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## Nutraceutical components, antioxidant activity, and color of 11 varieties of prickly pear (*Opuntia* sp.)

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### Summary

In Mexico, there are 50 recorded varieties of the prickly pear fruit. National production covers only the white-pulp fruit, but other red varieties have export potential; however, their nutraceutical properties are unknown. The pulp and peel (underutilized tissue) of the pigmented fruits of the genus *Opuntia* sp. are marketed on a limited basis. They represent an alternative source of stable pigments (betalains), which are associated with antioxidant properties, for the agroindustry. The objective was to assess the content of nutraceutical components, antioxidant activity, and peel and pulp color of 11 varieties of the prickly pear fruit that are marketed on a small scale. Statistical analysis revealed that Roja Villanueva peel had the highest betalain content (39.97 mg 100 g<sup>-1</sup> FW). Altea Blanca peel demonstrated the highest concentration of phenolic compounds (618.39 mg GAE 100 g<sup>-1</sup> FW), whereas Altea Roja had the highest ascorbic acid content (37.14 mg AAE 100 g<sup>-1</sup> FW). The greatest nutraceutical potential was observed in the pulp of the non-marketed Tzaponopal Rojo variety of the species *O. robusta* var. *larreyi*, due to the high antioxidant activity (0.0183 mg mL<sup>-1</sup>), as well as the darkest color (< hue value, 12.31) and the lowest lightness (< luminosity, 19.31), which coincides with the highest betalain concentration.

**Keywords:** antioxidant components, betalains, pulp, peel.

### Introduction

Mexico is considered the center of origin of the family *Cactaceae*, which includes approximately 1500 species of the genus *Opuntia* (ANOOP et al., 2012), both wild and cultivated (SÁENZ, 2006). The subgenera *Opuntia* and *Nopalea* are the main producers of the edible fruit known as prickly pear fruit, grouped by its pigmentation into purple, red, orange, yellow, and white fruit (SEGURA et al., 2007; CARUSO et al., 2010). Prickly pear grows in arid and desert regions. It has a photosynthetic metabolism of Crassulacean type called *crassulacean acid metabolism* (CAM), which allows the production of biomass in the arid and drought conditions characteristic of its habitat (ANDREU et al., 2017). During the photosynthetic process, CAM plants open their stomata during the desert nights and close them during the hot, dry days, thus efficiently using night moisture (TAIZ and ZEIGER, 2002).

In Mexico, white-pulp and green-peel prickly pears are highly appreciated for their market value. In 2014, prickly pear production in the country reached approximately 588 000 t (SIAP, 2014); however, despite the great production and variability of this fruit in the country, the levels of antioxidant components in most prickly pear species are unknown. Nonetheless, these components in various other fruits are known to provide important nutraceutical benefits to consumers (TSAO and AKHTAR, 2005; ANOOP et al., 2012). Consumption of these other fruits prevents some chronic-degenera-

tive diseases, due to the presence of certain metabolites (phenolic compounds, flavonoids, ascorbic acid, and betalains) (SUMAYA-MARTÍNEZ et al., 2011; ABOU-ELELLA and ALI, 2014). Biosynthetically, betalamic acid-derived betalains, which are pigments used in the agri-food industry as natural colorants (STRACK et al., 2003), also have antioxidant properties (LIVREA and TESORIERE, 2006; ALBANO et al., 2015). Moreover, color is part of fruit quality and may therefore be a factor in its preference, acceptance, or rejection, and play a decisive role in the failure or success of prickly pear marketing. The wide genetic diversity of the Mexican prickly pear justifies the study of nutraceutical content, particularly in pigmented prickly pear varieties. The objective of the present study was to assess the content of nutraceutical components, antioxidant activity, and pulp and peel color of 11 varieties of prickly pear marketed on a small scale, and to identify those with the greatest nutraceutical attributes. Such nutraceutical information will be of benefit to consumers.

### Material and methods

#### Plant material collection

Healthy and pest-free fruits were harvested in the Dr. Facundo Barrientos Pérez Experimental Station at the Universidad Autónoma Chapingo in Chapingo, State of Mexico (Tab. 1), located at 19° 29' N and 98° 53' W, at an altitude of 2240 m. The climate is C (Wo) (w) b (i') g, considered the driest of the subhumid climates, with summer rains and mild temperatures. The area has a mean annual temperature of 17.8 °C and rainfall of 644.8 mm per year.

