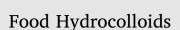
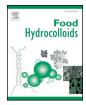
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The emulsifying performance of mildly derived mixtures from sunflower seeds



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ABSTRACT

Sustainability driven production of food ingredients is in the center of discussion the past years, with plants being a promising source, since they are widely available and have smaller environmental impact compared to animals. However, plant material consists of a sturdy configuration comprising many components, like proteins, which cannot be readily liberated. Thus, downstream processing of plants often involves intensive physico-chemical and thermal processing, which might be accompanied by alteration of protein properties, like emul-sification ability. Here, the aim was to investigate the emulsification ability of the native mixtures derived from sunflower seeds, obtained via simple separation steps and link their properties with their molecular composition. The investigated molecular mixtures were the cold-pressed sunflower cake, a protein-based and a fibre-based mixture. It was demonstrated that the residual oil in both the Sf cake and the protein-based mixture was present in the form of naturally emulsified oil droplets, so-called oil bodies. Oil bodies did not have a notable impact on the interfacial activity of the samples in contrast with the destabilization effect of polysaccharides. Despite their complex composition all mixtures could efficiently stabilize oil/water interfaces, showing similar properties compared to isolated proteins. This is an intriguing bottom line regarding the necessity for using pure emulsifiers. The findings prove that molecular mixtures which contain even minor amounts of proteins, can be used as ingredients for efficient emulsion stabilization.

1. Introduction

Global demand for food escalates and the need for sustainability driven production of food ingredients intensifies. Various aspects of food production can be fine-tuned, from the selection of the raw material source and the holistic valorisation of the material (Boye & Arcand, 2013). Animal sources of ingredients are extensively exploited, however they are considered less environmental friendly compared to ingredients derived from plants (Aiking, 2011). However, plant-based biomass requires different separation designs than those applied to produce animal ingredients, where for example, proteins from milk can be readily obtained in relative pure form (Mulvihill & Ennis, 2003). Plant materials have significantly different biomass configuration which does not breakdown readily, due to the sturdy cell walls, perplexing the separation of ingredients within the matrix (Campbell et al., 2011). In addition, the presence of a plethora of compounds, from polysaccharide to phenols, often results in complexation with proteins hindering their separation (Boland et al., 2013). For these reasons downstream processing of plant proteins often involves intensive physicochemical and thermal steps which are often accompanied by alteration of their functional properties, i.e. emulsification ability (Moure, Sineiro, Domínguez, & Parajó, 2006). Taking all these steps into account, separation processes of plant-based ingredients are not by default more sustainable than the production of animal ingredients (Apaiah, Linnemann, & van der Kooi, 2006). The design of less intensive ingredient separation steps, eventually leads to the reconsideration of the biorefinery philosophy. There is a need for less intensive and efficient practices that use minimum amounts of energy and chemicals (van der Goot et al., 2016).

As an example, by-products from sunflower oil industry (sunflower seed cake) constitute a worthwhile source of ingredients like proteins, fibres and phenols (Moure et al., 2006). Sunflower press cake is ranking fourth with a worldwide production of about 19 million metric tons in 2017 (USDA, 2018). The protein content is 25–50 wt% (Pickardt, Eisner, Kammerer, & Carle, 2015), fibres are ranging between 29 and 52 wt% and phenols reach up to 3 wt% (Weisz, Kammerer, & Carle, 2009), with chlorogenic acid being the dominant phenol (2.8 wt%). However, obtaining these components, is not trivial due to irreversible

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bonding with surrounding molecules (González-Pérez & Vereijken, 2007).

A solution to that problem could be a shift from designs that do not aim towards pure components but to multicomponent mixtures (mixtures of oil, proteins and fibres) (Pelgrom, Berghout, van der Goot, Boom, & Schutyser, 2014). As it has been shown in the case of emulsion stabilization, multicomponent fractions from yellow peas were capable to stabilize O/W emulsions (Geerts, Nikiforidis, van der Goot, & van der Padt, 2017). This indicates that there is no need to separate pure proteins in order to use them as emulsifiers and that potentially coarse protein mixtures have good emulsifying properties.

Highly purified sunflower seed proteins are being reported as efficient emulsifiers (González-Pérez and Vereijken, 2007). However, phenol oxidation occurs at protein extraction conditions (pH > 8) and covalent bonds between proteins and phenols are triggered, which affect protein properties, like chemical stability and solubility (Karefyllakis, Salakou, Bitter, van der Goot, & Nikiforidis, 2018; Sosulski, 1979). On the other hand, when mixtures of proteins and phenols are in native environment, only physical attractive forces between proteins and phenols occur, which has been proven to positively affect the protein interfacial and emulsifying properties (Karefyllakis, Altunkaya, Berton-Carabin, Van Der Goot, & Nikiforidis, 2017). Therefore, one realizes that full exploitation of all potentials of sunflower seeds and its by-products might require a different approach. Part of this novel approach is the investigation of the properties of sunflower proteins when natively embedded in complex molecular mixtures.

For the above reasons, the aim of this work was to investigate the interfacial properties and emulsification ability of molecular complex mixtures derived at neutral pH from sunflower seeds and link their properties with their molecular composition. The obtained mixtures were the solid residue after cold pressing (cake), which was then aqueously divided to a protein and a fibre-based mixture. All fractions showed high interfacial activity and relativity good emulsification ability, opening a new path towards efficient exploitation of plant-derived ingredients.

2. Materials and methods

2.1. Materials

Whole sunflower seeds (SF seeds) were granted from Cargill B-V (Amsterdam, The Netherlands). Sunflower oil (SF oil) was purchased from the local market and was filtered with silica (MP Alumina N-Super I, MP Biomedicals, Germany) as described by Berton et al. (Berton, Genot, & Ropers, 2011) to remove any polar compounds. Petroleum ether, ethanol, potassium monobasic dihydrate, potassium phosphate dibasic, sodium hydroxide and hydrochloric acid, Nile Blue were all purchased from Sigma Aldrich (Sigma, USA) and were of analytical grade. Qualitative filter paper Grade 595 1/2 was purchased by Whatman (GE Healthcare, USA). For all analyses ultrapure water was used.



Fig. 1. Process scheme of the steps to derive the three different complex molecular mixtures and pictures of these mixtures in powdered form.

2.2. Methods

2.2.1. Preparation of Sunflower press cake (SF cake)

2.2.1.1. Cold-pressing. The diagram of the processing steps that were followed and the composition of the samples are presented in Fig. 1. Whole (with hulls) sunflower seeds were pressed using an oil press (KK20 F Universal, Kern Kraft, Germany) with a standard seed screw. Pressing temperature was kept below 45 $^{\circ}$ C. The temperature was monitored with a thermometer and controlled with a water jacket. During the process, the oil and SF cake were collected in separate containers, as it is depicted in Fig. 1.

2.2.1.2. Milling. The SF cake was milled using a rotor mill (Pulverisette 14, Fritsch, Germany) at 8000 rpm and simultaneously sieved with an integrated sieve with 1 mm pore size.

2.2.2. Preparation of Protein and fibre-based mixtures

SF cake was dispersed in ultrapure water (1:20 w/v ratio) and the dispersion was stirred at 2000 rpm using an electric stirrer (IKA Eurostar digital Euro St-D, IKA, Germany) for 2 h. During this step the pH was monitored and constantly adjusted to 7.0 with small additions of NaOH solution (0.1 M). Afterwards, the dispersion was intensively

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