

## Oxidative stability of olive oil–lemon juice salad dressings stabilized with polysaccharides

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Received 26 September 2005; received in revised form 16 January 2006; accepted 13 March 2006

### Abstract

Lipid oxidation is a major cause of quality deterioration in food emulsions. Polysaccharides used to improve emulsion stability and texture may also affect lipid oxidation. In the present study, the oxidative stability of olive oil–lemon juice salad dressings, stabilized with gum arabic or propylene glycol alginate in admixture with xanthan, was investigated. Oil-in-water emulsions (50:50, v/v) were prepared with lemon juice and extra virgin olive oil and then homogenized at various homogenization rates to form different particle sizes. Keepability was followed by storing at room temperature for 6–8 months and measuring the formation of primary and secondary oxidation products. The shelf life was compared to that of the bulk olive oil. It was shown that the polysaccharides had the ability to inhibit lipid oxidation, probably due to their amphiphilic character (gum arabic and propylene glycol alginate) as well as their ability to induce viscosity increase. Olive oil–lemon juice emulsions were also assessed for consumer acceptance. The panellists were asked to smell the samples and rate them according to rancidity using a four-point (1 = no perception, 4 = extreme) intensity scale. The results were in accordance to those of chemical analysis. Lipid oxidation was not affected by the oil droplet size, as demonstrated by peroxide value measurements and sensory evaluation.

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*Keywords:* Salad dressing; Olive oil; Lemon juice; Gum arabic; Propylene glycol alginate; Xanthan; Oxidation; Keepability; Droplet size

### 1. Introduction

Emulsions are thermodynamically unstable systems and they tend to separate into two layers over time through a number of mechanisms (Dickinson, 1992; McClements, 1999). In order to make emulsions kinetically stable for a reasonable period of time, emulsifiers and/or stabilizers (e.g. proteins, phospholipids, polysaccharides) must be added. Polysaccharides are usually added to o/w emulsions to enhance viscosity of the aqueous phase, thus preserving desirable textural characteristics and stabilizing oil droplets against creaming (Dickinson, 1992; Paraskevopoulou et al., 2003; Paraskevopoulou, Boskou, & Kiosseoglou, 2005; Paraskevopoulou, Kiosseoglou, Alevisopoulos, & Kasapis, 1997).

Amphiphilic polysaccharides, such as gum arabic and propylene glycol alginate, act both as emulsifiers and stabilizers. Because of their surface-active properties they can adsorb onto the surface of freshly formed droplets during homogenization and prevent them from aggregation by steric and/or electrostatic forces. Studies have shown that polysaccharides are also capable of retarding lipid oxidation in o/w emulsions (Matsumura et al., 2003; Shimada, Fujikawa, Yahara, & Nakamura, 1992; Shimada et al., 1994; Shimada, Okada, Matsuo, & Yoshioka, 1996). The polysaccharide-induced viscosity increase of the aqueous phase inhibits oxygen diffusion and slows down the movement of oil droplets, thus reducing their collision probability. Additionally, the metal ion-chelating ability of polysaccharides has been proposed to account for their ability to inhibit lipid oxidation, while tragacanth gum has been found to act as a radical chain-breaker because of its ability to donate hydrogen (Shimada et al., 1992).

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Lipid oxidation is considered to be the predominant cause of quality deterioration of oils, fats and various fat-containing foods during storage. It involves the interaction between unsaturated lipids and oxygen-active species. It can cause alterations in the quality characteristics of foods, such as appearance, taste, texture, shelf life and nutritional profile and lead to the development of undesirable off-flavour (Min & Boff, 2002). Numerous studies have been carried out dealing with oxidation mechanisms in bulk oils although, in most processed foods, lipids are found as o/w or w/o emulsions. The oxidation of emulsified lipid is mechanistically different from that of bulk oils. In such products, the organization of the lipid molecules within the system and their interactions with other food components influences their susceptibility to lipid oxidation. A great number of studies have highlighted the impact of various factors in determining the oxidative stability of an oil-in-water emulsion, including chemical structure of lipids, oxygen concentration, antioxidants, interfacial characteristics, droplet characteristics (concentration, size), interactions with aqueous phase components (salts, sugars, polysaccharides, proteins.) and the presence of prooxidants (e.g. transition metal impurities) (McClements & Decker, 2000).

Salad dressings are very popular oil-in-water emulsion products, which may vary in fat content (20–65%) and viscosity (Dickinson & Stainsby, 1982). “Greek salad dressings” are, by definition, mixtures of virgin olive oil and lemon juice instantly prepared before use (Paraskevopoulou et al., 2005). Due to their composition, they are a good source of biophenols (Boskou & Visioli, 2003) as well as lipid-soluble and water-soluble vitamins (tocopherols,  $\beta$ -carotene, ascorbic acid). Besides, virgin olive oil, thanks to its balanced fatty acid composition, has highly appreciated nutritional characteristics, known for a long time to the people of the Mediterranean region, who use it daily for a variety of culinary purposes (Garcia Mesa, Jimenez-Marquez, Beltran-Maza, & Friaz-Ruiz, 1998; Romero, Guesta, & Sanchez-Muniz, 1998). Its consumption has also increased in non-Mediterranean areas because of the growing interest in the Mediterranean diet and the belief that it prevents certain diseases (Boskou & Visioli, 2003; Trichopoulos, 1995).

