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Emulsifying properties and structure changes of spray and freeze-dried peanut protein isolate

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ABSTRACT

The emulsifying properties of peanut protein isolate (PPI) prepared by spray- and freeze-drying methods were investigated together with the change in protein structure due to drying. Oil binding, water holding capacities and solubility of freeze-dried PPI were significantly higher ($p < 0.05$) than those of the spray-dried one. The spray-dried PPI had higher emulsifying activity index (EAI), whereas the freeze-dried PPI had higher emulsion stability index (ESI).

The freeze-dried PPI had significantly higher surface hydrophobicity, disulfide bonds and β-sheets than the spray-dried one ($p < 0.05$). While the latter contained more hydrogen bonds than the former ($p < 0.05$), as shown by the Fourier transform infrared spectroscopy, which suggested that the spray-dried PPI had relatively higher unfolded or flexible structure than the freeze-dried PPI.

Folded and wrinkled morphology of spray-dried PPI but plate-shaped structure in the freeze-dried PPI suggested that droplet shrinkage and solute concentration led to the distinct morphology, respectively. These two different drying processes greatly brought about different structure and properties thereof. Thus, the freeze-dried PPI produced more stable emulsions (higher ESI), while the spray-dried PPI occupied the oil water interface faster (higher EAI).

1. Introduction

Proteins are natural amphiphilic molecules with interfacial activity and colloid-stabilizing characteristics. Hence, proteins are increasingly used as emulsifiers. Proteins preferentially adsorb to the oil–water interface and form a viscoelastic film, which provides physical stability to the emulsion during subsequent processing and storage (Dickinson, 2001; Joshi et al., 2012). Vegetable proteins are being increasingly used in various food applications, owing to their acceptable emulsifying, fat and water absorbing, texture modifying and whipping properties (Joshi et al., 2012; Suresh Kumar et al., 2014). Peanut protein does not contain cholesterol and its nutritional value is similar to that of animal proteins. Peanut protein carries the characteristic aroma of peanut (He et al., 2014). Most of the currently available peanut protein is obtained from defatted peanut flour (DPF), which is protein rich (47–55%) and is an under-utilized byproduct of the peanut oil extraction industry (Feng et al., 2014). Due to its desirable functional properties, high nutritional value and low cost compared with other proteins, peanut protein is finding wider application in food industry.

Emulsifying properties of peanut protein has been studied and reported to considerable detail. Most of these studies have focused on quantifying and explaining the effects of different methods on the physicochemical characteristics of peanut protein (Fekria et al., 2012; Hu et al., 2014). The effects of physical methods such as ultrasound and high pressure or biological modification such as hydrolysis and transglutaminase crosslinking on the emulsifying properties have also been studied (He et al., 2014; Li et al., 2014). Only a few studies have noted that the extraction process also influences the functional properties of peanut proteins. For example, it has been suggested that the emulsifying activity of peanut protein might be associated with the tertiary structure of the protein and any alteration of this structure is expected to affect the emulsifying property (Kain and Chen, 2010; Liu et al., 2011).

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Nevertheless, there is not enough information in the literature to explain and relate the relationship between emulsifying properties and change in structure of peanut proteins.

Drying process exerts a very high degree of stress on the structure of extracted proteins and affects their functional properties (Caparino et al., 2012; Yu et al., 2007). Currently, spray-drying and freeze-drying are the two most commonly used methods to convert proteins into powder form. The thermal and dehydration related quality degradation of protein is relatively low when these two drying methods are used (Cohen and Yang, 1995). However, both spray and freeze drying processes alter the structure to some extent and bring about some changes in morphology (Haque et al., 2013; Shaviklo et al., 2010). Furthermore, these two drying methods exert a different degree of stress to the protein structure and, thus, differently affect the resultant functional properties. Studies have been carried out to compare the effect of these two drying methods on physicochemical and functional properties of lentil (Joshi et al., 2011), rice dreg (Zhao et al., 2013) and soy proteins (Hu et al., 2010; Liao et al., 2013). However, the effects of these drying methods on the emulsifying properties of peanut proteins have not been studied. Given that different proteins have different sensitivity to drying-related stresses, it is of practical importance to investigate the effect of spray and freeze drying methods on the structure of peanut proteins and relate these changes to the surface morphology of the dried product, hydrophobicity and emulsifying properties.

Hence, the effects of spray and freeze drying on the structural configuration (α-helix, β-sheet, hydrogen bonds) of peanut protein isolate (PPI) were investigated in this study. The effect of these two drying methods on the surface hydrophobicity, emulsifying properties and surface morphology of the dried PPI powders were also investigated in considerable detail. Thus, this study provides insight into how best to use spray and freeze drying technologies to produce PPI powders with best possible emulsifying properties.

