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Development of new apple beverages rich in isothiocyanates by using extracts obtained from ultrasound-treated cauliflower by-products: Evaluation of physical properties and consumer acceptance

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ABSTRACT

The objective of this study was to develop a new apple juice beverage enriched with isothiocyanates (ITC) – rich extracts obtained from cauliflower by-products. Ultrasound-assisted extraction (UAE) at different amplitudes (20–100%) and extraction times (0–10 min) at a frequency of 24 kHz was employed to obtain ITC-rich extracts. It was found that both amplitude and treatment time had a significant ($p < 0.05$) effect on the recovery of ITC from cauliflower leaves and stems, obtaining the highest yields of ITC from leaves (≈3000 μM) and stems (≈7000 μM) after UAE (80% amplitude, 3 min) and UAE (20%, 3 min), respectively. Moreover, the highest recovery of total phenolic compounds (TPC) (≈105 mg gallic acid equivalent (GAE)/L) μM) was found after UAE (100% amplitude, 3 min) of TPC from stems. ITC-rich extracts obtained from caulifower by-products at the optimum UAE conditions were added into apple juice (10–40%), thus increasing the ITC content of the juice and observing the highest values in the new beverage when the highest percentage (40%) was added. Significant differences in smell and taste were found in the apple juices containing 20% and 40% cauliflower extracts compared to control (0% UAE cauliflower waste extracts added) samples. However, the results showed that the beverage with 10% extract addition preserved well the sensorial properties with regard to control sample and no total colour differences (TCD < 3) were observed for any new sample compared to control. Therefore, the addition of extracts obtained after UAE of cauliflower wastes can be a useful tool to obtain new beverages rich in ITC although further research dealing with the bioaccessibility and bioavailability of these compounds in the new beverages should be conducted.

1. Introduction

A high consumption of Brassica vegetables has been shown to be correlated with a decrease of cancers such as lung, stomach, colon and rectal cancers (Holst and Williamson, 2004). These beneficial effects have been attributed to their high content in bioactive compounds such as polyphenols and glucosinolates. However, the biological activity of glucosinolates is not simple, as they are hydrolyzed by the endogenous enzyme myrosinase into isothiocyanates, which are also responsible of most of the anticarcinogenic properties of the Brassica vegetables (Zhang, 2004; Veeranki et al., 2015). Unfortunately, isothiocyanates can have undesirable sensory characteristics and tend to be disliked (Cox et al., 2012; Drewnowski and Gomez-Carneros, 2000; Reed et al., 2006). The solution is to extract these compounds from natural vegetable sources as well as from their derived wastes and by-products and use as health functional ingredients in food products (Barba et al., 2015a; Deng et al., 2014; Galanakis, 2013; Parniakov et al., 2015).

Cauliflower (*Brassica oleracea* var. Botrytis) is one of the main Brassicaceae crops and a great amount of cauliflower wastes and by-products are generated at various stages of production chain (Llorach et al., 2003). About 36% of waste is generated from total weight of cauliflower in terms of leaves and stems (Ospina Machado and Villamizar, 2004). Cauliflower has total glucosinolates content lower than other common Brassica vegetables (Tiwari and Cummins, 2013). However, although its total glucosinolates content is lower, cauliflower has the highest waste index (ratio of

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non-edible to edible portion after harvesting), which generates huge quantities of organic solid waste that creates an undesirable odour (Oberoi et al., 2007). Therefore, these wastes can constitute an important source of high-added value compounds which can be used as food additives for the development of new food products and/or nutraceuticals.

For the recovery of valuable compounds from plant food materials, wastes and by-products, several conventional extraction methods based on maceration and heat extraction at high temperatures (>60 °C) alone and/or combined with different solvents, which can be toxic (i.e., hexane, acetone, methanol, etc.) have been used. Moreover, the use of high temperatures may promote nutritional losses (Barba et al., 2014; Parniakov et al., 2014).

At this stage of development, there is a need to develop new extraction methods that can reduce the extraction time, process temperature and solvent consumption and contribute to higher extraction efficiency and lower energy consumption as compared to conventional extraction techniques (Deng et al., 2014; Galanakis, 2012; Koubaa et al., 2015).

For instance, both food researchers and food industry have shown an increased interest in the use of ultrasound-assisted extraction (UAE) as it may be a good alternative to conventional extraction methods to reocover bioactive compounds (Chemat et al., 2017; Pingret et al., 2013; Rombaut et al., 2014), especially phenolic compounds and isothiocyanates, from different plant food materials, wastes and by-products (Barba et al., 2015b; Deng et al., 2014; Roselló-Soto et al., 2014; Wang et al., 2011). This technique enhances extraction owing to cavitation phenomena, caused by creation of ultrasound pressure waves in the extraction solvents, thus facilitating cell disruption and solvent penetration. UAE can be used alone and/or combined with conventional extraction method as it requires low capital cost (Chemat et al., 2012; Tabasso et al., 2015). Moreover, this methodology can reduce the temperature and solvent amount, as well as the energy consumption, and the wastes generated when other processes are used (Chemat et al., 2017; Li et al., 2013). In addition, UAE can be combined with green solvents, thus improving high-added value compounds extraction yields (Barba et al., 2014; Grassino et al., 2016; Roselló-Soto et al., 2015; Šic-Žlabur et al., 2015).

