

## Antioxidant components and nutritional quality of 15 genotypes of Xoconostle (*Opuntia* spp.)

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### ABSTRACT

This study determined nutritional quality, content of antioxidant compounds and antioxidant activity in the rind (mesocarp) of the fruit of 15 genotypes of xoconostle to identify those that have greater nutraceutical potential. A proximal analysis was conducted. Contents of betalain, total phenolic compounds, flavonoids, ascorbic acid and antioxidant activity were quantified. The data obtained were analyzed with an analysis of variance (ANOVA), the Tukey test ( $P < 0.05$ ) and the Pearson coefficient of correlation. Significant differences in the proximal composition and antioxidant compounds were found among the 15 genotypes. The genotype Cambray had the highest total content of betalains (35.06 mg 100 g<sup>-1</sup> f.w.). The antioxidant activity observed in the other genotypes may be due to a synergetic effect of the presence of total betalains, phenolic compounds, flavonoids and vitamin C. The wild genotype Chaveñito (*O. sainaltense*), which had the highest percentage (95.88 %) of inhibition of the DPPH radical (indicating greater antioxidant activity), can be considered the genotype with the best nutraceutical quality.

**Key words:** phenolic compounds, flavonoids, vitamin C, antioxidant activity.

### INTRODUCTION

Mexico is the country with the largest and most important diversity of cacti (Bravo and Scheinvar, 1999; Esquivel, 2004; Reyes-Agüero *et al.*, 2006). The genus *Opuntia* is highly diverse and probably the most important because of its widespread distribution (Reyes-Agüero *et al.*, 2006; Sumaya-Martínez *et al.*, 2011). The xoconostle (*Opuntia* spp) is one of the natural resources of this genus that has been little used and commercialized.

Few studies have determined moisture, crude protein and fiber contents or the nutraceutical quality of the xoconostle fruit, in spite of the broad diversity of its varieties. Xoconostle has existed in Mexican cuisine since pre-Columbian times and as raw material for producing wines, liqueurs, candy, jams and jellies. The fruit is also consumed dry, crystalized or in syrup (Scheinvar *et al.*, 2009; Morales *et al.*, 2012). Moreover, medicinal properties (hypoglycemic and hypolipemic) have been attributed to xoconostle (Pimienta-Barrios *et al.*, 2008; Osorio-Esquivel *et al.*, 2011). Few commercial xoconostle varieties have been studied in terms of their phytochemistry or their antioxidant activity. Distribution of some phytochemicals varies depending on their location in the pericarp, endocarp (mucilage and seeds) or mesocarp (rind). The rind is the part of the fruit that is consumed ( $66.91 \pm 1.12$  % of the total fruit weight) (Osorio-Esquivel *et al.*, 2011). Recent studies highlight the presence of sugars, dietetic fiber, ascorbic acid, phenolic compounds and pigments (betalains) (Pimienta-Barrios *et al.*, 2008; Osorio-Esquivel *et al.*, 2011). Betalains are responsible for the array of fruit colors in the many species and varieties of the genus *Opuntia* (Stintzing and Carle, 2007). These pigments exhibit important antioxidant activity with non-toxic effects for humans (Sumaya-Martínez *et al.*, 2011). High levels of betalains help prevent cancer and lipid oxidation of membranes (Livrea and Tesoriere, 2006). Phenolic compounds are another group of secondary metabolites, identified in some fruits of the *Opuntia* genus (Osorio-Esquivel *et al.*, 2011; Pimienta-Barrios *et al.*, 2008), that protect plants from oxidative stress, and in human food, they contribute to preventing disease. Few studies describe the presence of flavonoids in cactus fruits (Moussa-Ayoub *et al.*, 2011). These metabolites are also important as they have antioxidant, anti-inflammatory, and anticancer properties (Crozier *et al.*, 2009).

Xoconostle could be considered a functional food if the presence and content of nutraceutical ingredients related to decreasing or preventing disease were known (Badimon *et al.*, 2010; Das *et al.*, 2012). Moreover, knowledge of the nutraceutical properties of the fruit could contribute to more efficient agroindustrial use (Bernal *et al.*, 2011). For these reasons, this study was conducted to determine the nutritional quality and content of antioxidant compounds in the pulp of the fruit of 15 genotypes of xoconostle to identify those with more nutraceutical attributes. This study contributes knowledge of a food resource used ancestrally and still a part of the cultural identity of some states of the Mexican Republic but today has little recognized potential.

## MATERIALS AND METHODS

### Plant material

Fruits of fifteen xoconostle *Opuntia* spp. genotypes (Table 1) were collected in four states of Mexico (Aguascalientes, Hidalgo, State of México and Zacatecas). The genotypes Chatito, Cuerón and El Aguacero were obtained from wild *Opuntia* populations in Palo Alto, municipality El Llano, Aguascalientes, located at  $21^{\circ} 54' N$  and  $101^{\circ} 58' W$  at an altitude of 2015 m. Climate is BS<sub>1</sub> kw(w)g, temperate, mean annual temperature  $17.2^{\circ} C$  and yearly precipitation is 485.7 mm. The xoconostles Invierno, Matizado and Del Borrego were harvested in a community collection in the municipality of Villa de Tezontepec, Hidalgo, located at  $19^{\circ} 53' N$  and  $98^{\circ} 49' W$ , 2320 m altitude. The climate is BS1hw, temperate

