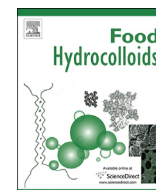
Food Hydrocolloids 71 (2017) 47–59

Contents lists available at ScienceDirect

Food Hydrocolloids

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



Design of novel emulsion microgel particles of tuneable size

Ophelie Torres, Brent Murray, Anwesha Sarkar*

Food Colloids and Processing Group, School of Food Science and Nutrition, University of Leeds, Leeds, LS2 9JT, UK

A R T I C L E I N F O

Article history:
Received 20 January 2017
Received in revised form
24 April 2017
Accepted 25 April 2017
Available online 26 April 2017

Keywords:
Emulsion microgel particles
Heat treated whey protein
Encapsulation
Cold gelation
Turbulent mixing

A B S T R A C T

In this study, we designed a one-step solvent-free route to prepare emulsion microgel particles, i.e., microgel particles containing several sub-micron sized emulsion droplets stabilised by heat-treated whey protein. The heat treatment conditions were optimized using aggregation kinetics via fluorimetry and dynamic light scattering. Emulsions were gelled and microgel particles were formed simultaneously via turbulent mixing with calcium ions using two specific processing routes (Extrusion and T-mixing). By varying the calcium ion concentration and mixing conditions, we identified the optimal parameters to tune the size and structure of the resultant emulsion microgel particles. Microscopy at various length scales (confocal laser scanning microscopy, scanning electron microscopy) and static light scattering measurements revealed a decrease in particle size (100–10 μm) with lower turbulent mixing time (ca. 4×10^{-4} s) and lower concentrations of calcium ions (0.1–0.02 M). Larger particle sizes (500–1000 μm) were achieved with an increase in the turbulent mixing time (ca. 2×10^{-2} s) and higher concentrations of calcium ions (1–1.4 M). Using gelation kinetics data (small deformation rheology) and theoretical considerations, creation of smaller sized emulsion microgel particles was explained by the increased flux of calcium ions to the denatured whey protein moieties coating the emulsion droplets, enabling faster gelation of the particle surfaces. These novel emulsion microgel particles of tuneable size formed as a result of complex interplay between calcium ion concentration, heat treatment of whey protein, gelation kinetics and mixing time, may find applications in food, pharmaceutical and personal care industries.

Crown Copyright © 2017 Published by Elsevier Ltd. All rights reserved.

1. Introduction

Lipophilic active molecules such as fat soluble vitamins, flavourings, fatty acids and essential oils pose challenges when incorporated into food, pharmaceuticals or other soft matter applications due to their partial or complete water insolubility. Besides oxidizing rapidly, most of these compounds are difficult to deliver in physiology and are generally only partially absorbed by the skin or via the gastrointestinal regime. Thus, their physiological activity is most often partly or fully lost before reaching the targeted physiological site (McClements, 2015). Consequently, there is a huge need to protect these lipophilic compounds from environmental degradation and tailor their release at particular biological sites (Sung, Xiao, Decker, & McClements, 2015). A wide range of technologies have been developed to encapsulate oil-soluble molecules, such as emulsions, emulsion gels, liposomes, micelles, nanoparticles, etc (McClements, 2011). Each of these has its own specific advantages and disadvantages in terms of degree of protection, delivery, cost, regulatory status, ease of use, biodegradability and biocompatibility (McClements & Li, 2010). Among these, emulsion microgel particles are vehicles that have not been explored as widely.

