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Original Research Article

Dietary fiber, phytochemical composition and antioxidant activity of Mexican commercial varieties of cactus pear

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ABSTRACT

Cactus pear is one of the most important fruit in Mexican culture; however the phytochemical characterization of the whole cactus pear (pulp, peel and seeds) has not been studied. Therefore, the goal of this research was to evaluate the dietary fiber, vitamin C, total phenolic (TP), flavonoid and betalain concentrations and antioxidant activity (AOX) in the pulp, juice, peel and seeds of four Mexican commercial varieties of cactus pear. Rojo Cenizo and Rojo San Martín pulps presented concentrations of total dietary fiber, vitamin C, TP, betalains, and AOX of 145–166 g, 3–7 g ascorbic acid, 3–6 g gallic acid equivalents (GAE), 500–3444 mg betanin equivalents and 1044–5954 mg indicaxanthin equivalents, and 46–67 mm Trolox equivalents (TE) per kg, respectively. Rojo San Martín peel presented TP content over two times greater than the pulp, whereas AOX levels of Rojo San Martín and Verde Villanueva peels were 4–9 higher than their respective pulp. Cactus pear peel was the most important source of phytochemical compounds and AOX of the fruit, because of that it has to be considered in future applications for the formulation of new health-promoting ingredients and/or foods.

1. Introduction

Cactus pear (*Opuntia ficus-indica*), the cactus fruit, is characterized by a fleshy, juicy pulp intermixed with a great number of small seeds, and enclosed by a thick peel with small thorns. The weight proportions of pulp, peel, and seeds are 28–58%, 37–67% and 2–10%, respectively. México is the main producer of cactus pear accounting for 44% of the world production (428,300 t/year), displaying vast genetic variability, which is reflected in the great diversity of pulp and peel colors: red, violet, green, yellow (Castellanos-Santiago and Yahia, 2008; El-Kossori et al., 1998; Financiera Rural, 2011). The annual harvest estimated of cactus pear in the San Sebastian Villanueva, Puebla area is around 85,054 t, making it one of the most important producer regions in México (SIAP-SAGARPA, 2010).

Cactus pear pulp has been studied due to the presence of numerous compounds (dietary fiber, vitamin C, phenolic compounds), with the potential to provide important benefits like intestinal, cardiovascular and hepatic health, antioxidant activity and cancer prevention have also been reported (Chang et al., 2008; Chavez-Santoscoy et al., 2009; El-Kossori et al., 1998; Fernández-López et al., 2010; Galati et al., 2005; Kuti, 2004). However, the information concerning the phytochemical characterization of peel and seeds is scarce, in spite of these residues constituting up to 60% of the total weight of the fruit (Felker et al., 2005).

The demand for healthful foods has been growing in recent years, for that reason the food industry is constantly looking for new sources of nutritional and healthful components (González-Díaz et al., 2012), one of them could be the cactus pear. Therefore the goal of this study was to evaluate the dietary fiber (soluble and insoluble), vitamin C, total phenolic, flavonoid, and total betalain concentration, as well as the antioxidant activity of the pulp, peel and seeds of four Mexican commercial varieties of cactus pear (*Opuntia ficus-indica*). The obtained information will permit us to establish which fruit components and varieties are suitable for elaborating new health-promoting ingredients and/or foods.

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2. Material and methods

2.1. Reagents

Dietary fiber Kit K-TDFR was obtained from Megazyme (Bray, Co. Wicklow, Ireland). Methanol and water used for high performance liquid chromatography (HPLC) were obtained from Honeywell Burdick and Jackson (Radnor, PA, USA). HPLC grade standards of quercetin (purity ≥ 98%) and kaempferol (purity ≥ 96%) were purchased from Sigma–Aldrich Corp. (St. Louis, MO, USA), and isorhamnetin (purity ≥ 99%) was obtained from Indofine Chemical Company, Inc. (Hillsborough, NJ, USA). Folin–Ciocalteu reagent (2 N), gallic acid (purity 97.5–102.5%), L-ascorbic acid (purity ≥ 98%), 2,2′-azobis(2-amidinopropane) dihydrochloride, Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, purity ≥ 97%), trichloroacetic acid, DL-dithiothreitol, N-ethylmaleimide, orthophosphoric acid, ferric chloride, 2,2′-bipyridyl, and HPLC-grade formic acid were purchased from Sigma–Aldrich Corp. (St. Louis, MO, USA). Fluorescein was obtained from Riedel de Haën (Seelze, Germany). All of the other reagents were of analytical grade unless otherwise stated.

2.2. Sample preparation

Four varieties (Rojo San Martín, Verde Villanueva, Rojo Cenizo, and Cristal) of cactus pear fruit (*Opuntia ficus-indica*) were harvested at San Sebastián Villanueva, Puebla (latitude 19°03′37″, longitude 97°42′58″ and altitude 2332 m) in the years 2010 and 2011. Cactus pear fruit was harvested by expert collectors and mixed randomly, and then ~15 kg of each variety were shipped in crates at 20 °C and delivered within 4 days. Fifteen fresh and ripe fruit were randomly selected to determine each weight with an electronic scale (Torrey, EQ-4HP, Monterrey, NL, Mexico), and measure the polar and equatorial dimensions with a Vernier. Peel was manually removed while pulp and seeds were separated with a juice extractor (Turmix, uso rudo, Ocoyoacac, Estado de Mexico, Mexico). Afterwards, half of the pulp obtained was filtered (Whatman no. 1 paper) to separate both the solid (cloud) and liquid (juice) fractions. Cactus pear fruit components (peel, seeds, pulp and juice) were freeze-dried at −86 °C/100 Pa (Vir Tis, FM 25EL, Gardiner, NY, USA). Liquid pulp and juice, and lyophilized samples (pulp, juice, peel and seeds) were stored at −80 °C until the time of their analysis. Lyophilized peel and seeds were crushed with a blender (Oster, BRLY07-200-013, Monterrey, NL, Mexico) prior to storage at freezing conditions.

