LWT - Food Science and Technology 84 (2017) 457–463

Contents lists available at ScienceDirect

LWT - Food Science and Technology

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

Effect of indirect cold plasma treatment on cashew apple juice (*Anacardium occidentale* L.)

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ARTICLE INFO

Article history:
Received 30 April 2017
Received in revised form
5 June 2017
Accepted 6 June 2017
Available online 8 June 2017

Keywords:
Indirect cold plasma
Vitamin C
Antioxidant potential
Sugar content
Cashew apple

ABSTRACT

The effects of the application of indirect cold plasma treatment on vitamin C, polyphenol and flavonoid contents, antioxidant activity (FRAP, DPPH, ABTS) and sucrose, fructose and glucose contents of cashew apple juice (*Anacardium occidentale* L) have been studied. Treatments were carried out by using a benchtop plasma system and two operative variables were considered: N_2 plasma flow rate (10, 30 and 50 mL/min) and treatment time (5, 10 and 15 min). The plasma treatment promoted an increment of the vitamin C, flavonoid and polyphenol content as well on the antioxidant activity. Overexposure to plasma resulted in a decrease of most bioactive compounds denoting the importance in optimizing the process conditions.

1. Introduction

The demand for juices in the world has been increasing due to higher availability of ready to drink juices in the supermarket, better nutritional quality when compared to soft-drinks, and increasing demand for more natural products. The increase in this demand has also increased the demand for products with a higher fresh-like characteristics of flavor, texture, color, aroma and overall appearance, instead of juices with off-flavors and artificial colors.

The production of juices with high fresh-like characteristics and long shelf-life is still a challenge in the food industry (Ramos, Miller, Brandão, Teixeira, & Silva, 2013). To address this challenge, new preservation techniques are being developed and tested. Power ultrasound (Fernandes, Rodrigues, Law, & Mujumdar, 2011; Ortuño, Martínez-Pastor, Mulet, & Benedito, 2012), high pressure (Liu, Zhang, Zhao, Wang, & Liao, 2016), pulsed ultraviolet light (Keyser, Műller, Cilliers, Nel, & Gouws, 2008; Tran & Farid, 2004) and microwave heating (Basak, Bhattacharya, & Panda, 2016) are among the new technologies that have been applied to fruit juice processing.

Cold plasma is an emerging technology that has been recently applied in many biological treatments including sanitization and surface modification. In brief, cold plasma is an ionized gas (carbon dioxide, argon, nitrogen, helium, oxygen or air) characterised by active particles such as electrons, ions, free radicals and atoms which are both in ground and excited states at ambient temperature (Niemira, 2012). The reactive species generated in the gas plasma such as singlet oxygen, OH radicals, and NO radicals have a direct effect on microbial inactivation and surface treatment of packing materials. These reactive species are produced by applying energy at specific frequencies to a gas or a gas mixture (Ramazzina et al., 2015; Takamatsu et al., 2015). The species generated and therefore their effect vary according to the parameters of plasma source such as voltage, frequency, flow rate and plasma generation method and depend mostly on the type of gas that is used (Pankaj et al., 2014; Ramos et al., 2013).

Cold plasma has been proven efficient in inactivating microorganism in several products like mango and melon (Perni, Liu, Shama, & Kong, 2008), lettuce (Grzegorzewski, Ehlbeck, Schlüter, Kroh, & Rohn, 2011), meat and chicken skin (Noriega, Shama, Laca, Díaz, & Kong, 2011), pear (Berardinelli, Vannini, Ragni, & Guerzoni, 2012), cabbage (Lee, Kim, Chung, & Min, 2015), strawberry (Misra et al., 2014), blueberry (Lacombe et al., 2015). According to the results presented by these authors, cold plasma is a valuable alternative for food decontamination due to its lethal microbiological power, reaching in some cases up to a 5-log

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http://dx.doi.org/10.1016/j.lwt.2017.06.010
0023-6438/

reduction in microorganism count.

More recently, the application of cold plasma has been focused on the preservation of qualitative characteristics of fresh cut fruits and vegetables. Studies were addressed to the enzyme inactivation in tomato (Pankaj, Misra, & Cullen, 2013) and apple (Tappi et al., 2014); morphology, microstructure and rheological properties of wheat flour (Misra et al., 2015), rice (Thirumdas, Deshmukh, & Annapure, 2015) and maize starch (Bie et al., 2016).

The evaluation of the effect of the reactive species on the nutritional content and selected compounds is still scarce but it has been carried out on kiwi (Ramazzina et al., 2015), orange juice (Almeida et al., 2015; Alves Filho et al., 2016), pomegranate juice (Bursać Kovačević et al., 2016; Herceg et al., 2016) and melon (Tappi et al., 2016). The findings of these authors showed that cold plasma is an interesting technology that can produce fresh-like products with minimal degradation of biocompounds like vitamin C, carotenoids, polyphenols, flavonoids and anthocyanins. Regarding fruit juices, only the results of two juices have been reported not allowing a strong conclusion about whether this technology would be also applicable to other juices and its effects might be similar from a matrix food to another. To continue the development of this technology, our study aimed to apply indirect cold plasma to a fruit juice that has a high vitamin C and polyphenols content.