semiarid, mean annual temperature 14.5 °C and 508 mm annual precipitation (Gallegos-Vázquez *et al.*, 2012). The genotypes Cuaresmeño Blanco and Cuaresmeño Rojo were collected from commercial plantations in the municipality of Otumba, State of Mexico, located at 19° 42' N and 98° 45' W, 2349 m altitude; climate is C(w2) temperate subhumid, mean annual temperature 14.8 °C and yearly precipitation 514.3 mm (García, 1988). Finally, the genotypes Cuaresmeño Zacatecano, Blanco Jaspeado, Rojo Sainero, Chaveñito, Café, Rosita and Cambray were obtained from wild populations growing on communal rangelands of the municipality Saín Alto, Zacatecas, located at 23° 34' N and 103° 14' W, at 2050 m altitude, where climate is BS1hw, temperate semiarid, mean annual temperature 16.0 °C and annual precipitation 500 mm (Table 1) (Gallegos-Vázquez *et al.*, 2012).

**Table 1.** Location of collection and characteristics of the 15 xoconostle genotypes

No.	Species	Genotype (Common name)	Origin	Condition	Color
1	<i>O. matudae</i> Scheinvar	Cuaresmeño blanco	Otumba, Méx.	Comercial	Green
2	<i>O. matudae</i> Scheinvar	Cuaresmeño zacatecano	Saín Alto, Zac.	Silvestre	Green
3	<i>O. matudae</i> Scheinvar	Rojo sainero	Saín Alto, Zac.	Silvestre	Red
4	<i>O. matudae</i> Scheinvar	Cuaresmeño rojo	Otumba, Méx.	Comercial	Red
5	<i>O. matudae</i> Scheinvar	Cuerón	Palo Alto, El Llano, Ags.	Silvestre	Pink
6	<i>O. matudae</i> Scheinvar	El aguacero	Palo Alto, El Llano, Ags.	Silvestre	Pink
7	<i>O. duranguensis</i> Britton & Rose	Cambray	Saín Alto, Zac.	Silvestre	Purple
8	<i>O. duranguensis</i> Britton & Rose x <i>O. joconostle</i> F.A.C. Weber	Blanco jaspeado	Saín Alto, Zac.	Silvestre	Yellow
9	<i>O. joconostle</i> Weber	Rosita	Saín Alto, Zac.	Comunitaria	Pink
10	<i>O. sainaltense</i> Scheinvar	Chaveñito	Saín Alto, Zac.	Silvestre	Red
11	<i>O. joconostle</i> Weber	Chatito	Palo Alto, El Llano, Ags.	Silvestre	Pink
12	<i>O. tezontepecana</i> Gallegos & Scheinvar	Invierno	Villa de Tezontepec, Hgo.	Comunitaria	Orange
13	<i>O. leucotricha</i> Salm-Dick x <i>O. joconostle</i> Weber	Café	Saín Alto, Zac.	Silvestre	Brown
14	<i>O. leiascheinvariana</i> Martínez & Gallegos	Matizado	Villa de Tezontepec, Hgo.	Comunitaria	Pink
15	<i>O. oligacantha</i> Föster	Borrego	Villa de Tezontepec, Hgo.	Comunitaria	Purple

The fruits of each genotype were harvested considering their commercial maturity index. For those that were non-commercial, the criteria used were those commonly sought for consumption (fruit development, shape, depth of receptacle and peel color). The fruits were sampled randomly and were free of pests and disease. The fruits were washed to remove glochids and the mesocarp was separated from the cuticle (pericarp; 1 to 2 mm thick), pulp and seeds. The rinds were sliced at uniform thickness ( $0.3 \pm 0.1$  cm) and, in jute bags, they were frozen by immersion in liquid nitrogen and later lyophilized (Labconco Lyophilizer, USA) at -40 °C and 12 Pa for 24 h. The samples were kept at room temperature away from light

until analysis. The fresh (f.w.) and lyophilized (d.w.) rinds of the xoconostle fruits were weighed to determine concentrations of the phytochemicals analyzed.

### **Proximal analysis**

Moisture, ash, crude protein, lipids and crude fiber contents were determined in the dehydrated samples (dry weight) following the AOAC (2000) methodology. The content of carbohydrates was calculated using the formula of Audu and Aremu (2011):  $TC = 100 - (CP + L + C)$ , where TC = total carbohydrates (%); CP = Crude protein (%); L = lipids (%), C = ash.

### **Quantification of total betalains**

Pigment contents were determined with a modified version of the method described by Stintzing *et al.* (2003). Ten mL of distilled water was added to 0.1 g lyophilized rind. This mixture was left in repose for 15 h at room temperature. The sample was sonicated for 20 min in an ultrasonic bath (Cole-Parmer, USA). Absorbance of the extracts was read in a spectrophotometer (Genesys 10 s) at the wavelengths of 480 nm (betaxanthins-BX) and 540 nm (betacyanins-BC). Quantification was done with the following formula: betaxanthins or betacyanins ( $\text{mg g}^{-1}$ ) =  $(A \times DF \times MW \times V) / (\epsilon \times L \times DW)^{-1}$ ; where A = absorbance at 480 and 540 nm for betaxanthins and betacyanins, respectively; DF = dilution factor; MW = molecular weight (indicaxanthin:  $308 \text{ g mol}^{-1}$  and betanin:  $550 \text{ g mol}^{-1}$ ); V = volume of extract (mL),  $\epsilon$  = coefficient of molar extinction  $48,000 \text{ L (mol cm)}^{-1}$  for indicaxanthin and  $60,000 \text{ L (mol cm)}^{-1}$  for betanin; L = cell length (1 cm); DW = dry sample weight (g) (Chauhan *et al.*, 2013).

