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Microencapsulation of astaxanthin with blends of milk protein and fiber by spray drying

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ABSTRACT

This study investigated the encapsulation of astaxanthin using spray drying method to enhance its stability and application in food systems. Blends of milk protein (either whey protein isolate, WPI or sodium caseinate, SC) and carbohydrate (soluble corn fiber, SCF 70) were applied as the wall materials. The results demonstrated that spray drying could be applied to transform the stable astaxanthin emulsions into powders with reasonably good properties, including water activity, surface morphology and oxidative stability. The reconstituted emulsions also showed good stability similar to the parent emulsions. The microencapsulation efficiency was high (~95%) for both wall systems under investigation, indicating the suitability of these wall matrices for encapsulating the hydrophobic astaxanthin.

1. Introduction

Astaxanthin is a type of carotenoids found in marine animals such as salmon, trout, shrimp and lobster, giving them a characteristic red pigmentation. It can be synthesised only by some microorganisms, especially the green alga *Haematococcus pluvialis*, which is able to accumulate up to 3% on a dry weight basis (Olaizola, 2009).

Astaxanthin has been shown to exhibit a stronger antioxidant activity than vitamin E and β-carotene (Shimidzu et al., 1996; Naguib, 2000), contributed by its strong quenching activity against singlet oxygen and active scavenging of reactive oxygen species (Shimidzu et al., 1996; Naguib, 2000; Guerin et al., 2003). It has also been shown to protect membrane phospholipids and other lipids against peroxidation (Palozza and Krinsky, 1992; Barros et al., 2001). Research have demonstrated positive effect between astaxanthin and a number of health conditions, such as cancer, age-related macular degeneration, inflammation and cardiovascular oxidative stress (Tanaka et al., 1995; Pashkow et al., 2008; Willcox et al., 2008), as well as for general enhancement of immune responses (Jyonouchi et al., 1995; Bennedsen et al., 2000). In recent years, this bioactive compound has gained popularity as a nutraceutical and pharmaceutical ingredient. However, astaxanthin has poor water solubility; this limits its application in the food industry. In addition, the presence of double bonds in its molecular structure has made astaxanthin susceptible to oxidants, light and heat, resulting in poor quality products with reduced health promoting properties. Therefore, the free astaxanthin requires some forms of protection from chemical damage or modification, before it can be applied in functional foods.

Microencapsulation technique has wide applications in food processing, aiming to protect food ingredients against deterioration. It involves a formation of wall system to enclose droplets or particles of the core material. This technique has been applied to improve the solubility and stability of astaxanthin using chitosan matrix (Higuera-Ciapara et al., 2004; Kittikaiwan et al., 2007), β-cyclodextrin and hydroxypropyl-β-cyclodextrin (Chen et al., 2007), emulsions (Clark et al., 2000; Ribeiro et al., 2005; Wackerbarth et al., 2009) and liposomes (Peng et al., 2010). However, in most of these studies, astaxanthin was dissolved in organic solvents, for example, in dichloromethane and chloroform, which are toxic to humans. The choice of encapsulants also limits the applications of astaxanthin in food products. For example, despite chitosan is a potential encapsulant for astaxanthin, consumption of chitosan may interfere with lipid digestion and adsorption (Mun et al., 2007), which is needed to facilitate digestion of carotenoids. In addition, food grade encapsulant is required for food applications; this makes the non-food grade hydroxypropyl-β-cyclodextrin an unsuitable choice despite its good encapsulation capacity. Synthetic emulsifiers such as Tween 20 and Tween 80 as applied in some studies (Ribeiro and Cruz, 2005; Ribeiro et al., 2005; Anarjan et al., 2010) are less favoured by consumers than natural emulsifiers. Furthermore, some of these studies may not be suitable for large scale production.

To fill the research gap, we aimed to study the microencapsulation of astaxanthin using spray drying and to evaluate the product stability. Spray drying is a relatively well-established technology

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with good equipment readily available to produce high quality powders in large scale production, which are suitable to produce functional ingredients for food industry since cost is frequently a major consideration. Spray drying has been reported as a suitable technique for microencapsulation of ingredients in food industry including flavor, colorant, antioxidant, essential oils, etc. Some recent literature include the encapsulation of açai (*Euterpe oleracea* Mart.) juice (Tonon et al., 2009; Tonon et al., 2010), jaboticaba (*Myrciaria jaboticaba*) peel extracts (Silva et al., 2013), and co-encapsulation of fish oil, phytosterol ester and limonene (Chen et al. 2013).

For encapsulating lipophilic core materials, the core materials is normally homogenized in a solution containing wall material to form a stable emulsion and then fed into a spray dryer where it is converted into dry powders. Therefore, wall materials with good emulsifying properties is desired. We propose the use of whey protein isolate (WPI) or sodium caseinate (SC) with soluble corn fiber (SCF70) as wall systems in current study. These are natural emulsifiers accepted by consumers; they are also cost effective, readily available and exhibit good nutritional values. The use of SCF70, a soluble corn fiber which is also a prebiotic, as wall material for encapsulation of bioactive, is a relatively new attempt (Chen, 2012).