The development of a “Greek salad dressing” containing olive oil and lemon juice that would exhibit reasonable physicochemical stability over prolonged storage has been the subject of a previous work (Paraskevopoulou et al., 2005). Different combinations of xanthan gum with gum arabic or propylene glycol alginate exhibited a positive effect on the creaming and rheological behaviour, rate of oil droplet coalescence and sensory properties of the salad dressings. Gum arabic (*gum Acacia*), a hydrocolloid produced by the natural exudation of acacia trees, is a complex mixture of six main carbohydrate components and a small but functionally important amount of protein (~2% w/w), which is found as an integral part of the structure (Wareing, 1999). Propylene glycol alginate, an esterified form of

alginate, is appreciated as a particularly effective thickener in several acidic food applications, such as salad dressings and lactic drinks (Onsøyen, 1999). Xanthan gum is an extracellular polysaccharide made by the bacterium *Xanthomonas campestris* (Urlacher & Noble, 1999).

The favourable effect of the above-mentioned polysaccharides, as well as the role of the oil droplet size on the keepability of “Greek salad dressings”, was the objective of this work. By modifying the emulsification energy, emulsions with different droplet sizes were prepared. Keepability was measured by periodically measuring peroxide value, acidity and specific extinction coefficient  $K_{232}$ . Furthermore, the sensory perception of lipid oxidation products of the emulsions by the consumers was assessed.

## 2. Materials and methods

### 2.1. Materials

Gum arabic (GA) and xanthan gum (X) were purchased from Sigma Chemical Co. (USA). Propylene glycol alginate (PGA) (degree of esterification 40–60%) was kindly supplied by FMC Biopolymer (Brussels, Belgium). Extra virgin olive oil was bought from the local market. Lemon juice was a commercial sample purchased in a local supermarket. Egg yolk (EY) was obtained by first breaking fresh hen’s eggs and, following complete removal of the adhering white by rolling the intact yolks on tissue paper, the vitelline membrane was punctured and the liquid yolks of a number of eggs were collected. Benzoic acid was obtained from Riedel de Haën (Germany).

### 2.2. Preparation of salad dressings

Oil-in-water emulsions (50% v/v) were prepared as follows: a lemon juice polysaccharide solution was first prepared by slowly dispersing 1% w/v gum arabic (or 1% w/v PGA) and 0.5% w/v xanthan gum, with stirring for at least 6 h, to ensure complete dissolution. The emulsions were prepared by adding dropwise, while mixing with a propeller-type mechanical stirrer, 50 ml of virgin olive oil to 50 ml of the polysaccharide solution. The droplet size of the resulting crude emulsion was then reduced further using an Ultra-Turrax T25 homogenizer (IKA Instruments, Germany), equipped with an S25KG-25F dispersing tool. By modification of the energy of emulsification three gum arabic/xanthan-stabilized (GA/X 9500 rpm, GA/X 13,500 rpm, GA/X 24,000 rpm) and three propylene glycol alginate/xanthan-stabilized (PGA/X 8000, PGA/X 9500, PGA/X 20,500 rpm) emulsions with different oil droplet sizes, were produced. Thus, it was possible to test the effect of droplet size and the effect of stabilizing agent on the oxidative stability of the produced Greek-type salad dressings. Benzoic acid was added at a concentration of 1‰ w/v in the continuous phase. The pH values of the final samples ranged between 3.4 and 3.6.

For comparison, another olive oil-in-lemon juice salad dressing (50:50 v/v) containing 1% w/v egg yolk solids, in the place of GA or PGA, was prepared by the same emulsification procedure, following homogenization at 13,500 rpm for 2 min.

Olive oil samples and salad dressings (100 ml) were stored in screw-capped glass containers (100 ml) prior to analysis.

### 2.3. Droplet size determination

The droplet size distribution of the salad dressings over the storage period was measured using a laser light scattering instrument (Malvern Instruments, UK). Measurements are either reported as the full particle size distribution or as the surface-volume mean diameter  $d_{3/2} = \frac{\sum n_i d_i^3}{\sum n_i d_i^2}$ , where  $n_i$  is the number of droplets of radius  $d_i$ . To prevent multiple scattering effects, the emulsions were diluted with deionized water prior to analysis so that the droplet concentration was less than about 0.02% v/v. The dilute emulsions were placed directly into the measurement cell of the instrument and stirred slowly during the measurement. Each sample was analyzed 4 times and the data are presented as averages. Droplet sizes were checked periodically to monitor emulsion stability.

### 2.4. Viscosity measurements

The viscosities of the salad dressings were determined at 25 °C at various shear rates (from 0.1 to 20.4 s<sup>-1</sup>) with the aid of a Brookfield DV-II, LV viscometer (USA), equipped with concentric cylinder geometry. The flow curves giving viscosity  $\eta$  (MPa s) as a function of shear rate  $\dot{\gamma}$  (s<sup>-1</sup>) were characteristic of shear-thinning behaviour.

### 2.5. Oxidative stability evaluation

#### 2.5.1. Experimental design

Twenty containers of each emulsion were stored in conditions similar to those in consumers' sales points: inside a chamber at 23 °C and placed in shelf units, 2.5 m high with illumination for ~12 h/day. Containers were sampled every three or four weeks for complete 3.5–6 months storage.

Furthermore, 100 ml of virgin olive oil were stored under the same conditions and sampled in a similar way.

#### 2.5.2. Analytical determinations

Free acidity, peroxide value (PV) and specific extinction coefficient  $K_{232}$  were determined according to the analytical methods described in Regulation EC/2568/91 of the Commission of the European Union (EU, 1991). Oil was separated from the salad dressings by repeated freeze–thaw cycles, followed by centrifugation (Jacobsen et al., 1999). Two containers of each sample were independently analyzed in each sampling and each parameter was measured twice. The results are expressed as mean values.

