Innovative Food Science and Emerging Technologies 32 (2015) 79–90

Contents lists available at ScienceDirect

Innovative Food Science and Emerging Technologies

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

Physical and oxidative stability of whey protein oil-in-water emulsions produced by conventional and ultra high-pressure homogenization: Effects of pressure and protein concentration on emulsion characteristics

Essam Hebishy [a,c], Martin Buffa [a], Buenaventura Guamis [a], Anabel Blasco-Moreno [b], Antonio-José Trujillo [a,*]

[a] Centre Especial de Recerca Planta de Tecnologia dels Aliments (CERPTA), XaRTA, TECNIO, MALTA Consolider, Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain

[b] Servei d'Estadística Aplicada, Universitat Autònoma de Barcelona, 08193 Bellaterra, Barcelona, Spain

[c] Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, RiobambaEC060150, Ecuador

ARTICLE INFO

Article history:
Received 7 June 2014
Received in revised form 5 August 2015
Accepted 19 September 2015
Available online 9 October 2015

Keywords:
Ultra high-pressure homogenization (UHPH)
Whey protein
Submicron emulsions and oxidative stability

ABSTRACT

Oil-in-water pre-emulsions (15% sunflower + 5% olive oils) obtained by colloid mill homogenization (CM) at 5000 rpm using whey protein isolate at different levels (1, 2 and 4%) were stabilized by ultra high-pressure homogenization (UHPH, 100 and 200 MPa) and by conventional homogenization (CH, 15 MPa). Emulsions were characterized for their physical properties (droplet size distribution, microstructure, surface protein concentration, emulsifying stability against creaming and coalescence, and viscosity) and oxidative stability (hydroperoxide content and thiobarbituric acid reactive substances, TBARs) under light (2000 lux/m^2 for 10 days). UHPH produced emulsions with lipid droplets of small size in the sub-micron range (100–200 nm) and low surface protein with unimodal distribution when produced at 4% whey proteins and 200 MPa. All emulsions exhibited Newtonian behavior ($n \approx 1$). Long term physical stability against creaming and coalescence was observed in UHPH-emulsions, compared to those obtained by CM and CH. However, CH emulsions were highly stable against creaming (days) in comparison to the CM emulsions (hours). UHPH resulted in emulsions highly stable to oxidation compared to CM and CH treatments, especially when 100 MPa treatment was applied.

Industrial relevance: In the food, cosmetic and pharmaceutical sectors, industrial operators are currently interested in developing encapsulating systems to delivery bioactive compounds, which are generally hydrophobic, unstable and sensitive to light, temperature or/and oxygen. Ultra high-pressure homogenization is capable of producing stable submicron emulsions (<1 μm) with a narrow size distribution, inducing more significant changes in the interfacial protein layer thus preventing droplet coalescence and also inhibit lipid oxidation. The present study suggests that emulsions produced by whey protein (4%) treated by ultra high-pressure homogenization have a good physical stability to flocculation, coalescence and creaming and also high stability to lipid oxidation, opening a wide range of opportunities in the formulation of emulsions containing bioactive components with lipid nature.

1. Introduction

Sub-micron emulsions have a number of unique functional attributes that have led them to be utilized within an increasing number of industrial products, including foods, pharmaceuticals, cosmetics, personal care products and chemicals. Due to their size characteristics, sub-micron emulsions are expected to maintain high stability against creaming and coalescence. Since they display large interfacial area/volume ratios, a sufficiently high concentration of surfactant (i.e. emulsifiers) has to be added to enable the new surface area developed by nano droplets to be rapidly coated during the emulsification process (Mason, Wilking, Meleson, Chang, & Graves, 2006); this should avoid or limit shear- and turbulence-induced coalescence, and ensure higher physical stability of emulsion for a long period of time.

