Food Chemistry 167 (2015) 340–348

Contents lists available at ScienceDirect

Food Chemistry

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

Effects of xanthan–locust bean gum mixtures on the physicochemical properties and oxidative stability of whey protein stabilised oil-in-water emulsions

Hanna Khouryieh [a,*], Goutham Puli [b], Kevin Williams [b], Fadi Aramouni [c]

[a] Food Processing and Technology Program, Western Kentucky University, Bowling Green, KY, USA
[b] Department of Chemistry, Western Kentucky University, Bowling Green, KY, USA
[c] Food Science Institute, Kansas State University, Manhattan, KS, USA

ARTICLE INFO

Article history:
Received 14 February 2014
Received in revised form 2 June 2014
Accepted 2 July 2014
Available online 11 July 2014

Keywords:
Menhaden oil
Xanthan gum
Locust bean gum
Emulsion
Oxidative stability

ABSTRACT

The effects of xanthan gum (XG)–locust bean gum (LBG) mixtures (0.05, 0.1, 0.15, 0.2 and 0.5 wt%) on the physicochemical properties of whey protein isolate (WPI) stabilised oil-in-water (O/W) emulsions containing 20% v/v menhaden oil was investigated. At higher concentrations, the apparent viscosity of the emulsions containing XG/LBG mixtures was significantly higher ($p < 0.05$) than the emulsions containing either XG or LBG alone. Locust bean gum showed the greatest phase separation, followed by XG. Microstructure images showed depletion flocculation at lower biopolymer concentrations, and thus led to an increase in creaming instability and apparent viscosity of the emulsions. Addition of 0.15, 0.2 and 0.5 wt% XG/LBG mixtures greatly decreased the creaming of the emulsions. The rate of lipid oxidation for 8-week storage was significantly lower ($p < 0.05$) in emulsions containing XG/LBG mixtures than in emulsions containing either of the biopolymer alone.

1. Introduction

The benefits of consuming long chain omega-3 polyunsaturated fatty acids (ω-3 PUFA) have been widely reported (Ruxton, Reed, Simpson, & Millington, 2004; Sahena et al., 2009; Shahidi & Miraliakbari, 2004; Siddiqui et al., 2004). However, PUFA are highly susceptible to oxidation during storage, which creates various aldehyde and ketone products that render unacceptable odour and flavour in PUFA containing foods (McClements & Decker, 2008). In addition, lipid oxidation products, such as lipid peroxides, unsaturated aldehydes and malonaldehydes, and several cholesterol oxidation products (Addis, 1986; Esterbauer, Schaur, & Zollner, 1990; Sangeetha, Das, Koratkar, & Suryaprabha, 1990) have a negative impact on human health due to their cytotoxic and genotoxic effects (Fang, Vaca, Valsta, & Mutanen, 1996; Kanner, 2007). The rate of oxidation of PUFA can be decreased by the addition of many synthetic or natural antioxidants.

Oil-in-water emulsions, in which the lipid portion is dispersed as miniscule droplets within an aqueous continuous phase, can be effective vehicles for delivering ω-3 PUFA into food systems as they can be easily incorporated into many food matrix types. One approach to inhibit lipid oxidation in oil-in-water emulsions is to use food biopolymers, such as proteins and polysaccharides, that can bind transition metals and take them away from the emulsion droplet surface (Chen, Mcclements, & Decker, 2010).

Several studies have demonstrated that proteins dispersed in the continuous phase of oil-in-water emulsions can inhibit deleterious oxidation reactions by a combination of metal chelation and free radical scavenging (Elias, McClements, & Decker, 2005; Elias, McClements, & Decker, 2007). Whey protein isolate (WPI) is a surface-active whey protein product that is commonly used as an emulsifier in the food industry because of its good emulsifying and stabilizing properties. The two major constituents of WPI are β-lactoglobulin and α-lactalbumin (Cayot & Lorient, 1997). They contain cysteyl residues, disulphide bonds and thiol functional groups, which can scavenge free radicals to inhibit lipid oxidation. Therefore, WPI-stabilised emulsions may act as an antioxidant system (Sun, Sundaram, & Mark, 2007).