Previous studies have assessed and added cauliflower wastes and by-products to foodstuffs such as beef sausage to enhance its nutritional value (Abul-Fadl, 2012) or even in cereal based ready-to-eat expanded snacks as a novel source of dietary antioxidants, fibre and proteins (Stojceska et al., 2008). Regarding beverages, cauliflower wastes and by-products have been only used to enrich a commercial chicken soup with its polyphenol extracts (Llorach et al., 2005). For instance, it will be interesting to work on the enrichment of liquid foods to enhance their nutritional quality using phytochemical extracts rich in bioactive compounds such as polyphenols and isothiocyanates.

The aims of the present work were 1) to prepare a powder rich in phenolic compounds and isothiocyanates from cauliflower wastes; 2) to study the effects of UAE on the recovery of phenolic compounds and isothiocyanates from cauliflower wastes powders and 2) to develop a beverage with the obtained extracts, rich in isothiocyanates (with potential health benefits). The physical properties (i.e. colour and/or viscosity); as well as the consumer acceptability of the new beverage were also studied.

2. Materials and methods

2.1. Chemicals and reagents

Distilled water, methanol, acetonitrile, potassium phosphate buffer, sulforaphane standard, 1,2-benzendithiol, acetate buffer, 2,4,6-tripyridyl-s-triazine, ferric chloride, ferrous sulphate, 2,2-diphenyl-1-picrylhydrazyl (DPPH), butylated hydroxyl anisole (BHA), Folin-Ciocalteu reagent, $N_{a2}CO_3$ and gallic acid were purchased from Sigma Aldrich (Dorset, UK).

2.2. Samples

2.2.1. Production of cauliflower by-product powder

Cauliflowers were purchased at a local market in Manchester, UK. These were separated into two parts, edible and non-edible. The non-edible part; stem and leaves constituted the cauliflower by-products. Prior to blanching, the stem and leaves were washed with water and immediately drained with paper towel to absorb the excess of water. The blanching procedure followed the conditions predetermined by several studies (Stojceska et al., 2008; Tanongkankit et al., 2010; Volden et al., 2008; Wennberg et al., 2006). Blanching was performed in a hot-water bath at 90 ± 2 °C for 1 min for both cauliflower stem and leaves. Blanched stem and leaves were immediately cooled in cold water, and chopped into small pieces. The prepared cauliflower by-products were dried within 10 min after preparation (Fig. 1). The samples were dried at 50–55 °C overnight using a Havest Maid harvest dryer (Hydraflow Industrial Ltd, Upper Hutt, New Zealand) to a final moisture content of approximately 10%. Following drying, the samples were milled using a 0.5 mm mesh screen ZM100 Mill (Retch, Dusseldorf, Germany) to develop a powder. They were then packed and sealed in polyethylene bags and kept at room temperature until needed for analysis or extraction. This process was conducted to enhance the myrosinase activity due to cell damage which occurs during blanching and drying accompanied by the hydrolysis for the formation isothiocyanates (Holst and Williamson, 2004).

2.2.2. Apple juice beverage containing isothiocyanates

Four beverages with cauliflower stem by-products extracts (having the highest isothiocyanates content) levels of 0% (control), 10%, 20% and 40% in apple juice, were prepared. The cauliflower extracts (stored at 4 °C) were added to the apple juice on the day of the analysis. Apple juice used was the Sainsbury's pure apple juice made from concentrate with a total sugar of 9.2 g/100 mL (packed in 1L box pack).

2.3. Ultrasound-assisted extraction

Fifty grams of cauliflower powder (leaves or stems) were weighed into a beaker and 100 mL of different solvents (distilled water, 70% methanol or 80% acetonitrile) were added. Each beaker was sonicated using the UP400S Ultrasonic Processor (Hielscher, Teltow, Germany) at a working frequency 24 kHz, and power 400W at 20% or 100% amplitudes for 5 min. The experiment was performed in duplicate for each solvent and amplitude. The mixture was then centrifuged (Rotanta 460R Centrifuge, Hettich lab Technology, Tuttlingen, Germany) at 1500*g* for 15 min. The supernatant solution was collected and placed into another beaker. The pellet was centrifuged for a second time at 1500*g* for 15 min with 100 mL of solvent. The second supernatant was also collected and combined with the previous one. The combined extracts were filtered through a Whatman® paper filter under vacuum conditions. For each sample, isothiocyanates, total phenols and antioxidant activity were determined.

2.4. Optimisation of solid-liquid extraction of isothiocyanates and phenolic compounds from cauliflower wastes powder

A response surface methodology (RSM) using a five-level-two-factor Central Composite Rotatable Design (CCRD) was used.