Free acidity, given as % oleic acid, was determined by titration of a solution of oil in ethanol/ether (1:1) with ethanolic potash.

Peroxide value, expressed as milliequivalents of active oxygen per kilogramme of oil (meq/kg), was determined as follows: a mixture of oil and chloroform/acetic acid was left to react with a solution of potassium iodide in darkness; the free iodine was then titrated with a sodium thiosulfate solution.

$K_{232}$  extinction coefficient was calculated from the adsorption of a solution of the oil in iso-octane at 232 nm, using a UV spectrophotometer (Hitachi, Japan) and a path length of 1 cm.

### 2.6. Organoleptic assessment

Organoleptic evaluation was performed for salad dressings and extra virgin olive oil, following their storage time, simultaneously with oxidative stability measurements. The analytical panel, consisting of 20 untrained members, was asked to make its evaluation on the basis of rancidity. The panellists were students and members of the staff of the Laboratory and were asked to score the samples by smelling them. Rancidity grading was based upon a four-point intensity scale: 1 = no perception; 2 = weak; 3 = medium; 4 = extreme (Jellinek, 1984). The dressings and olive oil samples were served in glass beakers (~20 ml in each beaker) at room temperature.

### 2.7. Statistical analysis

All experiments were performed on quadruplicate samples. Statistical analyses were conducted with the one-way ANOVA software package. Significant differences among means ( $p < 0.05$ ) were determined by LSD test.

## 3. Results and discussion

### 3.1. Properties of the salad dressings

Prepared salad dressings were monomodal with the droplet size distributions overlapping one another, as shown in Fig. 1. The emulsions prepared with a mixture of GA/X were physicochemically stable over the storage period as no coalescence and no droplet size or viscosity variations were observed over the time of experiments (Table 1, Fig. 2). On the other hand, the droplet size of the rest of the emulsions (prepared with a mixture of PGA/X or EY/X) increased significantly ( $p < 0.05$ ) during storage. From the droplet size data obtained by the laser diffraction, it was observed that GA/X-stabilized emulsions had the lowest droplet sizes, likely due to the fact that gum arabic is more surface-active than PGA. The larger droplet size of EY/X-stabilized emulsions could be attributed to their relatively low egg yolk content (1% w/v egg yolk solids).

Finally, their viscosity remained stable throughout the storage period (Fig. 2).

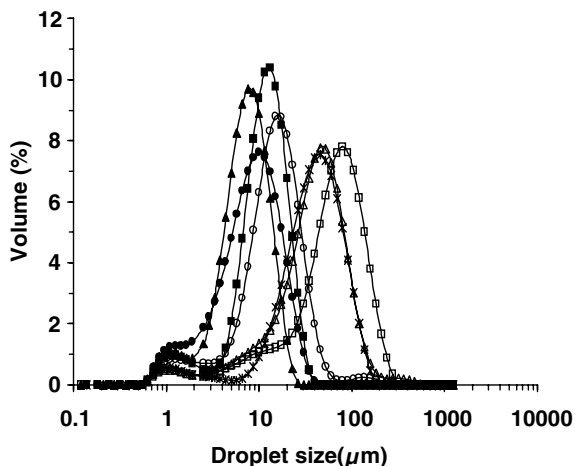


Fig. 1. Droplet size distribution of olive oil–lemon juice emulsions prepared at various energies of emulsification and stabilized with GA/X (■, ▲, ●), PGA/X (□, △, ○) and EY/X (✱) mixtures 24 h after preparation. Emulsions prepared with: *low energy input* (■, □); *medium energy input* (▲, △); *high energy input* (●, ○).

### 3.2. Acidity of the lipid phase

The salad dressings showed an increase of acidity with the storage time that became significant ( $p < 0.05$ ) at the end of the storage period (Fig. 3). This increase was attributed to the migration of acids/acidic compounds from lemon juice to olive oil droplets during their preparation.

### 3.3. Oxidative stability of salad dressings

#### 3.3.1. Quality characteristics of olive oil sample

The quality characteristics that influence the storage stability of olive oil were determined before the preparation of salad dressings and are shown in Table 2. Acidity, peroxide value and coefficients of specific extinction ( $K_{232}$ ,  $K_{270}$ ) were determined. All values were lower than the limits set by the EU Regulation 2568/91 for extra virgin olive oil.

Table 1  
Effect of energy of emulsification on the mean droplet diameter ( $d_{32}$ ) increase with time of olive oil–lemon juice salad dressings stabilized with mixtures of PGA/X, GA/X or EY/X