2. Materials and methods

2.1. Materials

The materials were obtained from the following sources: astaxanthin standard from United Bioresearch, NSW, Australia; Cynatech Bioastin astaxanthin from Hawkins Watts Limited, New Zealand (NZ); Pam's sunflower oil from Countdown supermarket, Auckland, NZ; whey protein isolate (WPI) from Reactiv, Auckland, NZ; sodium caseinate (SC) from Fonterra, NZ, and Promintor™ soluble corn fiber 70 (SCF70) of DE 20, a gift from Tate & Lyle, Ltd., NZ. MilliQ water was used for preparation of emulsion and in all analyses. All chemicals used were of analytical grade.

2.2. Preparation of astaxanthin emulsions

Wall material, consisting of SCF 70 with WPI or SC as emulsifiers (in a mass ratio of 1:1), was dissolved in 700 g distilled water. The solution was allowed to hydrate for at least 1 h before emulsion preparation and held in a 50 °C water bath to ensure a full dissolution of the emulsifiers, followed by cooling down to room temperature. Core material, including sunflower oil and Cynatech Bioastin astaxanthin, was added to the solution in a mass ratio to the wall material of 1:4. The total concentration of the dissolved solids (wall and core materials) was 30% of the weight of final emulsion. The pH of pre-emulsions was adjusted to around 6.7 using 1 M NaOH or 1 M HCl as required. Coarse emulsions were prepared using an Ultraturrax Janke & Kunkel T25 homogeniser (Germany) at 13,500 rpm for 2 min, and then passed through a two-stage homogeniser (APV, Denmark) at 80 and 800 bars.

The amount of Bioastin used in the emulsion was 2 wt%. Since Bioastin contains 5 wt% astaxanthin, the amount of astaxanthin is equal to 0.1% of the 30% total solid content. Assuming the core material was evenly and completely microencapsulated, the amount of encapsulated astaxanthin was approximately 0.33 wt% of the spray dried powders.

2.3. Spray drying of astaxanthin emulsions

The emulsions were converted into powders using a centrifugal spray drier (Sunshine model GZ-5, Jiangsu Wuxi Yangguang Co., Jiangsu, China). The feed and hot air were entering the spray drier in a co-current manner. The drying capacity was 5 kg water/h and the diameter of the drying chamber was 0.85 m. Compressed air was used as the drying medium. The spray drier was equipped with an atomizing spray nozzle with a rotational speed of 25,000 n/min. The effects of air inlet temperature was investigated at 160 °C, 170 °C and 180 °C while the outlet temperature was at 70 °C and 80 °C to find the optimum drying temperatures. The pump flow rate was controlled at 2 L/h. The dried powder was collected and stored in air tight desiccators at 4 °C for further analysis.

2.4. Droplet size, size distribution and zeta potential of emulsions

The average droplet size, size distribution and zeta potential of the emulsions (both the parent emulsions before drying and the reconstituted emulsions after drying) were determined using a Nano Zetasizer (Malvern Instruments Ltd., Worcestershire, UK). The droplet size and size distribution were measured at a fixed angle of 173° at 25 °C. The emulsions were diluted with Milli-Q water to avoid multiple scattering effects during measurement. The polydispersity index (PDI) is used to describe the droplet size distribution with the value reported to be between 0 and 1. The final particle diameter and PDI were calculated from the average of three measurements. For zeta potential measurement, the temperature was set at 25 °C, while the pH of the emulsions during the measurement was around 6.7. The final ζ-potential was averaged from three measurements with each measurement set for 50 runs.

2.5. Determination of microencapsulation efficiency (ME)

For determination of ME, it is assumed that all the initial core material was retained in the powder. The surface oil content of encapsulated powders was determined using extraction with petroleum ether (ECP-AR, New Zealand) according to the method of Jafari et al. (2008) with some modifications. One gram of spray dried powder was accurately weighed and added to 15 mL of solvent in a volumetric flask and the suspension was mixed with a vertax gently at room temperature for 10 min. The powder and the solvent were separated by filtration through filter paper (No. 41, Whatman, Maidstone, UK). The powder residue was washed with 2 × 5 mL petroleum ether and the filtrate solution containing the extracted oil was transferred to a round-bottomed flask, which was subsequently placed in a rotary evaporator (Buchi Waterbath B-480, Germany) to evaporate the organic solvent under vacuum condition. After evaporation, the flask was dried in an oven at 63 °C until constant weight was achieved (about 1 h). The extracted oil value was then calculated based on the difference between the weight of the initial clean flask and that containing the extracted oil residue. The microencapsulation efficiency (ME) was obtained from the following equation.

$$\%\mathrm{ME} = (\text{Total core content} - \text{Extracted surface oil})/(\text{Total core content}) \times 100 \quad (1)$$

The experiment was conducted in duplicate.

2.6. Water activity of powders (a_w)

Water activity of the powders was determined using a water activity analyzer (HydroLab 3, Rotronic, USA) at the temperature of 25 °C. Each measurement was repeated twice, and average a_w was reported.