Emulsion microgel particles are a relatively new class of soft solids (Torres, Murray, & Sarkar, 2016). The particles have a similar structure to emulsion gels, although their physical characteristics and scales differ. In emulsion microgel particles, emulsion droplets are stabilised by an emulsifier and gelling agent that create a soft solid shell around several emulsion droplets, which are then incorporated into a continuous gel matrix (Ruffin, Schmit, Lafitte, Dollat, & Chambin, 2014; Zhang, Zhang, Decker, & McClements, 2015). This soft solid shell has been demonstrated to protect lipophilic compounds such as polyunsaturated fatty acids against oxidation (Augustin & Sanguansri, 2012; Beaulieu, Savoie, Paquin, & Subirade, 2002; Velikov & Pelan, 2008). Additionally, the microgel particle allows swelling or de-swelling as a function of pH, ionic strength, temperature and enzymatic conditions via tuning

* Corresponding author.
E-mail address: A.Sarkar@leeds.ac.uk (A. Sarkar).

http://dx.doi.org/10.1016/j.foodhyd.2017.04.029
0268-005X/Crown Copyright © 2017 Published by Elsevier Ltd. All rights reserved.

the size and/or physicochemical properties (Ballauff & Lu, 2007; Wei, Li, & Ngai, 2016). Hence, these particles have great potential for site-dependent release of lipophilic active compounds in a range of food, pharmaceutical, personal care and other soft material applications (Ching, Bansal, & Bhandari, 2016).

Whey protein isolate (WPI) is widely accepted for research and commercial applications and its versatility as an emulsifier and gelling agent is well recognized (Sarkar, Murray, Holmes, Ettelaie, Abdalla, & Yang, 2016). Under heat-treatment WPI undergoes conformational changes, exposing its hydrophobic and sulfhydryl groups allowing irreversible aggregation and gel formation under specific conditions of protein concentration, ionic strength and temperature (Roefs & Peppelman, 2001). On addition of calcium (Ca^{2+}) ions, heat treated WPI (HT-WPI) undergoes further aggregation via Ca^{2+} cross-linking of the negatively charged carboxylic groups on the WPI. Protein-Ca^{2+}-protein complexes are formed, reducing the negative charge on the protein (Bryant & McClements, 2000; Hongsprabash, Barbut, & Marangoni, 1999; Phan-Xuan et al., 2014).

Several technologies have been developed for the production of WPI stabilised emulsion microgel particles. For instance, multistep emulsion-templating allows the formation of emulsion particles via $O_1/W/O_2$ emulsions (Sung et al., 2015). The WPI aqueous phase of the $O_1/W/O_2$ emulsion is typically gelled through heat treatment, forming (O_1/W) WPI stabilised emulsion microgel particles suspended in an external oil phase (O_2). The oil phase is then washed away with the use of organic solvents. Although this generates microgel particles of controlled size: the multiple processing steps causes the technique to be laborious; heat gelation renders it ineffective for the use of heat-sensitive compounds; the use of organic solvents limits its application in certain medical drugs and food products where biocompatibility is a key issue (Beaulieu et al., 2002). An alternative multistep emulsion-templating method was designed by Egan, Jacquier, Rosenberg, and Rosenberg (2013). The aqueous WPI phase of the $O_1/W/O_2$ emulsion was gelled via a cold set technique. The external oil phase (O_2) was then washed away with surfactants rather than solvents. Although this technique allows the encapsulation of heat-sensitive compounds and does not require the use of solvents, the multiple processing required still causes this method to be time consuming and laborious plus excess surfactant may need to be removed. Extrusion technologies allowing cold external gelation of heat-treated WPI emulsion microgel particles have also been developed (Egan et al., 2013). In this case, the heat-treated WPI stabilised emulsion was dropped into an ionic bath, allowing the gelation of the continuous phase, which entrapped oil droplets into microgel particles. Although this external gelation method was successful it produced large particles, of 1–2 mm in diameter, limiting their application in food systems. Other processing methods produce emulsion microgel particles by emulsifying the oil phase with WPI or sodium caseinate and gelling the emulsion into microgel particles with alginate or pectin (Ruffin et al., 2014; Zhang, Zhang, & McClements, 2016). The use of several different biopolymers causes this technique to be not very cost effective. Also, thermodynamic incompatibility between the protein at the interface and the gelling biopolymer might result in uncontrolled release behaviour.

