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Impact of pulsed electric fields, high hydrostatic pressure, and thermal pasteurization on selected characteristics of *Opuntia dillenii* cactus juice

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ABSTRACT

Besides its bioactivity, *Opuntia dillenii* cactus juice exhibits desirable physicochemical properties. In the present study, impact of innovative pasteurization techniques – pulsed electric fields (PEF) and high hydrostatic pressure (HHP) – on the microbial inactivation, selected physicochemical properties and rheological characteristics, ascorbic acid, flavonols, betacyanins, and the antioxidant activity was evaluated in comparison to the non-pasteurized fresh juice. Results showed that PEF and HHP, as well as thermal pasteurization (TP), reduced the microbial populations found in juice from 10^3 to less than 10 cfu/mL. The non-Newtonian pseudoplastic flow behaviour of juice was maintained by all three methods. PEF and HHP resulted in a better retention of ascorbic acid, while TP reduced 22% of the ascorbic acid content. In contrast, TP caused a slight transformation of isorhamnetin glycosides. PEF and HHP maintained antioxidant activity of juice comparatively better than TP.

1. Introduction

Health benefits of *Opuntia dillenii* cactus fruit have been reported in different studies (e.g., Loro, del Rio, & Pérez-Santana, 1999; Díaz Medina, Rodríguez Rodríguez, & Díaz Romero, 2007; Böhm, 2008). *O. dillenii* (sometimes misleadingly described as *O. macrorhiza*) fruit is rich in betacyanins, to some extent similar to commercial red beets (*Beta vulgaris*) (Moussa-Ayoub, El-Samahy, Rohn, and Kroh (2011). Its juice has desirable characteristics of a low pH (approx. 3.6), moderate total soluble solids TSS (approx. 10°Brix), plain acidic taste, a plentiful deep red-purple color and a high content of ascorbic acid. But because of the high viscosity of the *O. dillenii* fruit mash, an enzymatic maceration is needed for *O. dillenii* fruit juice production (Moussa-Ayoub et al., 2016).

Consumers are increasingly demanding minimally processed healthy foods with more natural flavour and color, high quality and long shelf-life. Therefore, improving quality of the food products and developing the appropriate technologies used for food preservation is of key importance. Although an efficient inactivation of spoilage or pathogenic microorganisms can be achieved, thermal treatments might have a negative impact. In the meantime, also emerging and innovative non-thermal preservation technologies such as pulsed electric fields (PEF), high hydrostatic pressure (HHP), cold atmospheric plasma, or ultrasound are increasingly considered for preserving different foods (Barba, Esteve, & Frigola, 2012; Barba et al., 2015; Knorr et al., 2011). PEF is increasingly used for fruit juice preservation with products already available on the market. The short pulses of the applied electric field impact the permeability of biological membranes causing reversible or irreversible permeabilization depending on the purpose of treatment. For preserving foods, a high electric field strength in the range of 15–40 kV/cm is needed to achieve a complete irreversible permeabilization of

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the biological membranes resulting in the inactivation of vegetative microbial cells (Topfl, Mathys, Heinz, & Knorr, 2006; Knorr et al., 2011; Barba et al., 2015). With regard to the use of HHP for the preservation of high quality foods, a significant microbial inactivation in food products (e.g., fruit juices) is achieved by a pressure at 400 MPa or higher even at ambient temperatures within a sufficient holding time of several minutes (Castro & Saraiva, 2005; Patterson, 2005; Töpfl et al., 2006; Knorr et al., 2011; Barba, Esteve, & Frigola, 2012). Acidic foods (i.e. fruit juices) are particularly good items for high pressure processing and a pressure around 600 MPa is commercially preferred for achieving microbial inactivation (Castro & Saraiva, 2005).

In the present study, the impact of PEF and HHP on improving or maintaining the fresh characteristics of the fruit juice produced from *O. dillenii* cactus was compared to conventional thermal pasteurization (TP). The microbial load, selected physicochemical properties, and rheological characteristics, content of selected bioactive compounds, and the antioxidant activity of the pasteurized juices were determined.

2. Materials and methods

2.1. Plant sample, juice extraction and pasteurization

Samples from *Opuntia dillenii* cactus fruits were collected from an experimental field owned by the Suez Canal University (Ismailia, Egypt). After washing and removing glochides and distal parts, the fruits were mashed by interval mixing for 10 s. Then, mash was macerated using a commercial preparation of pectolytic enzymes (Fructozym® color, Erbslöh Geisenheim AG, Geisenheim, Germany). A dosage of 200 μL for every kg fruit mash was applied for a holding time of 60 min at 50 °C. Juice was obtained by raising pressure gradually to a maximum pressure level of 750 MPa using a manual laboratory juice presser. The collected juice was diluted with deionized water (juice: water ratio 2:3) and divided into four parts. Three parts were treated directly with pulsed electric fields (PEF), high hydrostatic pressure (HHP), or thermal pasteurization (TP). The fourth part was used as a control sample.