### **Quantification of total phenolic compounds**

To determine total phenols, the method described by Waterman and Mole (1994) was used. Twenty-five mL 95 % aqueous ethanol (v/v) was added to 0.1 g lyophilized rind of each genotype. The mixture was sonicated for 20 min in an ultrasonic bath. The supernatant was gauged to 25 mL with 80 % aqueous ethanol (v/v). An aliquot of 0.5 mL of ethanolic extract was mixed in a vortex with 10 mL 10 % (p/v) sodium carbonate ( $\text{Na}_2\text{CO}_3$ ). Each sample was placed in a water bath at 38 °C for 15 min, after which 3 mL distilled water and 1 mL Folin-Ciocalteu solution and water (1:1, v/v) were added to 1 mL of the mixture. The mixture was left to repose for 15 min in the dark at room temperature. Absorbance was read at 660 nm in a spectrophotometer. The calibration curve was constructed with gallic acid for quantification of the metabolites. Total phenol content was expressed in mg gallic acid equivalents per 100 g fresh weight ( $\text{mg GAE } 100 \text{ g}^{-1} \text{ f.w.}$ ).

### **Quantification of flavonoids**

Flavonoid content was determined following the method proposed by Chang *et al.* (2002). To the ethanolic extracts prepared previously, 1.5 mL 95 % (v/v) ethanol, 0.1 mL 10 % (w/v) aluminum chloride ( $\text{AlCl}_3$ ), 0.1 mL 1.0 M potassium acetate solution, and 2.8 mL distilled water were added. The mixture was homogenized in a vortex and incubated for 30 min at room temperature. Absorbance was read at a wavelength of 415 nm in a spectrophotometer. The standard curve was constructed with quercetin as reference. The results were expressed in mg quercetin equivalents in 100 g fresh weight ( $\text{mg QE } 100 \text{ g}^{-1} \text{ f.w.}$ ).

### **Quantification of ascorbic acid**

Ascorbic acid (vitamin C) concentration was determined following the official AOAC (2002) method. Ten mL metaphosphoric acid-acetic acid ( $\text{HPO}_3\text{-CH}_3\text{COOH}$ ) were added to 1 g lyophilized rind. The mixture was filtered and gauged with the solution of metaphosphoric acid-acetic acid to a volume of 10 mL. The resulting mixture was titrated with a 0.05 % (w/v) solution of 2,5-dichloroindophenol. The metabolite was quantified by applying the following formula:  $\text{mg ascorbic acid g}^{-1} = (X - B) (F \times E^{-1}) (V \times A^{-1})$ , where X = average mL added for sample titration; B = average mL for titration of the blank; F = mg ascorbic acid equivalent to 1 mL standard solution of 2,5-dichloroindophenol; E = g sample; V = initial sample volume; Y = final sample volume after titration. The concentration of ascorbic acid was expressed as mg equivalents of ascorbic acid per 100 g dry sample weight mg AAE 100 g<sup>-1</sup> f.w.).

### **Antioxidant activity assessment**

Antiradical activity was determined by the free DPPH radical method described by Brand-Williams *et al.* (1995). Twenty mL methanol was added to 1 g lyophilized xoconostle rind; the mixture was sonicated for 20 min and the solvent was eliminated with a Büchi rota-evaporator. To each 1 mL of methanolic extract, 3 mL of 0.1 mM DPPH solution was added for the DPPH radical reduction reaction. The mixtures were maintained in reaction in darkness for 30 min at room temperature. Absorbance was then measured at 516 nm in a spectrophotometer. Low absorbance in the mixture reaction indicated high antioxidant activity. The standard DPPH curve (absorbance vs. quercetin) was constructed ( $y = -1.6801 x + 0.9377$ ;  $R^2 = 0.9961$ ) with quercetin as reference and the results were expressed in mg quercetin equivalents per 100 g fresh weight (mg QE·100 g<sup>-1</sup> FW).. Percentage of DPPH reduction was calculated with the formula  $\% \text{ DPPH} = [(A_{\text{blank}} - A_{\text{sample}}) * 100] / A_{\text{blank}}$ , where:  $A_{\text{blank}}$  = absorbance of the blank (DPPH 0.1 mM);  $A_{\text{sample}}$  = absorbance obtained after 30 min.

### **Statistical analysis**

The study of the variables with the proximal analysis considered three replications (n = 3) and, for the phytochemical analyses, four replications (n = 4), for each xoconostle genotype. The results were expressed as mean ± standard error. The analysis of variance (ANOVA), Tukey test ( $P \leq 0.05$ ) and Pearson coefficient of correlation were calculated in SAS (2000).

## **RESULTS AND DISCUSSION**

### **Proximal analysis**

The proximal analysis enabled determination of the nutritional quality of the rind of fruits of different xoconostle genotypes as well as comparison with other *Opuntia* spp. fruits. The differences ( $P \leq 0.05$ ) among the 15 genotypes analyzed were significant in contents of ash, crude protein, lipids and carbohydrate (Table 2).