Polysaccharides have been widely used as thickening, stabilising, and gelling agents in the food industry for the control of microstructure, texture, flavour and shelf-life. The function of polysaccharides as emulsion stabilisers is not only attributable to their ability to increase the viscosity of the continuous phase and inhibit coalescence (Chen et al., 2010), but also to their effect on protein adsorption at the oil/water interface. Addition of an incompatible

* Corresponding author. Tel.: +1 270 745 4126; fax: +1 270 745 5946.
E-mail address: hanna.khouryieh@wku.edu (H. Khouryieh).

http://dx.doi.org/10.1016/j.foodchem.2014.07.009
0308-8146/

polysaccharide can enhance protein adsorption at interfaces and increase emulsion stability (Tolstoguzov, 1997). The antioxidant ability of polysaccharides in emulsions has also been reported (Chen et al., 2010; Kishk & Al-Sayed, 2007; Paraskevopoulou, Boskou, & Parakesvopoulou, 2007).

Xanthan gum is an anionic heteropolysaccharide produced by the microorganism *Xanthomonas campestris*. Xanthan's backbone consists of (1 → 4) β-D-glucopyranosyl units, which is substituted at C-3 on every other glucose residue with a charged trisaccharide sidechain. The trisaccharide chain consists of a D-glucuronic acid unit between two D-mannose units. Approximately half of the terminal D-mannose unit contain a pyruvic acid residue linked via a keto group to the 4 and 6 positions, while the D-mannose linked to the main chain contains an acetyl group at position O-6 (Garcia-Ochoa, Santos, Casas, & Gomez, 2000). Locust bean gum (LBG) is a neutral polysaccharide composed of mannose and galactose units and, therefore, belongs to the category of galactomannans. The physicochemical properties of galactomannans are strongly influenced by the galactose content and the distribution of the galactose units along the main chain. Longer galactose side chains produce stronger synergistic interactions with other polymers and greater functionality (Renou, Odile, Malhiac, & Grisel, 2013). A synergistic interaction occurs between xanthan gum (XG) and galactomannans, such as guar and locust bean gum (LBG) in solutions to result in enhanced viscosity or gelation (Dea, Morris, Rees, & Welsh, 1997; Goycoolea, Richardson, & Morris, 1995; Khouryieh, Herald, Aramouni, & Alavi, 2006; Khouryieh, Herald, Aramouni, Bean, & Alavi, 2007a,b; Long et al., 2013; Tako, 1993; Tako & Nakamura, 1984, 1989). Despite the great number of studies dealing with milk protein–polysaccharide emulsions, relatively limited studies have been performed on the influence of combined ionic and neutral hydrocolloids in oil-in-water emulsions. The effect of the synergistic interaction between XG (negatively charged gum) and LBG (neutral gum) on the stability of oil-in-water emulsions has never been investigated. Therefore, the objectives of this study were to investigate the effect of XG/LBG mixtures (ranged from 0 to 0.5 wt%) on the stability of WPI stabilised oil-in-water emulsions and to determine their physicochemical characteristics. The ultimate goal is to determine if the oxidative stability of WPI stabilised oil-in-water emulsions could be increased better than using either hydrocolloid alone.

2. Materials and methods

2.1. Materials and chemicals

Menhaden oil (14:0 myristic acid 6–9%, 16:0 palmitic acid 15–20%, 16:1 palmitoleic acid 9–14%, 18:0 stearic acid 3–4%, 18:1 oleic acid 5–12%, 18:2 linoleic acid < 3%, 18:3 linolenic acid < 3%, 20:4 arachidonic acid < 3%, 18:4 octadecatetraenoic 2–4%, 20:5 eicosapentaenoic 10–15% and 22:6 docosahexaenoic 8–15%), xanthan gum, locust bean gum, iron(II) chloride tetrahydrate, xylenol orange disodium salt, 2-thiobarbituric acid, iron(III) chloride, methanol, 1-butanol, and 1,1,3,3-tetraethoxypropane were purchased from Sigma Aldrich, Co. (St. Louis, MO, USA). Whey protein isolate was obtained from Davisco Foods International, Inc. (Le Sueur, MN). Hydrogen peroxide was obtained from Fischer Scientific (Fair Lawn, NJ, USA). All other chemicals and solvents were of analytical grade. Deionised water was used to prepare all the emulsions.