The harvest criteria were the visual parameters used commercially in the producing region for each variety (completely characteristically developed fruit, with fruit weight ranging from 125 g to 230 g, depending on the variety) (CORRALES et al., 2006). After cutting, fruits were stored for 15 days. The pulp was then separated from the seed and peel, and the tissue (pulp and peel) was fractionated into small portions and frozen by direct immersion in liquid nitrogen. The samples were then stored at -20 ± 2 °C for later analysis.

#### Quantification of total betalains (betacyanins and betaxanthins)

Pigment content was determined according to the methodology described by CASTELLANOS-SANTIAGO and YAHIA (2008). The frozen sample (1 g) was mixed with 10 mL distilled water, subjected to sonication for 20 min, and centrifuged at 2200 g for 20 min. Absorbance of the extracts was measured in a Genesys 10S spectrophotometer (Thermo Scientific, Florida, USA) at 483 nm (betaxanthins) and 538 nm (betacyanins). In order to estimate the pigment concentration, the following equation was used:

betacyanins or betaxanthins (mg g<sup>-1</sup> FW) = (A × DF × MW × V) (ε × l × FW)<sup>-1</sup>,

where A = absorbance; DF = dilution factor; MW = molecular weight (betacyanin: 550 g mol<sup>-1</sup>, betaxanthin: 308 g mol<sup>-1</sup>); V = extract

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volume (mL);  $\epsilon$  = molar extinction coefficient (betacyanin: 60 000 L mol<sup>-1</sup> cm<sup>-1</sup>; betaxanthin: 48 000 L mol<sup>-1</sup> cm<sup>-1</sup>); FW = fresh sample weight (g); and  $l$  = cell length (1 cm). Total betalain content was expressed as mg per 100 g fresh weight (FW).

#### Extraction of total phenolic compounds and flavonoids

The sample (1 g) was dissolved in 25 mL ethanol (95% v/v) in an ultrasonic bath (Cole-Parmer 8892, Illinois, USA) for 15 min. After 24 h, the volume was brought to 25 mL with ethanol (80% v/v) and centrifuged at 1409 g. This extract was subsequently used to determine the content of total phenolic compounds and flavonoids.

#### Quantification of total phenolic compounds

The method proposed by WATERMAN and MOLE (1994) was followed. First, 10 mL Na<sub>2</sub>CO<sub>3</sub> (10% w/v) were added to 0.5 mL ethanolic extract, and the mixture was then incubated at 38 °C for 15 min. Subsequently, 3 mL water and 1 mL Folin-Ciocalteu reagent:water (1:1 v/v) were added to 1 mL of this mixture, which was left to stand for 15 min in the dark, and absorbance was then measured at 760 nm in a Genesys 10S spectrophotometer. Total phenolic compound content in the extract was expressed as mg gallic acid equivalents per 100 g fresh weight (mg GAE 100 g<sup>-1</sup> FW).

#### Quantification of flavonoids

Flavonoid content was quantified according to the method proposed by CHANG et al. (2002). First, 1.5 mL ethanol (95% v/v), 0.1 mL AlCl<sub>3</sub> (10% w/v), 0.1 mL 1 M potassium acetate, and 2.8 mL distilled water were added to 0.5 mL ethanolic extract. The mixture remained at room temperature for 30 min, and absorbance was measured in a Genesys 10S spectrophotometer at 415 nm. Results were expressed as mg quercetin equivalents per 100 g fresh weight (mg QE 100 g<sup>-1</sup> FW).

#### Quantification of ascorbic acid

The sample (1 g) was homogenized in 3 mL metaphosphoric acid (3% v/v) for 3 min. The mixture was filtered, and 1 mL supernatant was brought to 10 mL with metaphosphoric acid (3% v/v). Then 2 mL pH 4 buffer solution (glacial acetic acid:sodium acetate [5% w/v, 1:1]), 3 mL dichloroindophenol, and 15 mL xylene were added to 2 mL of

the mixture, which was shaken vigorously in a vortex (Thermolyne Type 6700, USA). Absorbance was measured in a spectrophotometer at 520 nm. Ascorbic acid was calculated with the following equation:

$$\text{mg total ascorbic acid} = (C \times V \times 100) / (A \times W),$$

where C = ascorbic acid in the sample; V = capacity volume; A = aliquot of the solution taken (mL); and W = sample weight or volume. Results were expressed as mg ascorbic acid equivalents per 100 g fresh weight (mg AAE 100 g<sup>-1</sup> FW).