The xoconostle rind of the different genotypes had higher contents of moisture (89.94 - 93.84 %) than those reported in other studies. Contreras *et al.* (2011) found lower moisture content

**Table 2.** Nutrient composition of the rind of fruits of 15 xocostle genotypes (*Opuntia* spp.).

Num.	Species	Genotype	Moisture (%)	Ash (%)	Crude fiber (%)	Protein (%)	Lípidos (%)	Carbohydrates (%)
1	<i>O. mattudae</i> Scheinvar	Cuaresmeño blanco	91.92 ± 0.04cde	0.82 ± 0.00gh	1.04 ± 0.08a	0.29 ± 0.00bc	0.32 ± 0.00a	5.62 ± 0.04de
2	<i>O. mattudae</i> Scheinvar	Cuaresmeño zacatecano	92.60 ± 0.09bc	0.89 ± 0.01fg	0.54 ± 0.01bc	0.21 ± 0.00efg	0.13 ± 0.00cdef	5.65 ± 0.07de
3	<i>O. mattudae</i> Scheinvar	Rojo saínero	91.16 ± 0.10ef	1.06 ± 0.02e	0.81 ± 0.00ab	0.39 ± 0.01a	0.16 ± 0.00cd	6.44 ± 0.07bc
4	<i>O. mattudae</i> Scheinvar	Cuaresmeño rojo	93.12 ± 0.18ab	0.67 ± 0.02ij	0.57 ± 0.02bc	0.20 ± 0.01fg	0.13 ± 0.00cdef	5.32 ± 0.12ef
5	<i>O. mattudae</i> Scheinvar	Cuerón	91.29 ± 0.04def	1.41 ± 0.00a	0.53 ± 0.03bc	0.16 ± 0.00gh	0.08 ± 0.00f	6.55 ± 0.09abc
6	<i>O. mattudae</i> Scheinvar	El aguacero	93.10 ± 0.08ab	0.92 ± 0.01fg	0.62 ± 0.00abc	0.19 ± 0.00gh	0.10 ± 0.00def	5.08 ± 0.07ef
7	<i>O. duranguensis</i> Britton & Rose	Cambray	89.94 ± 0.05g	1.29 ± 0.00bc	1.03 ± 0.00a	0.34 ± 0.01b	0.17 ± 0.00c	7.25 ± 0.05a
8	<i>O. duranguensis</i> Britton & Rose x <i>O. jocosostle</i> F.A.C. Weber	Blanco jaspeado	93.84 ± 0.12a	0.57 ± 0.00j	0.31 ± 0.00c	0.17 ± 0.00gh	0.09 ± 0.00ef	5.02 ± 0.10ef
9	<i>O. jocosostle</i> Weber	Rosita	93.39 ± 0.20a	0.72 ± 0.03hi	0.45 ± 0.00bc	0.28 ± 0.00cd	0.18 ± 0.02c	4.99 ± 0.13ef
10	<i>O. sainaltense</i> Scheinvar	Chaveñito	91.31 ± 0.21def	0.94 ± 0.02f	0.66 ± 0.04abc	0.26 ± 0.00cd	0.17 ± 0.00c	6.68 ± 0.14abc
11	<i>O. jocosostle</i> Weber	Chatito	93.80 ± 0.07a	0.69 ± 0.00i	0.49 ± 0.00bc	0.14 ± 0.00h	0.25 ± 0.00b	4.64 ± 0.08f
12	<i>O. tezontepecana</i> Gallegos & Scheinvar	Invierno	91.55 ± 0.04de	1.22 ± 0.00cd	0.72 ± 0.20abc	0.29 ± 0.00c	0.12 ± 0.00cdef	6.12 ± 0.24cd
13	<i>O. leucotricha</i> Salm-Dick x <i>O. jocosostle</i> Weber	Café	92.01 ± 0.02bcd	1.38 ± 0.00ab	0.71 ± 0.02abc	0.25 ± 0.00cde	0.13 ± 0.00cdef	5.53 ± 0.00de
14	<i>O. leiascheinvariana</i> Martínez & Gallegos	Matizado	90.98 ± 0.05fg	1.16 ± 0.00de	0.69 ± 0.01abc	0.25 ± 0.00cde	0.16 ± 0.00cd	7.08 ± 0.04ab
15	<i>O. oligacantha</i> Förster	Borrego	91.85 ± 0.06cde	0.94 ± 0.00f	0.42 ± 0.02bc	0.24 ± 0.01def	0.15 ± 0.00cde	6.41 ± 0.01bc

Values are means ± standard error of three replications per genotype. Means with the same letters in a column are not significantly different (Tukey, *P* < 0.05).

(87.30 - 89.05 %) in *O. joconostle*. However, Morales et al. (2012) reported similar values for *O. joconostle* and *O. matudae* (93.24 and 94.11 %, respectively) and in different varieties of cactus pear (84.70 - 87.10 %) (Esquivel, 2004; El-Samahy et al., 2009). The content of crude fiber in the xoconostle fruits was similar to reports on several varieties of cactus pear (0.18 %) (El-Samahy et al., 2009). It is important to highlight that the consumption of fiber decreases blood cholesterol and sugar levels, although how the mechanism works is still unknown (Duarte-Martino et al., 2012).

