Industrial Crops & Products 112 (2018) 402–411

Contents lists available at ScienceDirect

Industrial Crops & Products

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

Encapsulation templated approach to valorization of cocoa husk, poppy and hemp macrostructural and bioactive constituents

Ana Belščak-Cvitanović[a,*], Aleksandra Vojvodić[a], Arijana Bušić[a], Julia Keppler[b], Anja Steffen-Heins[b], Draženka Komes[a]

[a] Department of Food Engineering, Faculty of Food Technology and Biotechnology, University of Zagreb, Croatia
[b] Department of Food Technology, Institute of Human Nutrition and Food Science, Kiel University, Germany

ARTICLE INFO

Keywords:
Cocoa bean husk
Encapsulation
Hemp
Polyphenols
Poppy
Proteins
Recovery

ABSTRACT

In this study valorization of cocoa bean husk (*Theobroma cacao* L.), common poppy (*Papaver somniferum* L.) and industrial hemp (*Cannabis sativa* L.) as frequent waste and under-utilised raw materials was proposed by a recovery process of their macrostructural and bioactive compounds and their utilization in the formulation of alginate-based hydrogel particulate delivery systems. Compositional analysis of the raw materials was performed and a simple routine for simultaneous recovery of both macrostructural and bioactive fractions developed by sequential,solvent extraction and alkaline extraction-isoelectric precipitation at two different extraction temperatures (20 °C and 70 °C). An innovative approach to utilization of both recovered macrostructural and bioactive isolates in combination with alginate was examined, aiming to formulate particulate delivery systems constituted from the macrostructural fractions as the carrier adjuncts and encapsulating the produced bioactive extracts of used raw materials. Macrostructural isolates obtained at higher extraction temperature (70 °C) were characterized by better functional properties, i.e. higher recovery yields (< 12.53%), lower WHC, higher FAC, higher foaming capacities (< 280%) and stabilities (< 200%), confirming those extraction parameters as the preferred ones for obtaining macroconstituent isolates for encapsulation purposes. The encapsulation-case study revealed potential protein-polysaccharide interactions which did not affect markedly the physico-chemical properties, but improved the polyphenols retention in the formulated particulate delivery matrix.

1. Introduction

Currently the valorization of food waste materials and secondary agro-industrial by-products is in the focus of numerous research studies, aiming on recovery and exploitation of those materials for biotechnological, food, cosmetic or pharmaceutical purposes (Galanakis, 2012; Galanakis et al., 2018). The biorefinery concept of plant raw materials, food waste materials and by-products represents a challenging issue that comprises a multifunctional recovery of different groups of constituents, and aims at increased yields of each of those groups and their further diverse applications (Lin et al., 2013). In this context, secondary food waste materials and by-products are used often for recovery of macromolecular constituents (especially carbohydrates and polysaccharides) and bioactive ingredients (such as pigments and antioxidants) (e.g. Wong et al., 2015). According to Galanakis (2015) the 5-step universal recovery strategy includes the macroscopic pre-treatment, macro- and micro-molecules separation, extraction/isolation, purification and finally formulation. For this purpose, apart from the conventional processing techniques, emerging technologies such as low temperature plasma treatment, high-hydrostatic pressure, ultrasound-assisted extraction (Pan et al., 2012), pulsed electric field (Parniakov et al., 2014), or nanotechnology have been more often extensively reported (Galanakis, 2013).

Among the macroconstituents comprising the chemical composition of majority of plant derived food waste materials, the recovery of proteins has so far not been extensively reported. However, due to the increasing trend of protein supplementation in the modern diet, the interest of scientific community during recent years has been focused on protein isolation from diverse plant sources. Usually protein and peptide compounds have been produced from animal (meat, fish or milk derived) by-products by diverse filtration or membrane technologies (Saidi et al., 2014; Suwal et al., 2014; Alfaro et al., 2014; Galanakis et al., 2014). Protein isolates obtained from plant sources represent a growing ingredient market, in part due to consumer preferences and their relatively low cost compared to animal-derived proteins (Stone et al., 2015). So far for protein recovery, mostly pulses

* Corresponding author at: Department of Food Engineering, Faculty of Food Technology and Biotechnology, University of Zagreb, Pierrotijeva 6, 10 000 Zagreb, Croatia.
E-mail address: abelscak@pbf.hr (A. Belščak-Cvitanović).

https://doi.org/10.1016/j.indcrop.2017.12.020
Received 9 August 2017; Received in revised form 30 November 2017; Accepted 9 December 2017

and legumes (Kiosseoglou and Paraskevopoulou, 2011; Can Karaca et al., 2011; Guleria et al., 2009), soy (Bian et al., 2003; Deak and Johnson, 2007), pea (Stone et al., 2015) and hemp (Malomo and Aluko, 2015) were used as the raw materials, while the recovered protein fractions were rarely used in further reported applications. Due to their pronounced functional properties, proteins as biopolymers are frequently used as carrier materials in encapsulation purposes. Either as single encapsulants or in combination with other biopolymers, proteins can serve as a viable alternative to synthetic polymeric materials, which apart from providing a physical matrix and barrier to encapsulated substances can also contribute to the nutritive and added value of the delivery systems they constitute (Nesterenko et al., 2013). Macrostructural and bioactive compounds recovered from diverse plant materials and by-products have already been evaluated as potential constituents of drug delivery systems produced by encapsulation (de Almeida et al., 2015; Reátegui et al., 2017; de Barros Fernandes et al., 2016; Zhang et al., 2017). However, there is a lack of studies on the application of plant-derived proteins recovered from these materials, especially with regard to formulation of encapsulated delivery systems.