The studies in pulp and juice were carried out as follows: phytochemical content and antioxidant activity were determined from liquid samples, while dietary fiber and flavonoids were determined from the lyophilized samples. Similarly, in lyophilized peel and seeds evaluations were done on the dietary fiber, phytochemical composition and antioxidant activity.

2.3. Physical and physicochemical characterization

Total soluble solids, pH and titratable acidity were determined for the pulp from 15 fresh fruit. Total soluble solids (°Brix) were evaluated by using a digital refractometer (ATAGO, N-1α, Tokyo, Japan), pH was determined with a glass-electrode pH meter (Thermo Scientific, Inc., Orion 3-Star, Waltham, MA, USA), and titratable acidity was measured following AOAC (1995) procedure 942.15 (g of citric acid/kg fresh weight (fw)). Moisture was carried out in the pulp, juice, peel and seeds according to AOAC (1995) procedure 925.45 (% in fw).

2.4. Phytochemical characterization

2.4.1. Dietary fiber

Soluble (SDF) and insoluble dietary fiber (IDF) were determined according to AOAC (1995) method 991.43. Total dietary fiber (TDF) was the sum of SDF and IDF. Results were reported as g dietary fiber per kg of dry weight (dw).

2.4.2. Vitamin C

The extraction and quantification of vitamin C content were performed according to the method described by Gillespie and Ainsworth (2007) using a kinetic microplate reader (Bio-Tek Instruments, Inc., Synergy HT, Bad Friedrichshall, Germany). Results were reported as g ascorbic acid (AA)/kg (dw).

2.4.3. Total phenolics

Total phenolic (TP) extraction was carried out in peel and seeds, 1 g of lyophilized sample was added to 10 mL of methanol (80%), and mixed for 2.5 h (250 rpm, 25 °C) in a shaker lab (Lab Line Instruments, Inc., 26, Melrose Park, IL, USA). The mixture was centrifuged for 10 min at 5000 × *g* and 4 °C (VWR International LLC, Galaxy 16DH, Radnor, PA, USA), and the supernatants were collected. TP was also directly analyzed in pulp and juice. Total phenolic content was developed following the Folin Ciocalteu method, and measured after 90 min of reaction at 725 nm (Stintzing et al., 2005). Results were expressed in g of gallic acid equivalents (GAE)/kg (dw).

2.4.4. Flavonoids

Flavonoids were extracted and quantified according to a method adapted from Fernández-López et al. (2010), with the only difference that the acid hydrolysis of the flavonoid conjugates was carried out with 300 mg of sample mixed in 3 mL of methanol and 400 μL of HCl (6 N), in a water bath at 90 °C for 20 min (Thermo Fisher Scientific, Inc., 102, Waltham, MA, USA). Flavonoids were analyzed by HPLC-PDA (Agilent Technologies, Inc., 1200 Series, Santa Clara, CA, USA) using a column Zorbax SB-C_{18}, (3.0 mm × 100 mm, particle size 3.5 μm) and gradient elution with (A) HPLC-grade water with 0.1% (v/v) formic acid and (B) HPLC-grade methanol. Separation was achieved at 35 °C and with a flow of 0.4 mL/min using the following gradient: 30% B (0 min), 40% B (5 min), 45% B (10 min), 45% B (15 min), 50% B (20 min), 70% B (25 min), 100% B (30 min). Prior to the next injection, the gradient was backed to 30% B (2 min) and the column re-equilibrated at 30% B (5 min). Flavonoid identification was based on comparison of HPLC retention times and absorption spectrums with their respective standards. Chromatograms were obtained at 365 nm and the results were expressed in mg/kg (dw).

2.4.5. Total betalains

Total betalain extraction was carried out in peel and seeds; one gram of lyophilized sample was mixed with 5 mL of water for 2.5 h at 250 rpm and room temperature in a lab shaker. Betalains were directly analyzed in pulp and juice. The mixture was centrifuged for 10 min at 5000 × *g* and 4 °C, and supernatants were collected for betalain quantification as reported by Cassano et al. (2007). Pigment content was calculated using the molecular weight and molar extinction coefficient of betanin (λ_{max} = 538 nm; 550 g/mol; 60,000 L/mol cm) and indicaxanthin (λ_{max} = 480 nm; 308 g/mol; 48,000 L/mol cm) (Stintzing et al., 2005). Results were expressed as mg of betanin equivalents and as indicaxanthin equivalents/kg (dw).

2.5. Antioxidant activity

The analyzed samples for the evaluation of the antioxidant activity (AOX) were prepared with the same process as TP