The range in protein contents (0.39 – 0.14 %) found in the different xoconostle genotypes was lower than that reported by Contreras et al. (2011) and Morales et al. (2012) (0.66-1.56 %) in another species (*Opuntia matudae*; 0.56 %) (Morales et al., 2012) and in some varieties of cactus pear (0.67 %) (El-Samahy et al., 2009). The nutritional quality of the proteins in a food may be due to genetic factors (Duarte-Martino et al., 2012). However, our study did not determine the amino acid profile or the nutritive characteristic of the proteins, which we suggest should be studied in the xoconostle fruit.

Contreras et al. (2011) reported less than 0.1 % lipid content of *Opuntia joconostle*. In the xoconostle genotypes studied lipids were found in the range of 0.18 to 0.25 %, but the genotype Cuaresmeño Blanco had the highest percentage (0.32 %), similar to that reported for other commercial fruits (Morillas-Ruiz and Delgado-Alarcón, 2012).

Carbohydrate content (4.64 to 7.25 %) in the xoconostle genotypes was higher than that found in the species *O. matudae* and *O. joconostle* (3.93 and 3.69 %, respectively) (Morales et al., 2012). In contrast, the levels of these metabolites in papaya, orange and mango were 10 to 16 % higher than those found in the xoconostles studied, justifying their use in the treatment of diabetes in the traditional medicine of Mexico (Pimienta-Barrios et al., 2008). Consumption of the fruits of some xoconostle genotypes can provide benefits to the consumer for their nutritional quality when eaten together with other fruits and vegetables.

### **Total betalain content**

In this study, the differences in total betalain content among the xoconostle genotypes were significant ( $P < 0.05$ ) (Table 3). These pigments were found in the range of 1.71 to 35.06 mg 100 g<sup>-1</sup> f.w. Castellanos-Santiago and Yahia (2008) reported higher levels (17 - 815 mg 100 g<sup>-1</sup> d.w.) in ten Mexican varieties of cactus pear than those observed in the 15 genotypes of xoconostle of our study (21.19 - 348.13 mg 100 g<sup>-1</sup> d.w.).

Betalains derive biosynthetically from betalamic acid and group in betacyanins and betaxanthins. Betacyanins are red-purple and the betaxanthins are responsible for the orange-yellow color of the pulp and rind of these fruits (Zrýd and Christinet, 2004; Stintzing and Carle, 2007). The different coloring is due to the variability in betaxanthin and betacyanin contents in the genotypes studied, as in other fruits (Guzmán-Maldonado et al., 2010; Azeredo, 2009).

**Table 3.** Content of pigments (betalains, betacyanins and betaxanthins) in the fruit rind of 15 genotypes of xocostle (*Opuntia* spp.).

Num.	Species	Genotype	Betalains (mg 100 g <sup>-1</sup> f.w.)	Betacyanins <sup>b</sup> (mg 100 g <sup>-1</sup> f.w.)	Betaxanthins (mg 100 g <sup>-1</sup> f.w.)
1	<i>O. matudae</i> Scheinvar	Cuaresmeño blanco	1.71 ± 0.06 k	0.80 ± 0.03 k	0.91 ± 0.03 l
2	<i>O. matudae</i> Scheinvar	Cuaresmeño zacatecano	3.89 ± 0.09 hi	2.05 ± 0.04 i	1.84 ± 0.05 i
3	<i>O. matudae</i> Scheinvar	Rojo saimero	11.60 ± 0.15 d	7.28 ± 0.12 d	4.32 ± 0.03 d
4	<i>O. matudae</i> Scheinvar	Cuaresmeño rojo	5.74 ± 0.08 g	3.59 ± 0.05 g	2.16 ± 0.04 h
5	<i>O. matudae</i> Scheinvar	Cuerón	4.23 ± 0.08 h	2.56 ± 0.05 h	1.67 ± 0.04 j
6	<i>O. matudae</i> Scheinvar	El aguacero	3.51 ± 0.09 i	2.25 ± 0.06 hi	1.26 ± 0.03 k
7	<i>O. duranguensis</i> Britton & Rose	Cambray	35.06 ± 0.11 a	26.05 ± 0.06 a	9.01 ± 0.06 a
8	<i>O. duranguensis</i> Britton & Rose x <i>O. joconostle</i> F.A.C. Weber	Blanco jaspeado	±	±	±
9	<i>O. joconostle</i> Weber	Rosita	2.21 ± 0.08 j	1.04 ± 0.04 k	1.17 ± 0.04 k
10	<i>O. sainaltense</i> Scheinvar	Chaveñito	7.09 ± 0.09 f	4.70 ± 0.07 f	2.38 ± 0.03 g
11	<i>O. joconostle</i> Weber	Chatito	12.31 ± 0.06 c	7.22 ± 0.04 d	5.10 ± 0.02 c
12	<i>O. tezontepicana</i> Gallegos & Scheinvar	Invierno	2.39 ± 0.02 j	1.42 ± 0.01 j	0.97 ± 0.01 l
13	<i>O. leucotricha</i> Salm-Dick x <i>O. joconostle</i> Weber	Café	16.71 ± 0.13 b	11.14 ± 0.09 b	5.57 ± 0.05 b
14	<i>O. leiascheinvariana</i> Martínez & Gallegos	Matizado	8.92 ± 0.10 e	4.67 ± 0.09 f	4.25 ± 0.01 d
15	<i>O. oligacantha</i> Förster	Borrego	8.70 ± 0.04 e	5.92 ± 0.02 e	2.78 ± 0.02 f
			12.34 ± 0.16 c	8.67 ± 0.13 c	3.67 ± 0.03 e

<sup>a</sup>mg betalains in 100 g<sup>-1</sup> fresh weight; <sup>b</sup>mg betacyanins in 100 g<sup>-1</sup> fresh weight; <sup>c</sup>mg betaxanthins in 100 g<sup>-1</sup> fresh weight. Values are means ± standard error of four replications. Means with the same letters in a column are not significantly different (Tukey, *P* = 0.05).



