Food Hydrocolloids xxx (2013) 1–7

Contents lists available at ScienceDirect

Food Hydrocolloids

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

Studies on the viability of *Saccharomyces boulardii* within microcapsules in relation to the thermomechanical properties of whey protein

Diep Duongthingoc [a,b], Paul George [a], Elizabeth Gorczyca [a], Stefan Kasapis [a,*]

[a] School of Applied Sciences, RMIT University, City Campus, Melbourne, VIC 3001, Australia
[b] Faculty of Food Technology, Nong Lam University, Hochiminh, Vietnam

ARTICLE INFO

Article history:
Received 20 December 2012
Accepted 23 July 2013

Keywords:
Encapsulation
Spray drying
Whey protein isolate
Saccharomyces boulardii

ABSTRACT

Present work investigates the effectiveness of using whey protein isolate as an encapsulant for a biotherapeutic agent (*Saccharomyces boulardii*) under varying concentrations of protein and $CaCl_2$. Viability of *S. boulardii* within the matrix of whey protein is highly dependent on the physicochemical properties of the protein network, and its response to various environmental stress factors including processing temperature, moisture content and change in pH. Our interest is to optimise the spray drying conditions of whey protein isolate, through observations from differential scanning calorimetry and small deformation dynamic oscillation on shear, in relation to the denaturation and subsequent aggregation of the globular molecules. It is evident from this work that protein concentration is directly proportional to the strength of the network, but inversely proportional to the denaturation temperature. Increasing concentrations of calcium chloride have a direct influence on electrostatic repulsion between protein molecules thus creating a better protein structure. Development of a cohesive network was used as a basis of manipulating the viability of *S. boulardii* under given conditions of spray drying. Analysis on spray dried powders suggests that whey protein preparations at the isoelectric point (pH ~5) and with $CaCl_2$ additions of 50–100 mM act as an efficient encapsulant providing high viability of *S. boulardii*.

© 2013 Published by Elsevier Ltd.

1. Introduction

Whey protein isolate is a mixture of β-lactoglobulin (50–55% w/w), α-lactalbumin (20–25% w/w), bovine serum albumin and immunoglobulins (Abd El-Salam, El-Shibiny, & Salem, 2009; Qi & Onwulata, 2011). These globular proteins possess exposed hydrophilic and buried hydrophobic groups, which are capable of forming a wide range of cross-links when unfolded, and hence can produce water-soluble or water-insoluble matrices (de Witt, 1990). The structure and consistency of the whey protein networks vary depending on protein concentration, ionic strength, pH, temperature and degree of denaturation (Foegeding, Davis, Doucet, & McGuffey, 2002; de la Fuente, Singh, & Hemar, 2002).

In food systems, whey protein is commonly dispersed in an aqueous phase at ambient temperature, which upon thermal treatment transforms into a polymeric network (Kessler & Beyer, 1991; Tolkach & Kulozik, 2007). This occurs due to the unfolding of globular conformation thereby exposing the hydrophobic core along with sulfhydryl–disulfide interactions, under the stimulation effect of thermal energy (Bryant & McClements, 1998; Hudson, Daubert, & Foegeding, 2000; Lorenzen & Schrader, 2006). According to Kessler and Beyer (1991), once the undesirable effects of protein sedimentation is avoided, educated manipulation of the thermal transition in whey protein can be used to obtain desired textures for added value food products.

Microencapsulation is a technique of enclosing and protecting highly sensitive materials from different environmental stresses and finds wide application in food, drugs and many other products (Thies, 2005). Microencapsulation controls the release of the core elements depending on the physiochemical properties of the encapsulant (Fang & Bhandari, 2010). Several methods have been developed for microencapsulation and the most relevant are spray-drying, spray-coating, emulsion, extrusion, and gel-particle technologies (Champagne & Fustier, 2007). In the food industry, spray-drying is the most common microencapsulation method, since it is considered to be highly efficient and cost effective (Gharsallaoui, Roudaut, Chambin, Voilley, & Saurel, 2007). The technique has been widely applied in encapsulating probiotic bacteria where the

* Corresponding author. Tel.: +61 3 9925 5244; fax: +61 3 9925 5241.
E-mail address: stefan.kasapis@rmit.edu.au (S. Kasapis).