The formation of sub-micron emulsions requires high energy inputs. Current equipment used for emulsion preparation includes colloid mills, microfluidizers, sonicators or high-pressure homogenizers (Stang, Schuchmann, & Schubert, 2001). HP-homogenizers of the piston-gap type are developed by manufacturers such as Avestin™, APV™ or Stansted Fluid Power™; the processed liquid is brought to high pressure in few seconds in the pressure intensifier then forced through a very small orifice, the valve gap of few micrometres in width. The high pressure homogenizer developed by Stansted Fluid Power™ used in the present study consists of one or two piston intensifier(s) able to generate high pressure up to 400 MPa and a high-pressure valve (HP-

* Corresponding author at: Tecnologia dels Aliments, Departament de Ciència Animal i dels Aliments, Facultat de Veterinària, Universitat Autònoma de Barcelona, 08193 Bellaterra, Spain. Tel.: +34 93 581 32 92; fax: +34 93 581 20 06.

E-mail address: toni.trujillo@uab.es (A.-J. Trujillo).

http://dx.doi.org/10.1016/j.ifset.2015.09.013
1466-8564/

valve) equipped with ceramic needles and seat of specially studied design. In such HP-homogenizers, the fluid under pressure is forced through a small orifice of some micrometers width to the HP-valve gap (Floury, Bellettre, Legrand, & Desrumaux, 2004). The fluid accelerates in a very short distance to a very high velocity and the resulting strong pressure gradient between the inlet and outlet of the HP-valve generates intense shear forces and extensional stress through the valve gap (Stevenson & Chen, 1997). Cavitation, turbulence and impact with solid surfaces take place at the outlet of the valve gap (Diels, Callewaert, Wuytack, Masschalck, & Michiels, 2005; Floury et al., 2004). In different emulsification systems including UHPH, due to shear effects and conversion of kinetic energy into heat, the flow through the HP-valve is accompanied by short-life heating phenomena that can be controlled by efficient cooling devices (Grácia-Juliá et al., 2008; Picart et al., 2006). All these mechanical forces are expected to disrupt particles down to the submicron range (McClements, 2005; Perrier-Cornet, Marie, & Gervais, 2005), increasing the interfacial area and resulting in considerable interactions between the adsorbed emulsifier at the interface of the oil droplets forming a more rigid interfacial layer (Lee, Lefèvre, Subirade, & Paquin, 2009).

High-pressure homogenization is an important process used in the preparation or stabilization of emulsions and suspensions. The result, from a practical point of view, is a much reduced tendency for creaming, contributing to an enhanced physical stability of the homogenized emulsions (Lee et al., 2009).

The stability and the emulsion formation become easier when using an emulsifier, which is adsorbed at the interface between oil and water and can lower the interfacial tension and prevent coalescence of droplets by increasing repulsion forces between droplets. Globular proteins derived from milk are widely used as natural emulsifiers to enhance the formation and stability of oil-in-water (O/W) emulsions, e.g., whey proteins such as β-lactoglobulin, α-lactalbumin, and bovine serum albumin (Dickinson, 2003; Livney, 2010). Whey protein isolate (WPI) is an excellent emulsifier and widely used in food emulsions due to its surface-active property forming a protective film and provides structural support for oil droplets through a combination of electrostatic and steric interactions (Djordjevic, Kim, McClements, & Decker, 2004). Furthermore, WPI can inhibit the oxidation of dispersed phase by preventing the penetration of prooxidants within the emulsified droplet (Dickinson, 2008).

Contrasting results in whey protein-stabilized emulsions subjected to high-pressure homogenization can be found in the literature. Desrumaux and Marcand (2002) showed that during ultra high-pressure emulsification, the conformation of whey proteins is changed (denaturation occurs), affecting their emulsifying properties. They found an optimum pressure of ~100 MPa, in which *d*3,2 and Span values reached a minimum. However, Perrier-Cornet et al. (2005) proved that at pressures above 200 MPa, the adsorption rate of whey proteins significantly increased (60%) corresponding to a very narrow particle size of sunflower oil.

Although several researchers have examined the effect of homogenization pressures (up to 300 MPa) on the physical stability of emulsion (Cortés-Muñoz, Chevalier-Lucia, & Dumay, 2009 Desrumaux & Marcand, 2002; Floury, Desrumaux, Axelos, & Legrand, 2003; Kiokias, Reiffers-Magnani, & Bot, 2004), only some researchers have studied the effect of pressures up to 100 MPa on the oxidative stability of emulsions (Horn, Nielsen, Jensen, Horsewell, & Jacobsen, 2012; Kuhn & Cunha, 2012; Sørensen et al., 2007). There is not much literature evidence regarding any association of ultra-high homogenization pressures (up to 300 MPa) with oxidative deterioration of the emulsions. A further elucidation of the effect of varying homogenization pressure (that generated varying droplet sizes) on oxidative deterioration of whey protein-stabilized O/W emulsions could contribute to control processing parameters for the production of high quality emulsified foods.