#### Antioxidant activity assessment

The analysis was performed using the DPPH (2, 2-diphenyl-1-picrylhydrazyl, Sigma-Aldrich) free radical method described by AMICO et al. (2008). For analysis, 10 g of sample were macerated in methanol and subjected to sonication for 20 min. The mixture was filtered, and the supernatant was concentrated in a Büchi R-210 rotary evaporator (Flawil, Switzerland). From the methanol extract, the following solutions were prepared in methanol by serial dilution to obtain 0.2, 0.15, 0.1, and 0.05 mg mL<sup>-1</sup> concentrations. As references, 0.1, 0.001, and 0.0001 mg mL<sup>-1</sup> concentrations of quercetin and ascorbic acid were individually prepared. Next, 3 mL DPPH solution (0.1 mM) were added to 1 mL of each concentration of extracts and the individual references. Mixtures were left at room temperature for 30 min, and then absorbance readings were taken at 516 nm. DPPH percentage was determined by the following formula:

$$\% \text{ DPPH} = (A_{\text{blank}} - A_{\text{sample}}) \times 100 / A_{\text{blank}},$$

where A<sub>blank</sub> is the control absorbance (DPPH 0.1 mM) and A<sub>sample</sub> is the absorbance obtained for each sample after 30 min with DPPH 0.1 mM.

The antioxidant activity of the samples was determined by calculating the mean inhibitory concentration (IC<sub>50</sub>), which is the concentration required by the sample to decrease DPPH absorbance to 50%. Low absorbance of the reaction mixture indicated high antioxidant activity. The standard curve was built with 3.93 mg DPPH dissolved in 100 mL methanol to obtain a 0.1 mM concentration (stock solution). The following concentrations were prepared from this solution: 0.01, 0.02, 0.04, 0.06, 0.08, and 0.1 mM DPPH. Absorbance was measured at 516 nm in a Genesys 10S spectrophotometer, and readings were taken in triplicate; methanol was used as a blank.

**Tab. 1:** Descriptions of 11 varieties and species of prickly pear (*Opuntia* sp.) grown at the Universidad Autónoma Chapingo Experimental Station, Chapingo, Mexico.

Species	Variety	Origin	Color	Market value*
<i>Opuntia</i> sp.	Alteña Blanca	Celaya, Gto.	Yellow-greenish	LMP
<i>Opuntia</i> sp.	Alteña Roja	Celaya, Gto.	Orange	AMP
<i>O. ficus-indica</i> (L.). Mill	Huatusco	Huatusco, Ver.	Yellow	LMP
<i>O. ficus-indica</i> (L.). Mill	Solferino	Chapingo, Méx.	Orange	AMP
<i>O. ficus-indica</i> (L.). Mill	Roja Villanueva	Villanueva, Pue.	Orange	AMP
<i>O. ficus-indica</i> (L.). Mill	Jade	Desconocido	Pink	LMP
<i>O. ficus-indica</i> (L.). Mill	Copena CEII	Zacatecas, Zac.	Pink	AMP
<i>O. ficus-indica</i> (L.). Mill	Copena VI	Chapingo, Méx.	Red - purple	LMP
<i>O. megacantha</i> Salm	Morada	San Martín de las Pirámides, Méx.	Red	AMPP
<i>O. megacantha</i> Salm	Plátano	Ojuelos, Jal.	Yellow	LMP
<i>O. robusta</i> var. <i>Larreyi</i>	Tzaponopal Rojo	San Martín de las Pirámides, Méx.	Red	AMP

\*LMP: low market potential; AMP: average market potential; AMPP: average market potential as a source of pigments (CORRALES-GARCÍA and HERNÁNDEZ-SILVA, 2005).

### Measurement of color parameters

Pulp and peel color were determined by assessing lightness (*L*), tone angle (*hue*) and purity of color or chromaticity index (*chroma*) with a HunterLab colorimeter (MiniScan XE Plus 45/ 0-L, Reston, Virginia, USA). Readings from *a*\* and *b*\* were obtained to clearly quantify color differences among the tissues and varieties. Variables were calculated with the following equations:

$$\text{hue} = \tan^{-1}(a/b) \text{ and } \text{chroma} = (a^2 + b^2)^{1/2} \text{ (MC GUIRE, 1992).}$$

### Statistical analysis

An asymmetric 11 × 2 factorial design, in which the pulp or peel of each variety was considered as a treatment, was used, with four replications. Results were analyzed using an analysis of variance and Tukey's range test ( $P \leq 0.05$ ). The least significant difference (LSD) ( $\alpha = 0.05$ ) and principal component analysis were obtained with the SAS (Statistical Analysis System 9.0).