0268-005X/$ – see front matter © 2013 Published by Elsevier Ltd.
http://dx.doi.org/10.1016/j.foodhyd.2013.07.024

Please cite this article in press as: Duongthingoc, D., et al., Studies on the viability of *Saccharomyces boulardii* within microcapsules in relation to the thermomechanical properties of whey protein, *Food Hydrocolloids* (2013), http://dx.doi.org/10.1016/j.foodhyd.2013.07.024

microorganisms successfully surpass extreme thermal conditions (Boza, Barbin, & Scamparini, 2004; Riveros, Ferrer, & Bórquez, 2009). On application of whey protein as an encapsulant, it was found that *Bifidobacterium breve* exhibited a relatively high viability after being encapsulated through spray drying (Picot & Lacroix, 2004; Surawicz et al., 1989).

The yeast *Saccharomyces boulardii* is considered to be a biotherapeutic agent as it renders protection against antibiotic-induced diarrhoea, ulcerative colitis, and Crohn's disease (Castagliuolo, Riegler, Valenick, Lamont, & Pothoulakis, 1999; Guslandi, Mezzi, Sorghi, & Testoni, 2000). By conducting an *in-vitro* study, Fietto et al. (2004) and van der Aa Kühle, Skovgaard, and Jespersen (2005) have found that *S. boulardii* is relatively tolerant to applied temperature, acidic pH, and bile salt up to 0.3% (w/w) in preparations. In terms of the anti-diarrheal effect, this biotherapeutic agent is typically administered freeze-dried, in an unprotected state as part of a capsule (Candelli et al., 2003).

There is little knowledge regarding the microencapsulation of this probiotic using the energy efficient technique of spray drying, as compared to its costly counterpart of freeze drying. This understanding can serve as a means of improving the survival and hence the viability of this agent during gastrointestinal passage. The aim of this research, therefore, is to improve the delivery of *S. boulardii* through the use of microencapsulation technology employing a whey-protein based matrix. To this end, fundamental understanding of the rheological and thermal transition behaviour of the biopolymer matrix is required in order to develop an optimal capsule consistency.

2. Materials and methods

2.1. Materials

2.1.1. Whey protein isolate

The material used was a product from Fonterra, Waikato, New Zealand. According to the certificate of analysis from the supplier, it contains 91.3% protein, 0.7% fat, 3.5% moisture, 3.8% ash and 0.44% lactose. A solution of 10% (w/w) whey protein in distilled water gave a pH value of 6.3. The bulk density of the powder was reported to be 0.45 g/ml with a microbial plate count of 9900 cfu/g.

2.1.2. Saccharomyces boulardii

Lyophilised culture of *S. boulardii* was obtained from Biocodex (Gentilly, France). Yeast cells were cultured to a concentration of 10^8 cfu/ml at a late log phase in yeast nitrogen base (YNB) broth containing 1% D-glucose (BDH, Merck, UK), which was harvested as biomass for spray drying with whey protein isolate suspensions. All the microbiological analysis on *S. boulardii* was performed in triplicates.

2.1.3. Yeast malt extract agar (YMA)

Culture media YMA was obtained from Sigma Aldrich Private Ltd, New South Wales, Australia. According to the supplier, it contains peptic digest of animal tissue (0.5%), yeast extract (0.3%), malt extract (0.3%), dextrose (1%) and agar (2%).

2.2. Sample preparation and analysis

2.2.1. Protein solution

Whey protein dispersions were prepared by mixing the protein powder in distilled water at neutral pH and ambient temperature. Solutions were stirred for approximately 30 min using a magnetic stirrer to ensure proper dissolution. Dispersions were stored overnight at 4 °C to achieve thorough hydration. Series of samples were prepared at a wide pH range by the dropwise addition of 1 M NaOH or 1 M HCl. Samples with distinct ionic strength were obtained by the addition of calcium chloride at concentrations ranging from 0 to 200 mM at pH 7.

