative stability and thus the shelf life of a food emulsion.

So far, studies (such as those mentioned above) investigating the effects of salts on emulsion oxidation have all been using "interaction between salts and metal ions" to explain their oxidation results. This "interaction between salts and metal ions" includes: 1) the ability of chloride ions to increase the catalytic activity of metals (Osinchak, Hultin, Zajicek, Kelleher, & Huang, 1992), and 2) the ability of sodium to decrease iron binding at the droplet surface or the formation of iron-chloride complexes which decreased iron binding to the interface (Mei et al., 1998). This hypothesis has also been used in other systems such as cookies (Mesías, Holgado, Márquez-Ruiz, & Morales, 2015) and pork patties (Overholt et al., 2016). However, this hypothesis is not sufficient to explain various effects of salts mentioned above in emulsion systems at different pH. Thus our question is that except the direct interaction between salts and metal ions, are there any other mechanisms through which salts affect lipid oxidation of emulsions? For example, in emulsions stabilized by natural emulsifier lecithin, can salts affect lecithin's chelating ability and therefore the final oxidative stability of emulsions? To answer these questions and get a better understanding of the impact of salts on lipid oxidation of emulsions, this study used lecithin-stabilized oil-in-water emulsion at pH 7.0 as a model system to investigate the influence of KCl (as one of the potential substitutions for NaCl) and NaCl individually on the physical properties of emulsion, the chelating activity of lecithin and thus the oxidative stability of these emulsions.

## 2. Materials and methods

### 2.1. Materials

Purified walnut oil was prepared by activated silicic acid and charcoal column chromatography according to Cui, McClements, and Decker (2015). Commercial lecithin Epikuron™ (23.6% phosphatidylcholine, 18.4% phosphatidylethanolamine, 5.0% phosphatidic acid and 14.6% phosphatidylinositol) was purchased from Cargill Inc. (Hamburg, Germany). Sodium chloride, potassium chloride, ammonium iron(II) sulfate hexahydrate, cumene hydroperoxide and 1,1,3,3-tetraethoxypropane were purchased from Aladdin Co. Ltd. (Shanghai, China). All reagents were of analytical grade. Distilled and deionized water was used in all experiments.

### 2.2. Emulsions preparation

Oil-in-water emulsions were prepared using 5 wt% oil in 95 wt% aqueous phase. The aqueous phase was prepared by dispersing 1.5 wt% (of the aqueous phase) lecithin in 5 mM phosphate buffer at pH 7.0. Oil and aqueous phase containing lecithin were added to a beaker, and a coarse emulsion was made by blending with a Fluko superfine homogenizer (F6/10, Fluko Equipment Shanghai Co. Ltd., Shanghai, China). The coarse emulsion was then passed through a high pressure homogenizer (HP-4L, Xigaofluid Co. Ltd., Lanzhou, China) three times at 9000 psi. 0, 171 or 274 mM sodium chloride (0, 1.0 or 1.6 wt%) or potassium chloride was added to the emulsions. Samples were then stored at 45 °C in dark without stirring. For oxidation measurements, sampling was conducted every 24 h from the same sample bottle.

### 2.3. Determination of droplet size and ζ–potential of emulsions

The particle size distribution of all samples was measured by static light scattering using a commercial instrument (Mastersizer 2000, Malvern, Worcestershire, UK), with a small volume sample dispersion unit (Hydro 2000 MU(A)). All samples were diluted in 5 mM phosphate buffer adjusted to pH 7.0. The refractive indexes used for particle and dispersant phase are 1.53 and 1.33 respectively. Each sample was measured in duplicate and the results were reported as volume-weighted mean diameter $d_{43}$. The ζ-potential of all samples was measured by a zeta-potential analyzer (Zetasizer NANO-ZS 90, Malvern, UK). Prior to measuring, samples were diluted with 5 mM phosphate buffer at pH 7.0 to a droplet concentration of approximately 0.025 wt%. Each measurement was repeated twice.

### 2.4. Determination of iron chelation by lecithin

Iron chelation by lecithin was determined according to Demant (Jayasinghe, Gotoh, & Wada, 2013) with some modifications. A two-phase system consisting of 3 mL aqueous phase containing 100 ppm iron ($Fe(NH_4)_2(SO4)_2{\cdot}6H_2O$) with or without 171, 274, 1710 or 2740 mM NaCl or KCl, and 1 mL hexane phase containing 0, 0.5, 1.0, 1.5 or 2.0 wt% (of total solution) lecithin was prepared. The system was vigorously vortexed for 1 min with a 5 s break for a total of 6 min and then centrifuged (SC-06 Zonkia Inc., Anhui, China) at 3802*g* for 10 min.

The remaining iron in the bottom aqueous phase was then measured according to Castellani, Guérin-Dubiard, David-Briand, and Anton (2004). Briefly, 350 μL of bottom solution was pipetted into an empty tube, followed by addition of 125 μL of hydroxylamine hydrochloride and 250 μL of sodium acetate buffer (pH 4.5). After 5 min incubation, 250 μL of *o*-phenanthroline was added followed by another 20 min reaction time. The absorbance was then measured at 510 nm. Iron concentration was calculated from a standard curve prepared using ammonium iron (II) sulfate hexahydrate. Iron taken up by lecithin was calculated as: Iron chelated (%) $= \left(1-\frac{Cre}{Ci}\right) \times 100\%$, where $Ci$ and $Cre$ are the initial and remaining iron concentration (ppm) in the bottom aqueous layer. For comparing iron chelation by various lecithin concentrations, the control is the iron taken up into pure hexane (no lecithin added). For comparing iron chelation by 1.5 wt% lecithin influenced by various salts concentrations, the control is the iron taken up into hexane containing lecithin (no salts added).

### 2.5. Measurement of lipid oxidation parameters

Lipid hydroperoxides were measured as primary oxidation products according to Shantha and Decker (1994). In brief, 0.3 mL emulsion was added to a mixture of 1.5 mL isooctane/2-propanol (3:1 v/v), vortexed, and centrifuged at 1000*g* for 2 min. 200 uL of the upper phase was then added to 2.8 mL of a methanol/ butanol (2:1, v/v). Then 15 μL of 3.94 M ammonium thiocyanate and 15 μL of 0.072 M $Fe^{2+}$ (ferrous sulfate) were added and the solution was vortexed. After 20 min incubation the absorbance was measured at 510 nm. The concentration of lipid hydroperoxides was calculated from a standard curve prepared using cumene hydroperoxide.

Thiobarbituric acid reactive substances (TBARS) were measured as secondary oxidation products using a method adapted from Alamed, McClements, and Decker (2006). Two milliliter of emulsion was mixed with 4 mL of a TBA solution containing 15% w/v trichloroacetic acid and 0.375% w/v thiobarbituric acid in 0.25 M HCl mixed with 2% BHT in ethanol in screw-capped tubes, and placed in a boiling water bath for 20 min. The tubes were cooled at room temperature and then centrifuged at 1600*g* for 15 min. The absorbance was measured at 532 nm. Concentrations of TBARS were determined from a standard curve prepared using 1,1,3,3-tetraethoxypropane.