I) PEF was conducted using a laboratory continuous system with juice flow rate of 6.6 L/h and an electric field strength of 35 kV/cm. The system consisted of a 7 kW pulse modulator (Scandinova Systems AB, Uppsala, Sweden) providing rectangular monopolar pulses. A pulse width of 3 μs was used (frequency 45 Hz, pulse energy 3.5 J, current 58 A, voltage 21 kV). The juice was pre-heated from room temperature to 45 °C. An outlet temperature of 67 °C after PEF treatment was measured as a result of the dissipation of electrical energy based on a total specific energy input of 85 kJ/kg applied during the treatment. A co-linear treatment chamber was used. A detailed description including information on electric field strength distribution and flow characteristics can be found in Jäger, Meneses, and Knorr (2009).

II) HHP was conducted with a high pressure processing batch system U4000 (Institute for High Pressure Physics, Warsaw, Poland; Year of manufacture 2003; Max pressure of 800 MPa; Vessel volume of 0.75 L; Temperature range −25–100 °C). The juice was filled in sterilized plastic bottles and then putted in vacuumed plastic bags before treatment. A 1:1 mixture of deionized water and propylene-glycol was used as pressure-transmitting medium. The juice was subjected to pressure of 600 MPa at ambient temperature for a holding time of 10 min. The pressure vessel was at ambient temperature, juice temperature was 15 °C before pressure was built-up. Due to the adiabatic heat of compression, the temperature of the juice reached 35 °C under pressure and decreased slowly to 28 °C before pressure release after the holding time.

III) TP was conducted using continuous heating lab-scale system with a juice flow rate of 6.6 L/h. The pre-heating temperature of juice was 55 °C, followed by pasteurization at 95 °C of with a holding time of 3 min. Samples were collected in sterilized glass bottles.

2.2. Chemicals and reagents

The flavonol standards were purchased from Extrasynthese (Genay, France). HPLC solvents and further chemicals were purchased from Carl Roth GmbH & Co. KG (Karlsruhe, Germany).

2.3. Physicochemical properties

The pH and total soluble solids (TSS) was determined directly in the non-pasteurized and treated juices. Color parameters were measured directly in all juices using a Minolta Chroma Meter CR-300 (Tokyo, Japan). The results were expressed as tristimulus parameters (L*, a* and b*), where L* (indicates lightness), a* (red-green) and b* (yellow-blue).

2.4. Ascorbic acid content

As described by Moussa-Ayoub et al. (2016), an HPLC system, equipped with a simple pump, a UV detector (Knauer Wissenschaftliche Geräte GmbH, Germany) and a 250 × 4 mm Hypersil ODS column (Knauer, Germany), applying a tetrabutylammonium hydrogen sulfate/ddH_2O/methanol mixture as eluent at a flow of 1 mL/min, was used. Each juice sample was diluted (1:3) with *meta*-phosphoric acid (6%), vortexed and filtered prior to injection. An aliquot of dilution (20 μL) was applied. The ascorbic acid content was determined at a wavelength of 254 nm and expressed as mg/100 mL of fresh juice.

2.5. Rheological measurements

Rheological measurements were carried out using Rheometer MCR301 (Physica®, Anton Paar GmbH, Graz, Austria-Europe) equipped with a CC27/P1 cylindrical measuring system. Analysis of date was carried out using the software Rheoplus/32 Multi6 V3.40. The measurements were carried out at 20 °C. Shear rate test was performed at shear rate range of 0.1–100/s. The measurement time of each sample was 60 s with 30 measurement points (2 s for each measurement point). The flow behaviour data was produced according to the OSTWALD-DE WAELE model: $\tau = K \cdot \gamma^n$, and the apparent viscosity (η_{eff}) was calculated at shear rate 100/s by the equation: $\eta_{eff} = \tau/\gamma = K \cdot \gamma^{n-1}$, where: τ = shear stress (mPa), γ = shear rate (1/s), K = consistency index (mPa.s) and n = flow index.

2.6. Microbiologicial analyses

Yeasts and molds, acid tolerant microorganisms, and total colony count were determined in the non-pasteurized diluted juice and in the pasteurized juices after a storage at 8 °C for 15 d. A series of dilution up to 10^{-3} was made and 500 μL of diluted sample were plated on plate count agar for total colony count, orange serum agar for acid tolerant microorganisms, and oxytetracyline glucose yeast extract agar for determining yeasts and molds. Samples were incubated for 48 h at 25 °C (yeasts) and 32 °C for total colony count and acid tolerant microorganisms. The number of colony forming units was counted manually and the inactivation effect was