The genotype *O. duranguensis* (Cambray) had the highest values for betacyanins and betaxanthins, while *O. matudae* (Cuaresmeño Blanco) had the lowest values of the two pigment types. Because of their high levels and stability in aqueous systems, the Cambray genotype could be a source of these pigments for use in agro- and pharmaceutical industries (Castellar et al., 2003).

Osorio-Esquivel et al. (2011) found betacyanins and betaxanthins in *O. joconostle*. They found the highest content of these pigments in the fruit endocarp (23.03 mg 100 g<sup>-1</sup> f.w.), followed by the mesocarp (7.25 mg 100 g<sup>-1</sup> f.w.) and pericarp (4.56 mg 100 g<sup>-1</sup> f.w.). Different methods of extraction could be the reason that concentrations of these pigments in *O. joconostle* differ from those found in the xoconostle genotypes of our study and from those reported for other *Opuntia* spp. fruits. The betaxanthin and betacyanin contents (3.0-18.9 mg 100 g<sup>-1</sup> and 0.16-30.0 mg 100 g<sup>-1</sup>, respectively) found in nine varieties of *Opuntia* sp. (Chávez-Santoscoy et al., 2009) are similar to those found in our study.

Betacyanin dry weight contents (data not shown) found in the studied xoconostle genotypes (9.90 - 258.68 mg 100 g<sup>-1</sup> d.w.) were similar to those of red pitaya and orange pitaya (37.6 and 199.6 mg 100 g<sup>-1</sup> d.w., respectively). In contrast, the betaxanthin content observed in xoconostle (11.23 - 89.45 mg 100 g<sup>-1</sup> d.w. (data not shown) were notably lower than those reported for red and orange pitaya (147.61 and 177.37 mg 100 g<sup>-1</sup> d.w., respectively) (García-Cruz et al., 2013), associated to the intense coloring of the latter fruits compared with xoconostle. However, Stintzing and Carle (2007) point out that the pigments identified in *Opuntia* fruits are stable, while the betacyanins in pitaya tend to decompose rapidly when they are isolated from the fruit. Nevertheless, the rate of degradation of the pigments in xoconostle is unknown.

#### **Total phenolic compound contents**

Total phenolic compound contents of *O. matudae* (Rojo Sainero) and *O. sainaltense* (Chaveñito) were significantly different ( $P < 0.05$ ) from those of the rest of the genotypes (Table 4).

The range of total phenolic compounds found in the 15 xoconostle genotypes (132.84 – 231.37 mg GAE 100 g<sup>-1</sup> f.w.) was higher than that found in the mesocarp of *O. joconostle* fruits. However, the pericarp of the xoconostles had higher concentrations of these metabolites (207.0 mg 100 g<sup>-1</sup> f.w.) (Osorio-Esquivel et al., 2011). These authors point out that the degree of ripeness at harvest, the genetic differences and the environmental conditions, among other factors, affect the content of phenolic compounds in the fruits. These factors could explain the variation among the 15 genotypes.

In contrast, the total phenol values reported in different varieties of cactus pear (106.6 - 130.0 mg GAE 100 g<sup>-1</sup> f.w.) (Yahia and Mondragón-Jacobo, 2011) and red pitaya (106.0 mg GAE 100 g<sup>-1</sup> f.w.) (García-Cruz et al., 2013) were lower than those found in xoconostle. The differences observed among species and varieties of different genera may be due to 1) genetic factors (Osorio-Esquivel et al., 2011) and 2) harvest and handling of fresh fruits that

**Table 4.** Content of antioxidant components (total phenols, flavonoids and vitamin C) in the fruit rind of 15 genotypes of xocostle (*Opuntia* spp.).