## Results and discussion

### Total betalain content (betacyanins and betaxanthins)

Total betalain concentration was higher in red prickly pear fruits than in white and orange ones, which showed lower concentrations in pulp and peel (Tab. 2). The total betalain levels (92.08 mg 100 g<sup>-1</sup>

FW) in the pulp of the Tzaponopal Rojo variety surpassed those of the other varieties. KUTI (2005) reported a betalain concentration in *Opuntia* spp. fruits similar to that found in the present research (81.5 mg 100 g<sup>-1</sup> FW). Betalains are responsible for the array of fruit colors in the many species and varieties of the genus *Opuntia* (STINTZING and CARLE, 2007). These metabolites derive biosynthetically from betalamic acid and are categorized as either betacyanins or betaxanthins. Betacyanins are red-purple, and betaxanthins are responsible for the orange-yellow color of the pulp and rind of these fruits (ZRYD and CHRISTINET, 2004; STINTZING and CARLE, 2007).

The Alteña Blanca variety (yellow-green color) had the lowest content of betacyanins (responsible for red-purple color) and betaxanthins (responsible for yellow-orange color) in both pulp and peel; by contrast, the highest betacyanin content was found in the pulp of the Tzaponopal Rojo (red), Copena VI (red-purple), and Jade (pink) varieties. The highest betaxanthin content was found in the peel of Roja Villanueva and the pulp of Tzaponopal Rojo. These results support the view that the high content of pigments in the pulp and peel of these varieties might make them good sources of natural pigments for use in the food industry. Values for both pigments found in the present study (0.46 mg -49 mg 100 g<sup>-1</sup> FW for betacyanins and 0.5 mg -19.91 mg 100 g<sup>-1</sup> FW for betaxanthins) were superior to those found by LÓPEZ et al. (2015) for the Cambrey genotype "xocostle" (sour prickly pear fruit), which had values of 26.05 mg 100 g<sup>-1</sup> FW

**Tab. 2:** Content of total betalains, betacyanins, and betaxanthins in the fruit of 11 varieties and species of prickly pear (*Opuntia* sp.) harvested at the Universidad Autónoma Chapingo Experimental Station, Chapingo, Mexico.

Species	Variety	Tissue*	Total betalains (mg 100 g <sup>-1</sup> FW)	Betacyanins (mg 100 g <sup>-1</sup> FW)	Betaxanthins (mg 100 g <sup>-1</sup> FW)
<i>Opuntia</i> sp.	Alteña Blanca	Peel	0.99k	0.50g	0.50h
		Pulp	1.03k	0.46g	0.57h
<i>Opuntia</i> sp.	Alteña Roja	Peel	19.20hj	12.46ed	6.74dg
		Pulp	23.08gi	14.36cd	8.73cf
<i>O. ficus indica</i>	Huatusco	Peel	4.25jk	2.24eg	2.01gh
		Pulp	4.34jk	1.81fg	2.53gh
<i>O. ficus indica</i>	Solferino	Peel	22.98gi	15.24cd	7.74dg
		Pulp	19.08hj	11.35df	7.74dg
<i>O. ficus indica</i>	Roja Villanueva	Peel	39.97cg	23.97bc	16.01ab
		Pulp	35.30be	20.62bd	14.67ac
<i>O. ficus indica</i>	Jade	Peel	21.21ih	14.89cd	6.42eh
		Pulp	41.91bc	30.13b	11.79be
<i>O. ficus indica</i>	Copena CEII	Peel	31.57eh	21.78bc	9.79cf
		Pulp	40.36bd	29.04b	11.32be
<i>O. ficus indica</i>	Copena VI	Peel	38.85dg	27.14b	11.71be
		Pulp	43.73b	30.91b	12.82bd
<i>O. megacantha</i>	Morada	Peel	17.03hk	12.67ed	4.36fh
		Pulp	27.86fi	20.37bd	7.49dg
<i>O. megacantha</i>	Plátano	Peel	14.02ik	2.19efg	11.83be
		Pulp	11.48ik	1.54fg	9.95bf
<i>O. robusta/Larreyi</i>	Tzaponopal Rojo	Peel	33.41eh	23.50bc	9.90bf
		Pulp	69.06a	49.15a	19.91a
CV			21.62	23.89	26.27

\*Fresh weight; CV = Coefficient of variation. Means with the same letters in a column are not significantly different (Tukey,  $P \leq 0.05$ ).

for betacyanins and 9.01 mg 100 g<sup>-1</sup> FW for betaxanthins. KHATABI et al. (2016) demonstrated that differences in betalain content in the juice and pulp of prickly pear fruits could be attributed to variability in prickly pear ecotypes, physiologies, and growth conditions.