2. Materials and methods

2.1. Materials

Commercially produced defatted peanut flour (DPF) was obtained from Gaotang Lanshan Co., Ltd. (Shandong Province, China). The process of producing DPF consisted of cleaning, drying, cracking, dehulling and flaking at low temperature (60 °C). The oil was extracted by using a blend of butane and propane. The oil-extracted flour was further dried by cross-ventilation and was finely pulverized by using an ultra-micro pulverizer (LHM, ZhenYuan Powder Engineering Equipment Co., China). The oil content of DPF was less than 2.0% (w/w).

2.2. Preparation of peanut protein isolate (PPI)

DPF (90.0 g) was dispersed in 900 mL of distilled water (10% w/v) and the pH of the dispersion was adjusted to 8.0 with 1.0 N NaOH. The dispersion was stirred using a magnetic stirrer for 1 h at ambient temperature (25 ± 2 °C). The precipitated fraction of DPF was separated from the protein extract using a centrifuge (3K15, Sigma Instruments, Germany) at 10,000× *g* for 15 min. The supernatant was collected and was acidified with 1.0 N HCl to pH 4.5. The protein precipitate was then collected by centrifuging at 10,000× *g* for 10 min. Subsequently, the PPI was resuspended in distilled water while maintaining approximately 10% (w/v) total solid. The pH of this suspension was adjusted to 7.0. A sample of this resuspended protein solution was characterized as undried PPI, and the remaining solution was dried using spray- and freeze-drying methods.

2.3. Preparation of spray-dried and freeze-dried PPI

PPI solution (approximately 10%, w/v) was dried by using spray- and freeze-drying methods. In the freeze-drying operation, the protein solution was first put into a lyophilizer (LGJ-25, Sihuan Instruments, China) and the temperature of the sample was lowered to −40 °C. The freeze-drying process was stopped when the sample temperature reached 20 °C. Freeze-dried PPI was pulverized using a grinder (FW-100, Taisite Instruments, China) and sieved through a No. 40 mesh.

Spray-drying of PPI solution was carried out using a benchtop spray dryer (Büchi B-290, Büchi Labortechnik AGt, Switzerland). The outlet and inlet temperatures were maintained at 85 °C and 180 °C, respectively, during the spray-drying process. The flow rate of the feed solution was maintained at 6.5 mL/min by keeping the pump capacity at 16%.

2.4. Determination of protein content

The PPI sample (approximately 0.25–0.50 g for solid, 2.0–5.0 mL for liquid) was mixed with 6.0 g potassium sulfate and copper sulfate contact catalyst, and then 10 mL of sulfuric acid was added to the mixture. This mixture was digested in a digestion stove (SKD-20S2, Peiou Instruments, Shanghai, China). Once the digestion was completed, the protein content was determined using a fully automated Kjeldahl apparatus (Kjeltec 2300, Foss, Denmark).

2.5. Determination of water-holding and oil-binding capacities

The water holding capacity (WHC) of samples was determined as described by Beuchat (1977). The sample (approximately 1.0 g, W_0) was placed in a centrifugal tube and was weighed together with the centrifuge tube (W_1). Then, 10 mL distilled water was added to the PPI sample and was mixed using a vortex mixer. After the mixture samples were thoroughly wetted, they were allowed to stand at room temperature for 30 min and then centrifuged at 3000× *g* for 20 min. The supernatant was decanted and the centrifuge tube containing the sediment was weighed (W_2). The WHC (gram of water per gram of protein) was calculated as given by equation (1).

$$\mathrm{WHC} = \frac{W_2 - W_1}{W_0} \tag{1}$$

The oil binding capacity (OBC) was determined following the method of Yu et al. (2007). The sample (approximately 1.0 g, W_0) was placed in a centrifugal tube and was weighed together with the tube (W_1). Then, 5 mL soybean oil (Yihaijiali Co. Ltd., China) was added to the PPI sample and was mixed for 5 min using a vortex mixer. These PPI-oil samples were allowed to stand for 30 min at an ambient temperature. The fully mixed PPI-oil samples were then centrifuged at 3000× *g* for 20 min. The supernatant of soybean oil was decanted and the centrifuge tube containing the sediment was weighed (W_2). The OBC (gram of oil per gram of protein) was calculated using equation (2).

$$\mathrm{OBC} = \frac{W_2 - W_1}{W_0} \tag{2}$$

2.6. Solubility

PPI was dispersed in water (1%, w/v) by stirring for 30 min using a magnetic stirrer. Then, the pH was adjusted to 7.0 and the stirring was continued for 60 min. The pH was readjusted if necessary.