Num.	Species	Genotype	Total phenols (mg GAE 100 g <sup>-1</sup> f.w.) <sup>a</sup>	Flavonoids (mg QE 100 g <sup>-1</sup> f.w.) <sup>b</sup>	Ascorbic acid (mg AA 100 g <sup>-1</sup> f.w.) <sup>c</sup>
1	<i>O. matudae</i> Scheinvar	Cuaresmeño blanco	205.03 ± 2.90	bc 3.58 ± 0.10	de 13.46 ± 0.05
2	<i>O. matudae</i> Scheinvar	Cuaresmeño zacatecano	141.53 ± 2.67	hg 3.13 ± 0.19	ef 9.27 ± 0.05
3	<i>O. matudae</i> Scheinvar	Rojo sainero	224.32 ± 3.19	a 4.24 ± 0.11	bc 10.37 ± 0.03
4	<i>O. matudae</i> Scheinvar	Cuaresmeño rojo	206.14 ± 2.78	b 3.94 ± 0.11	cd 11.19 ± 0.03
5	<i>O. matudae</i> Scheinvar	Cuerón	199.47 ± 3.15	bc 2.18 ± 0.00	gh 10.13 ± 0.02
6	<i>O. matudae</i> Scheinvar	El aguacero	162.39 ± 4.99	ef 2.30 ± 0.09	gh 6.83 ± 0.01
7	<i>O. duranguensis</i> Britton & Rose	Cambray	176.86 ± 3.15	de 1.98 ± 0.00	h 11.50 ± 0.02
8	<i>O. duranguensis</i> Britton & Rose x <i>O. jocosostle</i> F.A.C. Weber	Blanco jaspeado	189.04 ± 1.93	cd 2.77 ± 0.16	fg 4.40 ± 0.02
9	<i>O. jocosostle</i> Weber	Rosita	151.20 ± 2.39	fg 2.93 ± 0.08	bc 8.59 ± 0.05
10	<i>O. sainaltense</i> Scheinvar	Chaveñito	231.37 ± 3.14	a 3.59 ± 0.19	de 9.56 ± 0.12
11	<i>O. jocosostle</i> Weber	Chatito	161.20 ± 4.47	ef 2.23 ± 0.14	gh 6.80 ± 0.01
12	<i>O. tezontepicana</i> Gallegos & Scheinvar	Invierno	140.48 ± 3.05	hg 3.63 ± 0.11	de 11.66 ± 0.06
13	<i>O. leucotricha</i> Salm-Dick x <i>O. jocosostle</i> Weber	Café	132.83 ± 2.88	h 2.26 ± 0.10	gh 9.62 ± 0.02
14	<i>O. leiascheinvariana</i> Martínez & Gallegos	Matizado	190.10 ± 3.37	bcd 4.97 ± 0.12	a 10.18 ± 0.04
15	<i>O. oligacantha</i> Förster	Borrego	196.62 ± 2.94	bc 4.77 ± 0.10	ab 9.56 ± 0.02

<sup>a</sup> mg gallic acid equivalents 100 g<sup>-1</sup> fresh weight; <sup>b</sup> mg quercetin equivalents 100 g<sup>-1</sup> fresh weight; <sup>c</sup> mg ascorbic acid equivalents 100 g<sup>-1</sup> fresh weight; Values are means ± standard error of four replications. Means with the same letters in a column are not significantly different (Tukey, *P* = 0.05).

can cause stress, which alters physiology and stimulates responses that cause phenolic compounds to accumulate (Pirovani et al., 2009).

Flavonoids have been studied little in *Opuntia*. The differences in flavonoid content (1.98 – 4.97 mg QE 100 g<sup>-1</sup> f.w.) found among the genotypes (Table 4) were significant ( $P < 0.05$ ). In all the genotypes, flavonoid concentrations were lower than that of their total phenolic compounds; this result may be due to enzymatic degradation (Jiménez and García-Carmona, 1999), or because some flavonoids are proanthocyanidins (condensed tannins), as occurs in other fruits (Cui et al., 2006).

#### **Flavonoid content**

The 15 xoconostle genotypes had a lower flavonoid content (1.98 a 4.97 mg QE 100 g<sup>-1</sup> f.w.) than that reported in cactus pears (1.34 and 27.73 mg QE 100 g<sup>-1</sup> f.w.) by Fernández-López et al. (2010), but similar to those found by Kuti (2004) in four cactus pear varieties (0.98 – 9.35 mg mg QE 100 g<sup>-1</sup> f.w.). The fruits of commercial white cactus pear (9.8 mg QE 100 g<sup>-1</sup> d.w.) and “manso” cactus pear (5.9 mg QE 100 g<sup>-1</sup> d.w.) had lower values (Guevara-Figueroa et al., 2010) than those found in xoconostle 19.68 - 58.57 mg QE 100 g<sup>-1</sup> de d.w.)(dry weight data not shown).

#### **Ascorbic acid content**

Differences among genotypes (Table 4) in ascorbic acid were significant ( $P < 0.05$ ). Of the 15 genotypes studied, Cuaresmeño Blanco (13.46 mg AA 100 g<sup>-1</sup> f.w.) had the highest content. Guzmán-Maldonado et al. (2010) and Morales et al. (2012) reported higher concentrations (31.8 mg AA 100 g<sup>-1</sup> f.w.) in Cuaresmeño Rojo. The observed differences may be due to several factors (geographic location, climate conditions, cultural practices, genotype, stage of maturity, postharvest handling, and method of analysis). However, the genotype is one of the most important factors for identification of the fruits with high vitamin C content (Latocha et al., 2011).

The ascorbic acid (AA) content in the studied xoconostles (4.41 and 13.46 mg AA 100 g<sup>-1</sup> f.w.) (Table 4) was similar to that reported for cactus pears with purple peel (1.0 a 11.1 mg AA 100 g<sup>-1</sup>) and with red peel (2.30 a 79.2 mg AA 100 g<sup>-1</sup>), Kuti (2004) considered these contents are high compared with other common fruits (peach, grapes and apple). Fernández-López et al. (2010) found higher values (14.5 a 23.3 mg 100 g<sup>-1</sup>) in species of red *Opuntia*.

#### **Antioxidant activity**

There were significant differences ( $P < 0.05$ ) in antioxidant activity among the genotypes (Table 5). It is important to highlight that some wild genotypes had higher antioxidant activity than other community or commercial genotypes (Table 1). Color was not a determining factor in identifying the varieties with higher antioxidant capacity, but a study to correlate xoconostle color with antioxidant activity is suggested. The Pearson correlation coefficient revealed positive correlation between antioxidant activity and betalain content (0.553) (Table 6), coinciding with Azeredo (2009) and Kuti (2004). However, antioxidant activity in some fruits is not associated only with betalains. Other metabolites, such as phenolic compounds, vitamin

**Table 5.** Antioxidant activity determined with the DPPH radical method in fruit rind of 15 genotypes of xoconostle (*Opuntia* spp.).

