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New approaches for the effective valorization of papaya seeds: Extraction of proteins, phenolic compounds, carbohydrates, and isothiocyanates assisted by pulsed electric energy

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ABSTRACT

The study compares the efficiency of common aqueous extraction (CE) at different pH (2.5–11) and temperatures (20–60 °C) and extraction assisted by pulsed electric energy (pulsed electric fields, PEF or high voltage electrical discharges, HVED) of nutritionally valuable and antioxidant compounds from papaya seeds. The exponential decay pulses with initial electric field strengths of ≈13.3 kV/cm and ≈40 kV/cm for PEF and HVED treatments, respectively, were used. The number of pulses *n* was changed within 1–2000. The impacts of temperature and pH on extraction efficiency of different components (proteins, total phenolic compounds, carbohydrates, isothiocyanates) and antioxidant capacity were ambiguous. The highest values of nutritionally valuable and antioxidant compounds were obtained for HVED-assisted extraction. However, the application of HVED-treatment may produce undesirable contaminants (chemical products of electrolysis, free reactive radicals, etc.) and extracts were unstable and cloudy. On the other hand, the application of the two-stage procedure PEF + supplementary aqueous extraction (+SAE) that include PEF-assisted extraction as the first step, and +SAE at 50 °C, pH = 7 during 3 h as the second step, allowed noticeable enhancement of the yields (+200%) and antioxidant capacities (+20%) even at neutral pH. This method has high prospects of industrial applications for release of valuable components from papaya seeds.

1. Introduction

A large amount of wastes and by-products are generated during all the food life cycle of exotic fruits, from agriculture phase, up to industrial manufacturing and processing, retail and household (Mirabella, Castellani, & Sala, 2014). Among these, papaya is gaining popularity, now ranked third with 11.22 Mt, or 15.36% of the total tropical fruit production (Evans & Ballen, 2012). During papaya processing, a large amount of by-products, especially seeds and peels, are produced and discarded into the environment causing organic pollution (Koubala, Christiaens, Kansci, Van Loey, & Hendrickx, 2014). Traditionally, these by-products have been considered as a problem. However, it is recognized that papaya seeds have some beneficial properties mainly attributed to their content in high-added value compounds, especially bioactive compounds such as isothiocyanates and phenolic compounds, which can be used for application in nutraceutical supplements, dietary additives, new food and pharmaceutical products (Ayala-Zavala et al., 2011). In fact, papaya seeds have been used for decades in parts of Asia and South America as vermifugal agent and they have been used in folk medicine to facilitate good menstrual flow (Adebiyi, Adaikan, & Prasad, 2003; Thomas et al., 2009).

However, at this stage of development there is a lack of information about the different extraction methods that can be used for the recovery of high-added value compounds from papaya seeds.

The classical treatments (grinding, heating), and the different alternative treatments currently used in industry to make extractions easier, degrade and disrupt the tissue structure (membranes and cellular walls) in an uncontrollable way. Unfortunately, entirely disrupted tissue losses its selectivity (capacity to sieve) and becomes permeable not just for the target cell compounds, but for undesirable compounds (impurities) passing into the extract.

In this line, pulsed electric field (PEF) assisted extraction seems to be rather promising. There are many successful examples of PEF application for the recovery of high-added value compounds from plant food

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materials and by-products (Donsi, Ferrari, & Pataro, 2010; Knorr et al., 2011; Martin-Belloso & Soliva-Fortuny, 2011; Vorobiev & Lebovka, 2011). PEF is a non-thermal treatment of very short duration (from several nanoseconds to several milliseconds) with pulse amplitude from 100–300 V/cm to 20–80 kV/cm. Under the effect of PEF, the biological membrane is electrically pierced and losses its semi-permeability. The electrical permeabilisation of biological membranes (called electroporation) may be reversible or irreversible (Pavlin, Kotnik, Miklavčič, Kramar, & Lebar, 2008). Some previous studies have demonstrated that electroporation induced by moderate electric fields (0.5–5 kV/cm) conserves the cell wall network (Corrales, Toepfl, Butz, Knorr, & Tauscher, 2008; Fincan & Dejmek, 2002; Toepfl, Mathys, Heinz, & Knorr, 2006; Vorobiev & Lebovka, 2010). For the purpose of extraction, the capacity of the cell network to act as a barrier for the passage of some undesired compounds is a big advantage and allows an improvement of the extraction selectivity. The preliminary PEF-assisted extraction experiments for some agricultural materials (grapes, grape by-products, sugar beets, yeast and orange peel among others) confirm the possibility of attaining selective extraction (Boussetta, Lesaint, & Vorobiev, 2013; Boussetta & Vorobiev, 2014; Boussetta et al., 2009; Cholet et al., 2014; Luengo, Alvarez, & Raso, 2013). Furthermore, plant materials treated by PEF seem to be less altered than thermally treated materials, and can be used in some new auxiliary applications as part of their bio-refinement.