| $t$ (days) | Propylene glycol alginate/xanthan |                   |                  | Gum arabic/xanthan |                  |                  | Egg yolk/xanthan    |
|------------|-----------------------------------|-------------------|------------------|--------------------|------------------|------------------|---------------------|
|            | 8000 rpm                          | 9500 rpm          | 20,500 rpm       | 9500 rpm           | 13,500 rpm       | 24,000 rpm       | 13,500 rpm          |
| 0          | 15.9 <sup>a</sup>                 | 12.4 <sup>a</sup> | 7.2 <sup>a</sup> | 6.3 <sup>a</sup>   | 4.5 <sup>a</sup> | 4.5 <sup>a</sup> | 15.1 <sup>a</sup>   |
| 30         | 16.3 <sup>a</sup>                 | 12.6 <sup>a</sup> | 7.2 <sup>a</sup> | 6.5 <sup>a</sup>   | 4.6 <sup>a</sup> | 4.6 <sup>a</sup> | 17.0 <sup>a</sup>   |
| 45         | 16.5 <sup>a</sup>                 | 13.5 <sup>a</sup> | 7.2 <sup>a</sup> | –                  | –                | –                | –                   |
| 60         | 16.6 <sup>a</sup>                 | 14.2 <sup>b</sup> | 7.5 <sup>a</sup> | 6.0 <sup>a</sup>   | 4.1 <sup>a</sup> | 4.9 <sup>a</sup> | 21.2 <sup>a,b</sup> |
| 90         | 21.9 <sup>b</sup>                 | 16.8 <sup>b</sup> | 8.0 <sup>a</sup> | 6.1 <sup>a</sup>   | 4.5 <sup>a</sup> | 5.0 <sup>a</sup> | 22.0 <sup>b</sup>   |
| 160        | –                                 | –                 | –                | 6.1 <sup>a</sup>   | 4.3 <sup>a</sup> | 5.1 <sup>a</sup> | 25.3 <sup>b</sup>   |

For each column, different superscripts indicate significant differences ( $p < 0.05$ ) among mean droplet diameters.

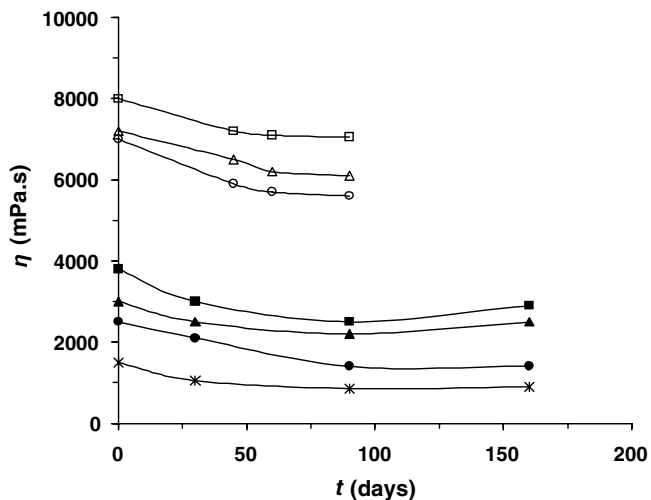


Fig. 2. Shear viscosity (at  $2 \text{ s}^{-1}$ ,  $25^\circ \text{C}$ ) of olive oil–lemon juice emulsions prepared with GA (■, ▲, ●), PGA (□, △, ○) or EY (✱) 1% (w/v) (on a dry basis) and xanthan 0.45% (w/v) as a function of storage time. Emulsions prepared with: *low energy input* (■, □); *medium energy input* (▲, △); *high energy input* (●, ○).

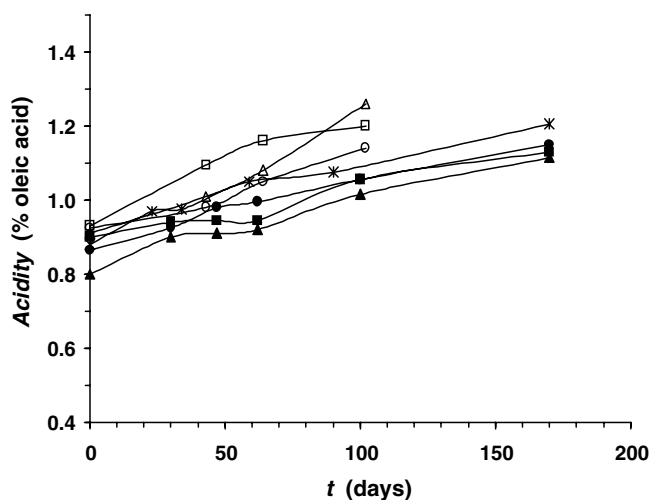


Fig. 3. Changes in acidity over storage time at  $23^\circ \text{C}$  in virgin olive oil sample and olive oil–lemon juice emulsions prepared with GA (■, ▲, ●), PGA (□, △, ○) or EY (✱) 1% (w/v) (on a dry basis) and xanthan 0.45% (w/v) as a function of storage time. Emulsions prepared with: *low energy input* (■, □); *medium energy input* (▲, △); *high energy input* (●, ○).

Table 2  
Quality characteristics of the virgin olive oil used in the experiments

| Quality characteristics                                  | Value | Quality limits <sup>a</sup> |
|--|-------|-----------------------------|
| Acidity (percentage oleic acid)                          | 0.49  | ≤1%                         |
| Peroxide value (meq O <sub>2</sub> /kg of oil)           | 9.65  | ≤20                         |
| K <sub>232</sub> [where K = absorbance/C (g/100 ml oil)] | 1.72  | ≤2.50                       |
| K <sub>270</sub> [where K = absorbance/C (g/100 ml oil)] | 0.20  | ≤0.20                       |

<sup>a</sup> Set by the EU Regulation 2568/91.

### 3.3.2. Primary oxidation

Lipid oxidation in oil-in-water emulsions is taking place at the surface of the oil droplets. Many studies suggest that the interaction between lipid hydroperoxides, the first products formed by oxidation, located at the droplet surface and transition metals, from the aqueous phase, is the most common cause of oxidative instability (Mei, McClements, & Decker, 1998a; Mei, McClements, & Decker, 1998b). The most likely mechanism is the decomposition of lipid hydroperoxides (ROOH) by the pro-oxidants into highly reactive peroxy (ROO<sup>•</sup>) and alkoxy (RO<sup>•</sup>) radicals, which react with unsaturated lipids within the droplets or at the oil–water interface, leading to the formation of lipid radicals. The lipid oxidation chain reaction propagates as these lipid radicals react with other lipids in their immediate vicinity.

