Food Hydrocolloids 38 (2014) 1–10

Contents lists available at ScienceDirect

Food Hydrocolloids

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

Premix membrane emulsification to produce oil-in-water emulsions stabilized with various interfacial structures of whey protein and carboxymethyl cellulose

Rikkert Berendsen, Carme Güell, Olivier Henry, Montserrat Ferrando*

Departament d'Enginyeria Química, Universitat Rovira i Virgili, Av. Països Catalans 26, 43007 Tarragona, Spain

ARTICLE INFO

Article history:
Received 16 September 2013
Accepted 6 November 2013

Keywords:
Membrane emulsification
Whey protein
Carboxymethyl cellulose
Protein–polysaccharide complexes
Oil-in-water emulsions
Multi-layer emulsions

ABSTRACT

Premix Membrane Emulsification (ME) with Shirasu Porous Glass membranes of 10 μm pore size enabled to produce oil-in-water (O/W) emulsions with different interfacial structures made of whey protein isolate (WPI) and carboxymethyl cellulose (CMC). Emulsions were stabilized by one interfacial layer, made of WPI (mono-layer emulsion) or 0.5 %wt WPI–0.25 %wt CMC complex (complex emulsion), or by two interfacial layers: one layer made of WPI and the second made of CMC (bi-layer emulsion) or WPI–CMC complex (sequential emulsion). Although the adsorption between the several layers was confirmed by Surface Plasmon Resonance (SPR), only O/W emulsions stabilized by one interfacial layer did not coalescence after homogenization. Mono-layer and complex emulsions were stable after emulsification with a 8.7 μm and 14.4 μm mean droplet size, respectively, although a significant amount of much smaller droplets contributed to increase droplet dispersion giving span values of 1.8 and 3.2 for mono-layer and complex emulsions, respectively.

Regarding oxidation rate, TBARS in complex emulsions increased much faster than in mono-layer emulsions. Adsorption data at a hydrophobic interface and the electrical charge of the WPI–CMC complex suggested that it formed a thick (2.2 nm) but less dense (1.40 g cm^{-3}) interface than WPI (2.59 g cm^{-3}) with a negative charge able to attract any transition metal ion and promote lipid oxidation. Premix ME should be further optimized to obtain multi-layered interfaces with an external positive layer, e.g. made of WPI.

1. Introduction

The use of emulsions is common in food industry, many products we daily consume are in fact oil-in-water (O/W) emulsions. Some typical examples are milk, cream, salad dressings, mayonnaise, soups and sauces.

In food emulsions, stability, quality and the ability to deliver functional compounds are of great importance (Guzey & McClements, 2006). Emulsions are kinetically unstable and tend to spontaneously coalesce minimizing the interfacial area (Capek, 2004). Average droplet size and droplet size dispersion are two properties which determine the physical stability of the emulsion. To prevent emulsion breakage, surfactants are added to provide a layer which lowers the interfacial tension between the oil and water phases (Kralova & Sjöblom, 2009).

Food emulsions are traditionally produced by colloid mills, rotor-stator systems and high pressure homogenizers, these produce emulsions with small droplet size but with a relatively high droplet size dispersion. Furthermore, they require a high energy input and high shear forces to produce emulsions. Membrane emulsification (ME) is a low energy technique that requires less surfactants, which produces emulsions with low polydispersity and reduces damage to emulsifiers sensitive to shear stress, in contrary to the traditional methods (Joscelyne & Trägårdh, 1999; Nazir, Schroën, & Boom, 2010; Silva, Cerqueira, & Vicente, 2012; Vladisavljević, Surh, & McClements, 2006).