A considerable interest was also accorded to the study of high voltage electrical discharges (HVED). HVED-technology has been recently studied for enhancing extraction of bioactive compounds from different raw materials. The HVED leads to the generation of hot, localized plasmas that strongly emit high-intensity UV light, produce shock waves, bubbles cavitations and generate hydroxyl radicals during water photo-dissociation (Boussetta & Vorobiev, 2014).

The main aim of the present research is to evaluate the potential of pulsed electric energy (PEF and HVED) assisted extraction for recovery the high-added value products from papaya seeds. The effects of pH (2.5–11) and temperature (20–60 °C) on efficiency of recovery was also studied.

2. Material and methods

2.1. Samples

Papaya fruits (*Carica papaya* L.) were purchased at a local supermarket (origin Ecuador) and seeds were separated mechanically from the pulp. Fresh seeds of ripe *C. papaya* L. fruits were washed in distilled water. The dry matter content, measured by drying 25 g of the papaya seeds at 105 °C to constant weight, was about 25 wt.%. A suspension of papaya seeds (solid/liquid ratio = 1/10) in distilled water was prepared immediately before experiments.

2.2. Treatment by pulsed electric energy

PEF and HVED treatments were done using the high voltage pulsed power 40 kV–10 kA generator (Tomsk Polytechnic University, Tomsk, Russia) in cylindrical batch treatment chamber using different types of electrodes. PEF treatment was carried out between two plate electrodes ($d = 110$ mm) with 3 cm distance between them (Fig. 1a). HVED treatment was done using electrodes in needle-plate geometry. The distance between the stainless steel needle ($d = 10$ mm) and the grounded plate ($d = 25$ mm) electrodes was fixed at 1 cm (Fig. 1a). Papaya seeds suspension (300 g) was introduced between the electrodes before treatment. Treatment comprised the application of n successive pulses ($n = 1$–2000). The damped oscillations with effective decay time $t_p \approx 0.5 \pm 0.1$ μs and the exponential decay of voltage $U \propto \exp(-t / t_p)$ with effective decay time $t_p \approx 8.3 \pm 0.1$ μs were observed in HVED and PEF treatment modes, respectively (Fig. 1b). The initial voltage peak amplitude was $U = 40$ kV and the corresponding electric field strengths E were $\approx$13.3 kV/cm and $\approx$40 kV/cm for PEF and HVED treatments, respectively. The distance between pulses was $\Delta t = 2$ s. Note that for PEF and HVED extraction time was calculated as $t = n\Delta t$.

The initial temperature before PEF or HVED treatment was 20 °C and the final temperature after electrical treatment never exceeded 35 °C. Suspension temperature was controlled by K-type thermocouple

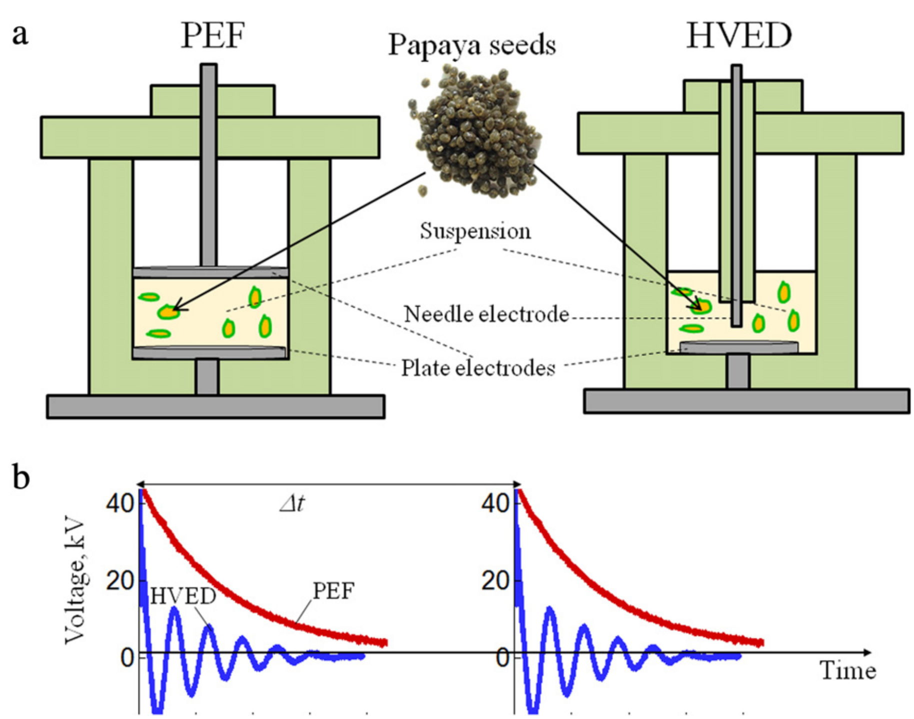

Fig. 1. PEF and HVED treatment chambers (a) and pulsed protocols (b).