Fig. 4 shows the storage stability of virgin olive oil and GA/X-, PGA/X- and EY/X-stabilized olive oil/lemon juice salad dressings. Oxidation was more rapid in olive oil sample than in salad dressings. The rate of hydroperoxide formation increased sharply in olive oil after an induction period of thirty (30) days. The polysaccharide- and egg yolk-stabilized emulsions exhibited a significantly lower peroxide value (PV) ( $p < 0.05$ ) than that of the oil throughout the storage period, which indicated the effectiveness of the above emulsifiers/stabilizers in the keepability of the

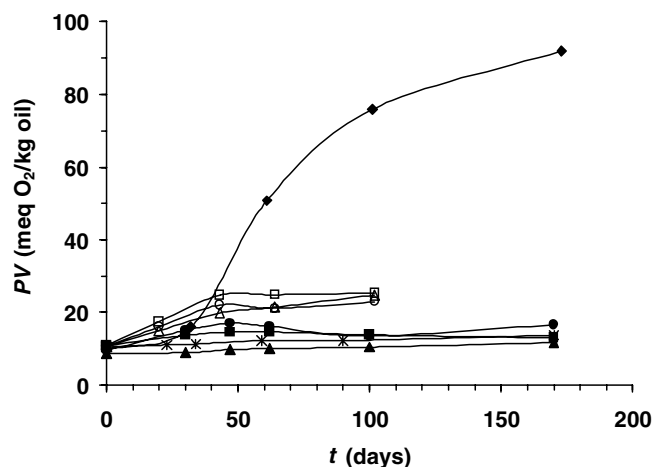


Fig. 4. Changes in peroxide values (PVs) over storage time at 23 °C in virgin olive oil sample (◆) and olive oil–lemon juice emulsions prepared with GA (■, ▲, ●), PGA (□, △, ○) or EY (✕) 1% (w/v) (on a dry basis) and xanthan 0.45% (w/v) as a function of storage time. Emulsions prepared with: low energy input (■, □); medium energy input (▲, △); high energy input (●, ○).

salad dressings. At the time (100 days) olive oil reached a peroxide value of 75 meq O<sub>2</sub>/Kg oil, GA/X- and EY/X-stabilized salad dressings had seven times lower peroxide values while PGA/X-stabilized emulsions four times.

### 3.3.3. Viscosity

The viscosity increase of the continuous phase (lemon juice) and hence the slowing down of the movement of the reactants present in it may account for their ability to retard lipid oxidation. The effect of polysaccharide addition on the viscosity of lemon juice/olive oil emulsions has already been examined (Paraskevopoulou et al., 2005). Measurement of viscosity of the salad dressings at various stages of storage indicates that increased viscosity contributes to an increased stability to lipid oxidation (Fig. 2).

### 3.3.4. Droplet surface

Additionally, the presence of GA and PGA at the oil droplet surface, due to their surface-active parts in their molecules, may also contribute to the oxidative stability of the salad dressings. Lipid hydroperoxides are surface-active and therefore migrate to and concentrate at the surface of the emulsion droplets, being susceptible to interactions with aqueous phase oxidation catalysts, such as iron (Nuchi, Hernandez, McClements, & Decker, 2002). Proteins, as well as polysaccharides, are often used in food emulsions to stabilize droplets against flocculation or coalescence and to improve their textural properties (Dickinson, 1992; McClements, 1999). According to McClements and Decker (2000), adsorbed emulsifiers are likely to be particularly effective at retarding lipid oxidation because of the ability of the interfacial membrane to act as a physical barrier that separates lipid substrates from pro-oxidants in the aqueous phase. Additionally, certain types of emulsifier molecules, containing either sugar or amino acid moieties (e.g. gum arabic, soluble soybean polysaccharide and proteins), may act as a chemical barrier to lipid oxidation by scavenging free radicals (Matsumura, Satake, Egami, & Mori, 2000; McClements & Decker, 2000). The inhibiting effect of peptide-bound polysaccharide GA against lipid oxidation in methyl oleate or methyl linoleate emulsions emulsified by  $\beta$ -casein or other surfactants (sugar ester S-1670, Tween 20) has been reported by Matsumura et al. (2000, 2003). Another mechanism that may account for the capability of the three polysaccharides to inhibit lipid oxidation is metal ion chelation. Xanthan has been shown to suppress oil peroxidation by inactivation of Fe<sup>2+</sup> ions, present in the system (Karadjova, Zachariadis, Boskou, & Stratis, 1998), due to its ability to chelate metal ions at negatively charged pyruvate sites (Shimada et al., 1992, 1994).

### 3.3.5. Emulsifier/stabilizer type

As can be seen (Fig. 4), hydroperoxide formation in the salad dressings was significantly affected by the emulsifier/stabilizer type used for their preparation. During the first days of storage all the emulsions appeared to be equally

effective at stabilizing olive oil droplets. After 20 days of storage, GA/X and EY/X were found to be more effective than PGA/X since both are better emulsifiers than PGA. Their ability to protect lipid hydroperoxides is likely due to the ability of these molecules to adsorb at the oil droplet surface, forming a film of high surface shear viscosity.