There are different methods to operate membrane emulsification (ME): Direct (or cross-flow) ME and Premix ME. In Direct ME, the dispersed phase is forced, using low pressure, to permeate through a membrane into the continuous phase. In Premix ME, emulsions are produced in a two-step process: i) production of a coarse emulsion, and ii) the entire coarse emulsion passes through the pore structure of the membrane. Premix ME has some advantages over direct ME: the optimal transmembrane fluxes with regard to droplet size uniformity are one or two orders of magnitude

* Corresponding author. Tel.: +34 977558505.
E-mail addresses: rikkert.berendsen@urv.cat (R. Berendsen), carme.guell@urv.cat (C. Güell), olivier.henry@urv.cat (O. Henry), montse.ferrando@urv.cat (M. Ferrando).

0268-005X/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.foodhyd.2013.11.005

higher than those of direct ME, and the experimental set-up is generally simpler and easier to control and operate than in direct ME (Nazir et al., 2010; Vladisavljević et al., 2006).

In food industry, whey proteins have been widely used to stabilize O/W emulsions because they show good surface active properties and form a viscoelastic layer at the interface (Bos & van Vliet, 2001) that improves the encapsulation properties due to a barrier effect. Nevertheless, whey protein-stabilized emulsions are rather heat sensitive when whey protein denaturates on the O–W interface, the protein unfolds and can form irreversible aggregates between emulsion droplets (Dickinson & Parkinson, 2004). A strategy to improve interfacial properties has been the use of protein–polysaccharide complexes which combines the positive properties of each of them: i) proteins show good adsorption to O/W interfaces and ii) polysaccharides enable to form a thicker stabilizing layer that is capable of protecting droplets against aggregation over a wide range of unfavourable conditions, such as thermal shock treatment and the addition of calcium salts (Chanamai & McClements, 2002). In addition, emulsions stabilized by complex interfaces made of protein and polysaccharides have been suggested to improve chemical stability against oxidation reactions, e.g., interactions between lipids and metal ions can be minimized by controlling the interfacial charge, thickness and density (Dickinson, 2008, 2011; Grigoriev & Miller, 2009; Katsuda, McClements, Miglioranza, & Decker, 2008; McClements & Decker, 2000; Rodríguez Patino & Pilosof, 2011).

The formation of protein–polysaccharide electrostatic complexes and coacervates mainly depends on pH, ionic strength and protein to polysaccharide ratio (Schmitt & Turgeon, 2011; Semenova, 2007). Applications of these complexes in food industry encompass: to form and stabilize emulsions possibly in combination with the design of multi-layered structures, micro- and nano-encapsulation processes, to form gels and to recover proteins from industrial by-products (Turgeon, Schmitt, & Sanchez, 2007).

In this study we used whey protein isolate (WPI) which is a by-product in cheese-making and has excellent emulsifying properties (Dybowska, 2011), and carboxymethyl cellulose (CMC) which is widely used in food for its physical properties and its low cost. In addition CMC can form electrostatic complexes with WPI over a range of pH and ionic strength (Hansen, Hidalgo, & Gould, 1971).

The aim of this study was to produce O/W emulsions with complex interfacial structures made of WPI and CMC using Premix ME, and determine droplet and oxidation stability of those emulsions. To do so, we determined the state diagram of the WPI/CMC system and set the conditions (WPI to CMC ratio, pH and ionic strength) required to form WPI–CMC complexes. The adsorption properties of the different interfacial structures made of WPI and CMC were measured by Surface Plasmon Resonance. Moreover Premix ME was applied to produce O/W emulsions stabilized by the different interfacial structures. To the author's knowledge no application of Premix membrane emulsification to obtain single emulsions stabilized by protein-polysaccharide complexes has been previously reported in literature. To determine to what extent the several interfacial structures affected the physical and oxidative stability of O/W emulsions, we monitored during 2-weeks droplet size distribution and the oxidative degradation of the oil phase.