Thus, external gelation has considerable potential if it can be made facile, rapid and allow processing of clean emulsion microgel particles. Careful optimization of temperature, shear and WPI and Ca^{2+} concentration might also allow the tailoring of the size of emulsion microgel particles. The objective of this study was to design and characterize HT-WPI emulsion microgel particles of tailored sizes and examine the complex interplay between whey protein concentration, Ca^{2+} concentration ($[Ca^{2+}]$) and turbulent mixing conditions.

Commercial whey protein isolate was heat treated at different temperatures and times and its unfolding and aggregation rate were monitored using a fluorescent probe method and dynamic light scattering, respectively. The gelation kinetics of HT-WPI stabilised emulsions with different concentrations of Ca^{2+} ions were examined using small deformation shear rheology. These rheological experiments showed the effect of $[Ca^{2+}]$ on the type of gels formed. Finally, two different turbulent mixing processing techniques involving extrusion or T-mixing were tested, hypothesized to offer different mixing times. The emulsion microgel particles were examined using confocal laser scanning microscopy and scanning electron microscopy. Theoretical considerations, such as the Kolmogorov mixing time and the flux of Ca^{2+} ions to HT-WPI interfaces were used to explain the differences in particle size of emulsion microgel particles, obtained with both processing routes.

2. Materials and methods

2.1. Materials

Whey protein isolate (WPI) powder containing 96.3 wt% protein (Molecular mass: 18.4 kDa) was a kind gift from Fonterra Limited (Auckland, New Zealand). Sunflower oil was purchased from Morrisons supermarket (UK). Calcium chloride, 8–aniline–1–naphthalenesulfonic acid (ANS), sodium hydroxide, hydrochloric acid, sodium chloride, hexane anhydrous, 95% were purchased from Sigma-Aldrich (Gillingham, UK). Silicone oil 350 CST was purchased from VWR international S.A.S (Fontenay-sous-Bois, France). All solutions were prepared with Milli-Q water having ionic purity of 18.2 MΩ cm at 25 °C (Milli-Q apparatus, Millipore, Bedford, UK). Nile Red was purchased from Sigma-Aldrich (Steiheim, Germany). Dimethyl sulfoxide (DMSO) was purchased from Fluorochem (Hadfield, UK). All other chemicals were of analytical grade and purchased from Sigma-Aldrich unless otherwise specified.

2.2. Analysis of whey protein aggregation

2.2.1. ANS fluorescence method

Different concentrations of WPI (9.6 and 12 wt%) were diluted into Milli-Q water at pH 7.8–aniline–1–naphthalenesulfonic acid (ANS) (1 mg mL^{-1}) were dissolved into 0.1 M NaCl. Spectrofluorimetric measurements were made using a Fluorescence spectrophotometer (Perkin-Elmer, LS-3, Waltham, USA) following the method of Nyman and Apenten (1997). The ANS fluorescence measurements involved a fluorescence excitation wavelength of 280 nm and an emission wavelength of 470 nm. The final concentration of ANS was determined by fluorescent titration of 12 wt % WPI heated at 85 °C for 40 min. Increasing amounts of ANS stock solution were added to WPI samples (3 mL) in a quartz cuvette. Fluorescence emission intensity (ΔF) was recorded in relative fluorescence units (rfu). A graph of volume ANS (x-axis) vs ΔF provided a value for the maximum volume of ANS needed (150 μL) as the curve reached a plateau (result not shown). The concentration of ANS was determined using equation (1):

$$[ANS] = \frac{V_{ANS} \times C_{ANS}}{(V_{ANS} + V_{WPI})} \quad (1)$$

where, C_{ANS} is the concentration of ANS stock solution (3.2 mM), V_{WPI} is the volume of protein and V_{ANS} is the volume of ANS added to the protein solution. This final concentration of ANS (0.15 mM) was used for the subsequent measurement.

12 wt% and 9.6 wt% WPI solutions were heated at different temperatures (75, 80 or 85 °C) for different time periods (0, 8, 15,

Download English Version:

https://daneshyari.com/en/article/4983769

Download Persian Version:

https://daneshyari.com/article/4983769

Daneshyari.com