2.2. Emulsion preparation

Stock solutions WPI (10%, w/v), XG (1%, w/v) and LBG (1%, w/v) were prepared separately by dissolving the required amount of WPI, XG and LBG powders into deionised water at room temperature, followed by continuous stirring with a magnetic stirrer for 6 h to ensure complete dispersion. The stock solutions of XG and LBG were heated in a water bath at 80 °C for 30 min and 0.02% (v/v) sodium azide was added to prevent microbial growth.

The oil-in-water emulsions were prepared first by slowly mixing the required amount of menhaden oil into WPI solution; to this either XG, LBG, or XG-LBG mixtures solution was added. The mixed solutions were first emulsified using a lab scale power homogenizer (PowerGen 500, Fischer Scientific, Fair Lawn, NJ, USA) for 5 min at 300 W output power. The emulsions were then sonicated in ultrasonic water bath (B 1500A-MT, VWR, San Francisco, CA, USA) for 1 min at high speed. The final composition of the oil-in-water emulsions was 20% v/v menhaden oil, 2 wt% WPI, and either 0, 0.05, 0.1, 0.15, 0.2 and 0.5 wt% XG, LBG or XG/LBG mixtures (total weight of both gums). The pH of the final emulsion was 6.6. XG and LBG solutions were blended in 50:50 synergistic ratios for the XG/LBG mixtures. Emulsions were stored in closed 20 ml bottles at room temperature (23 °C) in the dark for 8 weeks for the measurements of lipid oxidation.

2.3. Particle size characterisation

The droplet size of the emulsions was determined using the Malvern Mastersizer 2000 (Malvern Instruments, Ltd., Worcestershire, UK). The surface-area-average diameter (d_{32}) of the emulsion droplets was calculated as $d_{32} = \sum n_i d_i^3 / \sum n_i d_i^2$, where n_i is the number of droplets of diameter d_i. To prevent multiple scattering effects, the emulsions were diluted prior to particle size measurements with distilled water to keep the droplet concentration below 10^{-2} volume fraction. The refractive indices of the dispersed and continuous phases used in the calculations were 1.45 for oil and 1.33, respectively.

2.4. Viscosity measurements

Emulsion viscosity was measured using a Brookfield DV-II + Pro Viscometer (Brookfield Engineering, Middleboro, MA, USA). Readings were taken at a speed of 100 rpm using spindle No. 2 at 22 °C within 24 h of the emulsion preparation. Viscosities were determined at day 1 of emulsion preparation, making sure that all samples were measured within the same timeframe.

2.5. Creaming index

Creaming index (CI) provides indirect information about the extent of the droplet aggregation in an emulsion. The more the aggregation, the larger the flocs and the faster the creaming (Sun et al., 2007). Immediately after preparation, emulsions were transferred to 21 mm diameter, 70 mm high glass test tubes and were sealed to prevent evaporation. They were kept at ambient temperature and the movement of any creaming boundary was tracked for period of 15 d. Emulsion separation into a top cream layer and a bottom serum layer was monitored. The total emulsion height (H_T) and serum layer height (H_S) were measured. The creaming index was reported as CI (%) = $100(H_S/H_T)$.

2.6. Microstructure

Microstructure of the emulsions was studied using a deconvolution microscopy (Zeiss Axioplan IIe imaging, Carl Zeiss microscopy LLC, Thornwood, NY, USA) equipped with an attached camera. A 10× objective magnification was used to visualise the images at 50 mm below the cover slip. A 50 µl of freshly made emulsion was placed on a 1.2 mm thick glass slide. A coverslip was placed on the top of the droplet ensuring no air gap or bubbles are seen.