### 3.3.6. Oil droplet size

Oil droplet size influence on hydroperoxide formation rates is also seen in Fig. 4. For a fixed oil droplet concentration (50% in our experiments), an increased rate of oxidation was expected as the droplet size decreased, because of the increased surface area that was exposed to the aqueous phase. However, no dependence of the lipid oxidation rate on droplet size of olive oil/lemon juice emulsions was observed at any point of time during their storage. This was explained by the limited amounts of lipid hydroperoxides ( $\sim 10$  meq  $O_2$ /kg oil on preparation day) that were available in the emulsion systems and might have been present at the droplet surface. Other researchers have reported similar droplet effects on lipid oxidation (Osborn & Akoh, 2004; Roozen, Frankel, & Kinsella, 1994). If there was a high concentration of hydroperoxides in the systems, then increasing the surface area by decreasing the oil droplet size might increase their concentration at the interface, leading to the expected increase of lipid oxidation with decreasing droplet size.

### 3.3.7. Oxidation products

During lipid oxidation, a number of decomposition reactions occur simultaneously, that in turn result in the generation of a wide variety of different molecules, including aldehydes, ketones, alcohols and hydrocarbons. These oxidation products are responsible for the characteristic physicochemical and sensory properties of oxidized oils, since they are likely to be more surface-active than the initial lipid and some of them are water-soluble. The measurement of the  $K_{232}$  specific extinction coefficient was used to determine the level of conjugated dienes present in the emulsified oil. It is known that the oxidation products of oils and fats, which may result from their decomposition, display characteristic spectra in the ultraviolet region and at about 232 nm. Therefore, a determination of the absorbance at 232 nm is an indication of the state of oxidation of a fat.

The  $K_{232}$  coefficient of specific extinction showed a significantly ( $p < 0.05$ ) higher initial value for olive oil–lemon juice salad dressings stabilized with both GA ( $\sim 3.2$ ) and PGA ( $\sim 4.0$ ), in comparison to olive oil ( $\sim 1.75$ ). This is probably due to carbonyl compounds present in lemon juice being different from those due to oil rancidity (Fig. 5).

The  $K_{232}$  coefficient in virgin olive oil increased with storage time and the limit of 2.50 was exceeded after 100 days of storage. In salad dressings, on the other hand, the  $K_{232}$  coefficient remained practically constant during the storage period, indicating a delay in the formation of oxidation products. This was expected because of the delayed development of hydroperoxides in the emulsions (Fig. 4). Emul-

sions stabilized with a mixture of PGA/X exhibited  $K_{232}$  values ( $\sim 4.0$ ) significantly higher than the corresponding ones for GA/X-stabilized emulsions ( $\sim 3.5$ ). Likewise, EY/X-stabilized emulsions registered a constant value for  $K_{232}$  ( $\sim 3.4$ ) throughout the storage period. This was probably due to the fact that egg yolk proteins are below their isoelectric point in the pH ( $\sim 3.5$ ) of the emulsion systems, thereby producing positively charged emulsion droplets. As Osborn and Akoh (2003) have pointed out, at this pH value, the positively charged metal ions can not bind to the emulsion droplets, which explains the decreased oxidation rates in the EY/X-stabilized salad dressings.

### 3.4. Sensory studies

Sensory evaluations have been carried out to characterize the salad dressings and to find a correlation with chemical analysis. The sensory evaluation of the salad dressings appears to be necessary since the increased shelf life of these products by the addition of polysaccharides, such as GA and PGA, is futile if sensory properties are unacceptable to consumers. For this reason, a questionnaire was conducted to detect the acceptability of lemon juice–olive oil emulsions (4-point intensity scale: 1 = no perception; 2 = weak; 3 = medium; 4 = extreme). The results are presented in Table 3. Storage of virgin olive oil at 23 °C revealed the appearance of rancid odour ( $2.33 \pm 1.07$ ) in almost 9 weeks. This was also indicated by the peroxide value and  $K_{232}$  measurements. Lower values for rancidity were measured in all emulsions compared to olive oil even after 33 weeks.

On the other hand, in the emulsions stabilized with PGA/X, the intensity of rancidity was not changed significantly during storage ( $1.33 \pm 0.65$ – $1.58 \pm 0.90$  after 15

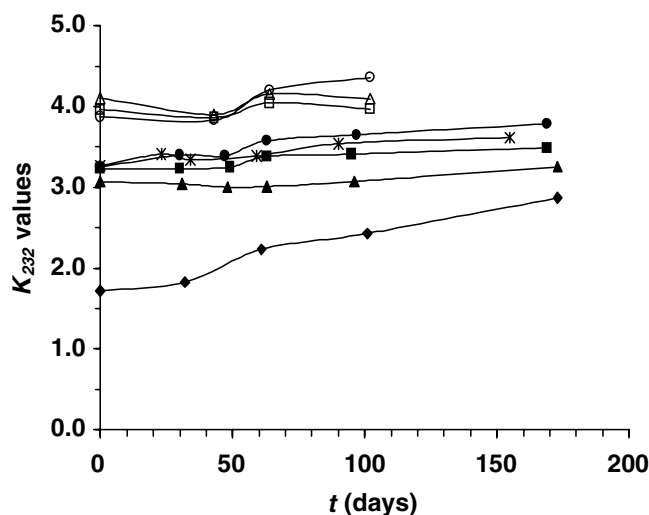


Fig. 5. Changes in specific extinction coefficient values ( $K_{232}$ ) over storage time at 23 °C in virgin olive oil sample (◆) and olive oil–lemon juice emulsions prepared with GA (■, ▲, ●), PGA (□, △, ○) or EY (X) 1% (w/v) (on a dry basis) and xanthan 0.45% (w/v) as a function of storage time. Emulsions prepared with: low energy input (■, □); medium energy input (▲, △); high energy input (●, ○).