2. Materials and methods

2.1. Materials

Whey protein isolate (BiPRO) lot no. JE 034–7–440–6 (Davisco Foods International. Inc., Le Sueur, MN), with a reported protein content of 98.1% on dry basis, was dissolved in distilled water to obtain a 2.0%wt solution, stirred for 2*h* at room temperature, and kept overnight in a fridge. Carboxymethyl cellulose, sodium salt (Acros Organics), with a M_w of 250,000 and a degree of substitution of 0.7, was dissolved in distilled water to obtain a 2.0 %wt solution and stirred for 2 h at room temperature. Acetic acid 96% (Panreac) together with sodium azide (Sigma–Aldrich), were dissolved in distilled water to obtain 0.02 M acetic acid and 0.04 %wt sodium azide (NaN_3). WPI and CMC solutions were mixed with the acetic acid solution to obtain 1.0 %wt WPI, 0.25 %wt CMC and 0.5 %wt WPI–0.25 %wt CMC in 0.01 M acetic acid and 0.02 %wt NaN_3. The solution pH was adjusted to 3.80 with 1 M HCl (Panreac) or 1 M NaOH (Sigma–Aldrich). All distilled water used in this research had an electrical conductivity of 2 μS cm^{-1}. Crude sunflower oil was kindly provided by Cargill S.L.U. Reus, Spain.

2.2. Methods

2.2.1. WPI/CMC state diagram

Aqueous solutions containing 0.5 %wt of WPI and different concentrations of CMC were prepared to obtain a final ratio of 1:0, 1:1, 2:1, 3:1 and 4:1, respectively. All mixtures contained 0.01 M acetic acid, 0.02 %wt NaN_3 and the pH was adjusted from 6 to 3 with 1 M HCl or 1 M NaOH solutions. Turbidity and pH of each mixture was measured right after preparation to identify a clear solution, a cloudy solution or the formation of a precipitate. Turbidity was quantitatively measured from the Transmission (T) profiles obtained with a Turbiscan Lab Expert (Formulaction, France).

2.2.2. Zeta-potential measurement

Laser Doppler micro-electrophoresis was used to measure ζ-potential of WPI, CMC and a WPI-CMC complex by means of a Zetasizer Nano ZS (Malvern Instruments). Particles were detected at a measurement position of 2 mm using a 633 nm laser at 25 °C. Solutions of 1 %wt WPI, 0.25 %wt CMC and 0.5 %wt WPI–0.25 %wt CMC in 0.01 M acetic acid, 0.02 %wt NaN_3 at a pH of 3.8 were prepared and centrifuged at 1000 *g* for 30 min to remove impurities. After that, the ζ-potential distribution of each of them was measured. The reported ζ-potential values are an average of six measurements and calculated by the Smoluchowski equation. All measured ζ-potential distributions gave a monomodal distribution.

2.2.3. Surface plasmon resonance measurements

Surface plasmon resonance is an optical measurement technique highly sensitive to localized refractive index changes occurring at the surface of a metal-dielectric interface. A typical set-up includes shining a monochromatic light beam through a high refractive index glass prism coated with a 50 nm thick gold layer. Under total internal reflectance conditions, the evanescent wave created at a specific incident angle will result in a plasmonic wave seen as a sharp decrease in intensity of the reflected beam. The position of the incident angle is strongly dependent on the refractive index changes occurring at the metal surface making surface plasmon resonance an ideal tool for studying the adsorption of biomolecules and biopolymers at surfaces (Fang, Wang, Du, & Zhu, 2009; Gopinath, 2010; Jung, Campbell, Chinowsky, Mar, & Yee, 1998).

2.2.3.1. Surface plasmon resonance (SPR). A Biacore 3000 (GE Healthcare, Uppsala, Sweden) was used to study the surface adsorption of the various biopolymers to a gold sensor chip. The microfluidic cell divided the bare gold sensor chip in four flow cells (each of them with a surface area of 1.2 mm^2) used in series at a flow rate of 40 μL min^{-1}. Baseline was acquired by flowing 0.01 M acetic acid (pH 3.8) with 0.02 %wt NaN_3 (running buffer) over the