2.2.2. Rheological measurements

Aggregation and subsequent structure formation of whey proteins were observed using AR-G2, a controlled strain rheometer with magnetic thrust bearing technology (TA Instruments, New Castle, DE). Small amplitude oscillation on shear was the technique of choice to characterize the viscoelastic properties of the protein network as a function of temperature. Whey protein samples were analysed using a parallel plate geometry of 40 mm diameter. Samples were loaded onto a Peltier plate at 25 °C and the exposed edges were covered with silicone oil from BDH (50 cS) to prevent moisture loss. They were heated to 80 °C at a ramp rate of 1 °C/min at a constant angular frequency and strain amplitude of 1 rad/s and 0.1%, respectively. This was followed by an isothermal run at that temperature for 30 min and a cooling stage to 5 °C at the same scan rate.

2.2.3. Modulated differential scanning calorimetry

Thermal measurements on whey protein systems were carried out with a modulated differential scanning calorimeter (DSC Q2000) from TA instruments (New Castle, DE). To enable accurate measurements, a traceable indium standard with $\triangle H_f$ of 28.3 J/g and a sapphire standard were used to calibrate the heat flow signals and the heat capacity response, respectively. Hermetic aluminium T_{zero} pans for a sample size of 8 mg were employed. Weighed samples were equilibrated at 25 °C for 2 min and then heated to 100 °C at a scan rate of 5 °C/min. During the experiment, at every 40 s, a modulation rate of 0.53 °C was applied.

2.2.4. Isolation and culturing of the yeast cells

Commercially available freeze-dried yeast cells (0.1 g) were transferred into a sterile YNB broth (10 ml), which was incubated for 24 h at 37 ± 1 °C. One ml of sample from the above incubated broth was diluted in 9 ml of saline (0.85%) solution and plated onto YMA culture media, which was incubated for 48 h to identify the colonies of the yeast. According to Larone (2011), yeast colonies can be distinguished by their whitish cream colour and circular shape, and upon magnification each yeast cell possesses an elliptical morphology. Yeast colonies obtained on YMA media were confirmed by a microscopic investigation for cell morphology and purity.

Identified colonies of yeast were then transferred to YNB broth (10 ml) and mixed thoroughly using a vortex mixer. Following mixing, an aliquot of 1 ml was inoculated into the same broth line (YNB, 99 ml), and was incubated at 37 ± 1 °C for 48 h. This served as the seed culture for the derivation of growth curves and subsequent microencapsulation studies.

2.2.5. Microencapsulation of yeast with spray drying

Microcapsules were prepared by spray drying a whey protein suspension containing the culture of *S. boulardii*. Yeast cell preparations (100 ml) in their late log phase with an average cell count of 3×10^8 cfu/g were centrifuged at 6000 rpm at 4 °C for 10 min. Pellets obtained after centrifugation were washed twice using a sterile saline (0.85%) solution before resuspending into a whey protein dispersion (10%, w/w) at the required pH and concentration of $CaCl_2$.

Spray drying of the above sample was performed on LabPlant Spray Dryer SD-Basic (Labplant UK Ltd), which was operated in accordance with the instructions of the manufacturer. To ensure an even distribution of microorganisms throughout this process, the feed solution was continuously stirred using a magnetic stirrer.

Please cite this article in press as: Duongthingoc, D., et al., Studies on the viability of *Saccharomyces boulardii* within microcapsules in relation to the thermomechanical properties of whey protein, *Food Hydrocolloids* (2013), http://dx.doi.org/10.1016/j.foodhyd.2013.07.024

Download English Version:

https://daneshyari.com/en/article/603861

Download Persian Version:

https://daneshyari.com/article/603861

Daneshyari.com