Table 3  
Sensory scores for rancidity for salad dressings and virgin olive oil during storage at 23 °C (sensory scale 1–4; mean  $\pm$  SD)

| Virgin olive oil                  |                              | Egg yolk/xanthan             |                                |                                  |                                  |                                |
|-----------------------------------|------------------------------|------------------------------|--------------------------------|----------------------------------|----------------------------------|--------------------------------|
| Storage period (weeks)            |                              | Storage period (weeks)       |                                |                                  |                                  |                                |
| 0                                 | 9                            | 0                            | 4                              | 14                               | 21                               | 33                             |
| 1.42 $\pm$ 0.51 <sup>a</sup>      | 2.33 $\pm$ 1.07 <sup>b</sup> | 1.30 $\pm$ 0.47 <sup>a</sup> | 1.47 $\pm$ 0.61 <sup>a,b</sup> | 1.88 $\pm$ 0.89 <sup>a,b,c</sup> | 2.00 $\pm$ 0.93 <sup>b,c</sup>   | 2.09 $\pm$ 0.94 <sup>c</sup>   |
| Propylene glycol alginate/xanthan |                              | Gum arabic/xanthan           |                                |                                  |                                  |                                |
| Storage period (weeks)            |                              | Storage period (weeks)       |                                |                                  |                                  |                                |
| 0                                 | 6                            | 0                            | 4                              | 14                               | 21                               | 33                             |
| 8000 rpm                          | 1.18 $\pm$ 0.39 <sup>a</sup> | 8000 rpm                     | 1.30 $\pm$ 0.47 <sup>a</sup>   | 1.21 $\pm$ 0.42 <sup>a</sup>     | 1.50 $\pm$ 0.73 <sup>a</sup>     | 1.63 $\pm$ 0.75 <sup>a,b</sup> |
| 9500 rpm                          | 1.35 $\pm$ 0.49 <sup>a</sup> | 9500 rpm                     | 1.48 $\pm$ 0.85 <sup>a</sup>   | 1.58 $\pm$ 0.77 <sup>a</sup>     | 2.25 $\pm$ 0.93 <sup>a,b</sup>   | 2.45 $\pm$ 1.21 <sup>b</sup>   |
| 20,500 rpm                        | 1.24 $\pm$ 0.44 <sup>a</sup> | 24,000 rpm                   | 1.48 $\pm$ 0.67 <sup>a,b</sup> | 1.11 $\pm$ 0.32 <sup>a</sup>     | 1.56 $\pm$ 0.63 <sup>a,b,c</sup> | 2.00 $\pm$ 0.89 <sup>c</sup>   |

a–c: Different superscripts mean that the results in each row for each polysaccharide are significantly different ( $p < 0.05$ ).

weeks), whereas in the EY/X or GA/X-stabilized emulsions the panellists reported a weak perception of rancid flavour only after 21 weeks ( $2.00 \pm 0.93$  and  $1.63 \pm 0.75$ – $2.25 \pm 1.28$ , respectively). Finally, the statistical analysis between salad dressings of similar initial droplet size, stabilized with mixtures of GA/X or PGA/X and homogenized at 20,500 rpm and 9500 rpm, respectively, revealed no significant differences among them.

#### 4. Conclusions

It can be concluded that the polysaccharides, gum arabic and propylene glycol alginate, have the ability to inhibit lipid oxidation in addition to their stabilizing/emulsifying capacity. This is probably due to their amphiphilic character and partly due to the presence of xanthan. Gum arabic, in particular, more effectively suppressed the oxidation during the storage period. This is attributed to its better surface-active properties in comparison to propylene glycol alginate. Lipid oxidation was not affected by the oil droplet size, as demonstrated by peroxide value measurements and sensory evaluation.

#### References

- Boskou, D., & Visioli, F. (2003). Biophenols in olive oil and olives. In M. Pilar Vaquero, T. Garcia-Arias, A. Carbajal, & F. J. Sanchez-Muniz (Eds.), *Bioavailability of micronutrients and minor dietary compounds. Metabolic and technological aspects* (pp. 161–169). Kerala: Research Signpost.
- Dickinson, E. (1992). *An introduction to food colloids*. Oxford: Oxford University Press.
- Dickinson, E., & Stainsby, G. (1982). *Colloids in food*. London: Applied Science Publishers.
- EU. (1991). Regulation 2568. Characteristics of olive oil and olive residue oil and the relevant methods of analysis. Official J. L. 248.
- Garcia Mesa, J. A., Jimenez-Marquez, A. J., Beltran-Maza, G., & Friaiz-Ruiz, L. (1998). Thermooxidation of different vegetable oils used in deep frying on dietary fat intake. *Nutrition Reviews*, 49, 375–376.
- Jacobsen, C., Hartvigsen, K., Lund, P., Meyer, A. S., Adler-Nissen, J., Holstborg, J., et al. (1999). Oxidation in fish-oil-enriched mayonnaise. 1. Assessment of propyl gallate as an antioxidant by discriminant partial least squares regression analysis. *European Food Research Technology*, 210, 13–30.
- Jellinek, G. (1984). *Sensory evaluation of food*. Chichester: Ellis Horwood.
- Karadjova, I., Zachariadis, G., Boskou, G., & Stratis, J. (1998). Electrothermal atomic adsorption spectrometric determination of aluminium, cadmium, chromium, copper, iron, manganese, nickel and lead in olive oil. *Journal of Analytical Atomic Spectrometry*, 13(3), 201–204.
- Matsumura, Y., Egami, M., Satake, C., Maeda, Y., Takahashi, T., Nakamura, A., et al. (2003). Inhibitory effects of peptide-bound polysaccharides on lipid oxidation in emulsions. *Food Chemistry*, 83(1), 107–119.
- Matsumura, Y., Satake, C., Egami, M., & Mori, T. (2000). Interaction of gum arabic, maltodextrin and pullulan with lipids in emulsions. *Bioscience Biotechnology and Biochemistry*, 64(9), 1827–1835.
- McClements, D. J., & Decker, E. A. (2000). Lipid oxidation in oil-in-water emulsions: impact of molecular environment on chemical reactions in heterogeneous food systems. *Journal of Food Science*, 65(8), 1270–1282.
- McClements, D. J. (1999). *Food emulsions: principles, practice and techniques*. Boca Raton: CRC Press.