Cocoa bean husk is today one of the most abundant secondary agro-industrial by-products, remaining after processing of cocoa beans (*Theobroma cacao* L.) in confectionery industry as one of the largest food industries. So far, this agro-industrial waste has been used for mushrooms cultivation and animal feed, while its use as a source of dietary fiber that might be used to supplement fiber intake has been proposed recently (Collar et al., 2009). Apart from the food waste materials and by-products, there is a big diversity of geographically traditional raw materials that have limited applications and whose nutritive composition is not yet well explored. Among such materials, opium poppy (*Papaver somniferum* L.) exhibits a big potential due to its very specific proximate composition and characteristic aroma, however apart in households it is still industrialy underutilised. On the other side, industrial hemp (*Cannabis sativa* L.) belongs to one of the most widely used and exploited raw materials, primarily for its high fat and protein contents, among which the protein products are already widely produced and available commercially (Teh and Birch, 2013). A joint characteristic of all three stated raw materials is primarily a high fat content and abundance in specific nutrient constituents, such as carbohydrates and proteins. Although the proteins recovered from hemp have an extensive application, it is much less known that cocoa husk and poppy are also abundant in these highly-valuable nutrients. Additionally, all of those plant substrates contain a natively present amount of bioactive compounds, whose composition needs further research and exploitation.

In the present study, a valorization approach of cocoa bean husk, poppy and industrial hemp is proposed by recovery of highly-valuable macroconstituents (especially proteins) and bioactive compounds from the three materials, aimed at their more prominent and efficient utilization in the future. For that purpose a simple sequential recovery process of separate macroconstituents (fat, extractable bioactive compounds, proteins) will be examined and the isolated protein and bioactive fractions utilised as carrier adjuncts and active ingredients in encapsulation purposes. Apart from providing a general macroconstituent composition of the three raw materials, the extraction efficiency of proteins by two extraction methods relying on alkaline extraction-isoelectric precipitation will be evaluated and functional properties of the isolated protein fractions characterized. The obtained protein fractions (isolates) will be used as carrier/encapsulant adjuncts for improving the encapsulation efficiency of bioactive compounds, primarily polyphenols in ionically-crosslinked alginate beads.

2. Materials and methods

2.1. Chemicals and materials

All chemicals used for the experimental procedures were of analytical or HPLC grade. Sodium alginate (medium viscosity) was purchased from Sigma-Aldrich (USA). Cocoa bean husk (CBH) was donated by a national confectionery industry (Kandit d.o.o., Croatia) in dried form, while dried poppy seeds (Franck, d.o.o., Croatia) and industrial hemp powder (Solae LLC, Switzerland) were purchased at a local market.

2.2. Determination of proximate composition of samples

All samples were dried in a laboratory oven at 50 °C to unify the moisture content, ground and homogenized by using a ball mill and sieved through 0.45 mm pore-size sieve. The proximate composition of all three raw materials was evaluated by using standard AOAC methods. Dry matter and crude mineral content were determined gravimetrically according to the modified AOAC 930.15 and AOAC 942.05 methods, respectively (AOAC, 1990). Crude fat content was determined gravimetrically after continuous extraction with petroleum ether in Soxhlet apparatus, using modified AOAC 920.39 protocol (AOAC, 1990). Crude protein content of secondary raw materials was determined according to the AOAC 976.05 method (AOAC, 1990) by using semi-automatic Kjeldhal protocol and a standard factor of 6.25 for calculation. All analyses were performed in duplicate and the results were expressed as average value on dry matter basis (% dmb). Crude carbohydrate content was determined indirectly by subtracting the contents of the previously determined major constituents (fat, protein and mineral content) from 100% dmb. The results were expressed as average percentage of dry matter (% dmb).

2.3. Determination of extractable compounds (preparation of bioactive extracts)

The samples previously deffatted by Soxhlet extraction were extracted repeatedly (3-fold extraction) with a 70% aqueous ethanol solution (v/v) for 15 min at ambient temperature. For the extractions 1:10 sample-solvent (w/v) ratio was used and the extractions performed on a magnetic stirrer operating at 500 rpm. After each extraction, the mixture was vacuum filtered through Whatman no. 4 filter paper, and the crude residue reused for subsequent extraction. Following the final, third extraction, the supernatants were combined, and ethanol removed via vacuum evaporation. The obtained aqueous extract was filled up to a defined volume and dry matter of the obtained extracts determined using the previously described AOAC method, in a 10 mL aliquot (AOAC, 1990).

2.4. Preparation of protein isolates

For the recovery of protein fractions, the alkali extraction–isoelectric precipitation (AE-IP) was employed as the protein isolation technique, according to the procedure reported by Stone et al. (2015). Briefly, to 50 g of all three raw materials which were previously defatted and extracted by 70% ethanol 750 mL of water were added and the mixture adjusted to pH 9.50 with 1.0 M NaOH, following by stirring at 500 rpm for 1 h at 20 °C or 70 °C. After the extraction, the mixture was centrifuged at 4500 × *g* (Sorvall ST 8 centrifuge, Thermo Fisher Scientific, Asheville, USA) for 20 min at 4 °C and the supernatant collected. For precipitation of proteins, the pH was adjusted to pH 4.50 using 1.0 N HCl and kept at 4 °C for 1 h followed by centrifugation at 4500 × *g* for 20 min at 4 °C. The crude pellet was collected, redissolved in distilled water and freeze dried (Labconco Corp., Kansas City, MO, USA). The obtained freeze dried isolates were pulverized in a mortar and pestle and kept well closed at refrigerated conditions until the analyses or further use.

2.5. Chemical and functional characterization of protein isolates

The yield of recovered protein fractions (%) was determined