Num.	Species	Genotype	DPPH inhibited (%)	Antioxidant activity (mg QE 100 g <sup>-1</sup> f.w.)*
1	<i>O. matudae</i> Scheinvar	Cuaresmeño blanco	40.78 ± 0.05	214.37 ± 0.86 i
2	<i>O. matudae</i> Scheinvar	Cuaresmeño zacatecano	96.77 ± 0.03	313.49 ± 0.45 a
3	<i>O. matudae</i> Scheinvar	Rojo sainero	76.09 ± 0.11	276.88 ± 1.99 e
4	<i>O. matudae</i> Scheinvar	Cuaresmeño rojo	62.55 ± 0.63	252.91 ± 11.15 fg
5	<i>O. matudae</i> Scheinvar	Cuerón	76.83 ± 0.84	278.18 ± 14.90 e
6	<i>O. matudae</i> Scheinvar	El aguacero	96.77 ± 0.03	313.49 ± 0.45 a
7	<i>O. duranguensis</i> Britton & Rose	Cambray	83.79 ± 0.18	290.51 ± 3.07 d
8	<i>O. duranguensis</i> Britton & Rose x <i>O. joconostle</i> F.A.C. Weber	Blanco jaspeado	96.90 ± 0.03	313.71 ± 0.52 a
9	<i>O. joconostle</i> Weber	Rosita	62.45 ± 0.05	252.73 ± 0.90 g
10	<i>O. sainaltense</i> Scheinvar	Chaveñito	95.88 ± 0.17	311.91 ± 2.98 a
11	<i>O. joconostle</i> Weber	Chatito	89.05 ± 0.22	299.82 ± 3.77 c
12	<i>O. tezontepicana</i> Gallegos & Scheinvar	Invierno	49.26 ± 0.05	229.39 ± 0.86 h
13	<i>O. leuotricha</i> Salm-Dick x <i>O. joconostle</i> Weber	Café	93.78 ± 0.25	308.18 ± 4.49 b
14	<i>O. leiascheinvariana</i> Martínez & Gallegos	Matizado	90.07 ± 0.41	301.61 ± 7.17 c
15	<i>O. oligacantha</i> Förster	Borrogo	64.10 ± 0.13	255.65 ± 2.36 f

\*mg quercetin equivalents 100 g<sup>-1</sup> fresh weight; Values are means ± standard error of four replications. Means with the same letters in a column are not significantly different (Tukey, *P* 0.05).

C, other pigments (carotenes) and some sulfur compounds, as well as a synergic effect among the various antioxidant compounds present, are considered (Brat et al., 2005; Kuti, 2004) and may explain the higher antioxidant activity in the genotypes with higher contents of phenolic compounds, flavonoids and vitamin C.

**Table 6.** Pearson correlation coefficients between phytochemical compounds in fruit rind of 15 varieties of xoconostle (*Opuntia* spp.).

	<b>Total betalains</b>	<b>Phenols</b>	<b>Flavonoids</b>	<b>Vitamin C</b>	<b>Antioxidant activity</b>
Betalains	1.0000	0.0013	-0.0424	0.3913	0.5536*
Phenols		1.0000	0.4526	0.1994	0.2727
Flavonoids			1.0000	0.3349	0.0626
Vitamin C				1.0000	0.2263
Antioxidant activity					1.0000

\*  $P < 0.05$ : Statistically significant correlation at a confidence level of 95 %.

Finally, we recommend intensifying research on the nutrient and nutraceutical quality of other genotypes, aiming to increase their demand, to generate new spaces for commercialization, and to contribute to conservation of the country's cultural identity. The high content of stable pigments (betalains), important for the food industry, the presence of antioxidant compounds, and the low water requirements give advantages to the cultivation of *Opuntia* spp. fruits.

These advantages make them an option for agriculture in arid and semiarid regions of the country. Considering the contents of antioxidant components obtained in this study, the consumption of fruits of some xoconostle genotypes can provide health benefits, together with consumption of other fresh fruits.

## CONCLUSIONS

Moisture (89.94 - 93.84 %) and crude fiber (0.42 – 1.04 %) contents were similar to those found for varieties of cactus pear. The carbohydrate content (4.64 - 7.25 %) was lower than that documented in commercial fruits (papaya, orange and mango). Significant differences in betalain content, phenolic compounds, flavonoids and vitamin C were found among the 15 xoconostle genotypes. The betalain content (1.71 a 35.06 mg 100 g<sup>-1</sup>f.w.) was similar to that reported in pitaya and lower than that found in cactus pear varieties. Although, the Pearson correlation coefficient revealed a positive relationship between antioxidant activity and betalains; the high antioxidant activity observed may be a synergic effect due to the levels found of total betalains, phenolic compounds, flavonoids and vitamin C. The contents of these antioxidants may point to the selection of genotypes for agroindustrial and commercial use. The wild genotype Chaveñito (*O. sainaltense*) inhibited the DPPH radical (antioxidant activity) to a greater extent (95.88 %), giving it higher nutraceutical quality, the genotype Cambray (*O. duranguensis*) is outstanding for its nutritional quality.

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