- Mei, L. Y., McClements, D. J., & Decker, E. A. (1998a). Iron-catalyzed lipid oxidation in emulsion as affected by surfactant, pH and NaCl. *Food Chemistry*, 61(3), 307–312.
- Mei, L. Y., McClements, D. J., & Decker, E. A. (1998b). Evidence of iron association with emulsion droplets and its impact on lipid oxidation. *Journal of Agricultural and Food Chemistry*, 46(12), 5072–5077.
- Min, D. B., & Boff, J. M. (2002). Lipid oxidation of edible oil. In C. C. Akoh & D. B. Min (Eds.), *Food lipids – chemistry, nutrition and biotechnology* (pp. 335–363). New York: Springer.
- Nuchi, C. D., Hernandez, P., McClements, D. J., & Decker, E. A. (2002). Ability of lipid hydroperoxides to partition into surfactant micelles and alter lipid oxidation rates in emulsions. *Journal of Agricultural and Food Chemistry*, 50(19), 5445–5449.
- Onsøyen, E. (1999). Alginates. In A. Imeson (Ed.), *Thickening and gelling agents for food* (Vol. 2, pp. 22–44). Gaithersburg: Aspen Publishers Inc.
- Osborn, H. T., & Akoh, C. C. (2003). Copper-catalyzed oxidation of a structured lipid-based emulsion containing  $\alpha$ -tocopherol and citric acid: influence of pH and NaCl. *Journal of Agricultural and Food Chemistry*, 51(23), 6851–6855.
- Osborn, H. T., & Akoh, C. C. (2004). Effect of emulsifier type, droplet size, and oil concentration on lipid oxidation in structured lipid-based oil-in-water emulsions. *Food Chemistry*, 84(3), 451–456.
- Paraskevopoulou, A., Athanasiadis, I., Blekas, G., Koutinas, A. A., Kanellaki, M., & Kiosseoglou, V. (2003). Influence of polysaccharide addition on stability of a cheese whey kefir–milk mixture. *Food Hydrocolloids*, 17(5), 615–620.
- Paraskevopoulou, A., Boskou, D., & Kiosseoglou, V. (2005). Stabilization of olive oil–lemon juice emulsion with polysaccharides. *Food Chemistry*, 90(4), 627–634.
- Paraskevopoulou, A., Kiosseoglou, V., Alevisopoulos, S., & Kasapis, S. (1997). Small deformation properties of model salad dressings prepared with reduced cholesterol egg yolk. *Journal of Texture Studies*, 28(2), 221–237.
- Romero, A., Guesta, C., & Sanchez-Muniz, F. J. (1998). Behaviour of extra virgin olive oil in potato frying: thermooxidative alteration of the fat content in the fried food. *Grasas Aceites*, 49, 370–378.
- Roozen, J. P., Frankel, E. N., & Kinsella, J. E. (1994). Enzymic and autoxidation of lipids in low fat foods: model of linoleic acid in emulsified hexadecane. *Food Chemistry*, 50(1), 33–38.
- Shimada, K., Fujikawa, K., Yahara, K., & Nakamura, T. (1992). Antioxidative properties of xanthan on the autoxidation of soybean oil in cyclodextrin emulsion. *Journal of Agricultural and Food Chemistry*, 40(6), 945–948.
- Shimada, K., Muta, H., Nakamura, Y., Okada, H., Matsuo, K., Yoshioka, S., et al. (1994). Iron-binding property and antioxidative activity of xanthan on the autoxidation of soybean oil in emulsion. *Journal of Agricultural and Food Chemistry*, 42(8), 1607–1611.
- Shimada, K., Okada, H., Matsuo, K., & Yoshioka, S. (1996). Involvement of chelating action and viscosity in the antioxidative effect of xanthan in an oil/water emulsion. *Bioscience, Biotechnology and Biochemistry*, 60(1), 125–127.
- Trichopoulou, A. (1995). Olive oil and breast cancer. *Cancer Causes Control*, 6, 475–476.
- Urlacher, B., & Noble, O. (1999). Xanthan gum. In A. Imeson (Ed.), *Thickening and gelling agents for food* (Vol. 13, pp. 284–311). Gaithersburg: Aspen Publishers Inc.
- Wareing, V. M. (1999). Exudate gums. In A. Imeson (Ed.), *Thickening and gelling agents for food* (Vol. 5, pp. 86–99). Gaithersburg: Aspen Publishers Inc.