Cashew apple is a hard, pear-shaped, small and non-climacteric pseudo-fruit (peduncle) associated to the cashew nut (the real fruit of the cashew tree), which is well known around the world (Assunção & Mercadante, 2003a). According the Food and Agriculture Organization of the United Nations (FAO) the world production of cashew apple in 2013 was of 2,001,905 tons, with India, Vietnam and Brazil being main producers (FAO, 2013). The cashew apple corresponds to 90% of the weight of the fruit and has an exotic aroma and flavor and it is preferably marketed as frozen pulp, juice, and nectar.

Cashew apple is a rich source of ascorbic acid, minerals, organic acids, phenolic compounds, carbohydrates and reducing sugars like fructose and glucose (Queiroz, Lopes, Fialho, & Valente-Mesquita, 2011; Rabelo, Fontes, & Rodrigues, 2009; Schweiggert et al., 2016). Cashew apple juice has been considered as a good source of vitamin C (1.21–2.00 mg/g) by Assunção and Mercadante (2003b) and by Queiroz et al. (2011). Moo-Huchin et al. (2014) observed that cashew apple exhibited a very high content of antioxidant compounds among several tropical fruits. The fruit contains about 1.8 mg/100 g of total anthocyanins, 287 mg GAE/100 g of phenolic compounds, 345 mg/100 g (quercetin equivalent) of flavonoids and 11.6 mg/100 g of carotenoids. The antioxidant capacity of the fruit is also high among several commercial fruits, presenting antioxidant activities measured by DPPH EC_{50}, ABTS and FRAP assays of 9397 g/g DPPH, 7.8 μmol Trolox/g, and 22.9 μmol Fe_2SO_4/g, respectively (Rufino et al., 2010).

The main objective of this study was to evaluate the effect of cold plasma (N_2) application (N_2 flow, treatment time) on vitamin C, glucose, sucrose and fructose contents, antioxidant activity, and polyphenol and flavonoid contents of cashew apple juice.

2. Materials and methods

2.1. Cashew apple

Fresh, non-pasteurized frozen pulp of cashew apple (*Anacardium occidentale* L.) was obtained from Kipolpa (Fortaleza, Brazil), which was stored frozen in a refrigerator. The moisture and soluble solid contents of the juice were of 88.5% and 10.4%, respectively and its pH was of 4.7.

2.2. Indirect cold plasma treatments

Cold plasma treatments were carried out by using a benchtop plasma system PE-100 (Plasma Etch, USA). Plasma was generated in nitrogen (grade FID 4.5, purity 99.95%, White Martins, Brazil) applying an 80 kHz electric field through the electrode. The process was carried out in indirect plasma mode meaning that the plasma was generated and then fed to the processing chamber, which contained the sample. Three different gas flow rates (10, 30 and 50 mL/min) were studied. Three polypropylene falcon tubes containing 10 mL of juice were indirectly treated with the cold plasma during 5, 10 and 15 min under vacuum conditions (30 kPa). After treatment, samples were centrifuged at 1699 CFG for 5 min in a 2–16 LK centrifuge (Sigma, Germany) and then filtered. The supernatant was used as extract for the subsequent chemical analysis.

2.3. Vitamin C determination

Vitamin C content in the cashew extracts was determined by using a modification of the method proposed by Salkić, Keran, and Jašić (2009). An aliquot of 1.0 mL of the extract was homogenized with 10 mL of 0.056 mol/L sodium oxalate solution using an Ultra-Turrax T25 Digital (IKA, Germany) at 13000 rpm for 30 s. The extraction mixture was left standing for 5 min and then it was filtered. An aliquot of 0.5 mL of the extract was diluted to 5.0 mL with 0.056 mol/L sodium oxalate (this dilution was done because of the high vitamin C content of the juice as to avoid out of scale UV-Vis absorbance measurements). Absorbance readings were made in an UV–Vis spectrophotometer Evolution 201 (Thermo Scientific, China) at 266 nm, 25 °C, using a 0.056 mol/L sodium oxalate solution as blank, and 10 mm quartz cuvettes. A 5-point calibration curve was made using L-ascorbic acid as standard.

2.4. Antioxidants and antioxidant capacity

The antioxidant potential content was measured by means of the total phenolic content (TPC) according to the Folin-Ciocalteu assay proposed by Rufino et al. (2010) and by means of the total flavonoid content (TFC) according to the aluminum chloride assay proposed by Paz et al. (2014).

The antioxidant activity was measured by FRAP, DPPH and ABTS assays following the methods described by Queiroz et al. (2011), Martínez et al. (2012) and Moo-Huchin et al. (2014), respectively. The absorbance measurement was taken in an UV–Vis spectrophotometer Evolution 201 (Thermo Scientific, China), using 10 mm quartz cuvettes.

2.5. Sugar contents determination

The HPLC determination of the sugars was carried out following the method reported by Almeida et al. (2015). Samples were filtered through a C18 cartridge and a 0.45 μm HA membrane cellulose ester (13 mm in diameter, white, smooth) prior to injection. HPLC analyses were carried out in a 1260 Infinity Quaternary LC System (Agilent technologies, USA) equipped with a pump system and IR detector. Sugars were analyzed onto a Supelcogel Ca column (300 × 7.8 mm) (Supelco Analytical) and kept at 80 °C. The injection volume was of 20 μL. An isocratic elution was performed with deionized water as mobile phase for 40 min (flow rate of 0.5 mL/min). Determination of sucrose (retention time: 9.0 min), glucose (retention time: 11.0 min) and fructose (retention time: 14.0 min) was based on calibration curves were obtained mixing these sugars at different concentrations: 1.0–10.0 g/L.