The aim of the present work was to study the physical and oxidative stability of emulsions produced by whey protein isolate (WPI) under various protein concentrations and pressures using the UHPH technology in comparison with conventional homogenization.

2. Material and methods

2.1. Materials

Whey protein isolate (WPI) was obtained from Lactalis (Prolacta 90, Retiers, France). The WPI contained 95.9 g dry solids per 100 g powder, and in dry basis (*w/w*), 1.04% non-protein nitrogen (NPN), 89.3% protein [(total N − NPN) × 6.38], 1.1% ash (including 0.27% calcium) and 1.6% lactose, as given by the producer. Protein constituents in the WPI corresponded mainly to β-lactoglobulin (β-Lg) and α-lactalbumin (α-La) (i.e. 68.5% β-Lg and 21.5% α-La per 100 g soluble protein) in addition to small amounts or traces of immune globulins, bovine serum albumin and lactoferrin.

Refined sunflower and olive oils were purchased from Gustav Heess Company (Barcelona, Spain). The characteristics and composition of oils according to the producer were: density (20 °C) = 0.921 and 0.913; acid value = 0.09 (mg KOH/g) and 0.11%; peroxide value (meqO_2/kg) = 0.02 and 0.5; absorbance (270 nm) = not determined and 0.29; unsaponifiable (% m/m) = <0.05 and <1.5%; C16:0 (%) = 6.34 and 11.94; C18:0 (%) = 3.97 and 3.30; C18:1 (%) = 26.65 and 75.23; C18:2 (%) = 61.02 and 6.75; C18: 3 (%) = 0 and 0.38 for sunflower and olive oils, respectively.

2.2. Preparation of emulsions

2.2.1. Preparation of protein dispersions.

WPI dispersions containing 1, 2 and 4% (*w/v*) were prepared using decalcified water by agitation with high speed mechanical blender (Frigomat machine, Guardamiglio, Italy) with two blenders at room temperature avoiding foam formation. Protein dispersions (pH ≈ 6.5–7) were stored overnight at 4 °C to allow protein hydration.

2.2.2. Homogenization treatments

After rehydration, protein dispersions (1, 2 and 4%) and oil (20%) were equilibrated at 20 °C before mixing. To prepare the pre-emulsions (or coarse emulsions), the oily dispersed phase (3 sunflower : 1 olive oil) was progressively added to the aqueous continuous phase containing WPI at room temperature and stirred for 5 min using the colloid mill (CM) homogenizer (E. Bachiller B. S.A, Barcelona, Spain) at maximum power (5000 rpm) (CM emulsion). This stainless toothache, corundum or perforated plate mill is used for fine grinding and homogenization of liquid, pasty and viscous products with even fineness up to 1 μm. The secondary or final emulsions were produced by the use of the following equipments.

Pre-emulsions were treated by UHPH using a Stansted high-pressure homogenizer (Model/DRG number FPG 11300:400 Hygienic Homogenizer, Stansted Fluid Power Ltd., UK) with a flow rate of 120 l/h. To minimize temperature retention after treatment, two spiral type heat-exchangers (Garvía, Barcelona, Spain) located behind the second valve were used. Emulsions were UHPH-treated at pressures of 100 and 200 MPa (single-stage) with inlet temperature (Tin) of 25 °C (UHPH emulsions). In the present study, other pressures above 200 MPa (i.e. 300 MPa) were also tested. However, in these emulsions particle size was excessively large so we decided not to continue working on this treatment.

Throughout the experiment, the Tin, the temperature after the homogenization valve (T1) and the temperature of the outlet product (T2) were monitored.

The conventional homogenization of the pre-emulsions was performed using APV Rannie Copenhagen Series Homogenizer (Model 40.120 H, single stage hydraulic valve assembly, Copenhagen, Denmark) with Tin of 60 °C at 15 MPa for a single stage (CH emulsions).