Pigments identified in prickly pear (*Opuntia*) fruits are stable, while the betacyanins in pitaya (*Stenocereus pruinosus*) tend to decompose rapidly when they are isolated from the fruit (STINTZING and CARLE, 2007). Nevertheless, the rate of degradation of the pigments in *Opuntia* and *Stenocereus* fruits is unknown. These pigments exhibit important antioxidant activity and are non-toxic to humans (SUMAYA-MARTÍNEZ et al., 2011). Moreover, high levels of betalains help prevent cancer and lipid oxidation of membranes (LIVREA and TESORIERE, 2006). Additionally, fruit quality is based partly on color, which may be a factor in its preference, acceptance, or rejection, thus playing a decisive role in the failure or success of prickly pear marketing.

### Total phenolic compound content

Phenolic compounds are another group of secondary metabolites, identified in some fruits of the *Opuntia* genus (PIMIENTA-BARRIOS et al., 2008; OSORIO-ESQUIVEL et al., 2011; ANDREU et al., 2017; MENA et al., 2018), that protects plants from oxidative stress, and their consumption contributes to preventing disease in humans. It is important to point out that in every variety, phenolic compound levels were much higher in the peel than in the pulp; this is consistent

with the work of MOUSSA-AYOUB et al. (2011), who observed similar behavior in *O. macrorhiza* fruits. Alteña Blanca peel had the highest concentration of these metabolites (618.39 mg GAE 100 g<sup>-1</sup> FW); on the other hand, Copena CEII pulp had the lowest concentration (106.60 mg GAE 100 g<sup>-1</sup> FW) (Tab. 3). Results were superior to those obtained by LÓPEZ et al. (2015) for 15 xoconostle cultivars whose phenolic compound contents ranged between 132.83 mg and 231.37 mg GAE 100 g<sup>-1</sup> FW.

YAHIA and MONDRAGÓN-JACOBO (2011) also reported total phenol values in different varieties of prickly pear (106.6 mg-130.0 mg GAE 100 g<sup>-1</sup> FW), and concentrations of phenolic compounds measured by GARCÍA-CRUZ et al. (2013) in red pitaya (106.0 mg GAE 100 g<sup>-1</sup> FW) were lower than those found in prickly pear fruits. The differences observed among species and varieties of different genera may be due to (a) genetic factors (MOUSSA-AYOUB et al., 2014; OSORIO-ESQUIVEL et al., 2011), and (b) stress caused by harvest and handling of fresh fruits, which alters physiology and stimulates responses that cause phenolic compounds to accumulate (PIROVANI et al., 2009). The effect of genotypic differences on the phenolic profiles of prickly pear fruits has also been investigated (MOUSSA-AYOUB et al., 2014; STINTZING et al., 2005). However, several studies point out that the concentration of phenolic compounds in prickly pear depends on the environmental conditions, as well as the part of the cactus plant under consideration (KHATABI et al., 2016; MOUSSA-AYOUB et al., 2014; STINTZING et al., 2005).

There was no relationship between the content of these phyto-

**Tab. 3:** Content of total phenolic compounds, flavonoids, and ascorbic acid in the fruit of 11 varieties and species of prickly pear (*Opuntia* sp.) harvested at the Universidad Autónoma Chapingo Experimental Station, Chapingo, Mexico.

Species	Variety	Tissue*	Phenolic compounds (mg GAE 100 g <sup>-1</sup> FW)	Flavonoids (mg QE 100 g <sup>-1</sup> FW)	Ascorbic acid (mg AAE 100 <sup>-1</sup> g FW)
<i>Opuntia</i> sp.	Alteña Blanca	Peel	618.39 <sup>a</sup>	27.73a	2.26de
		Pulp	165.56ef	1.34f	3.74ce
<i>Opuntia</i> sp.	Alteña Roja	Peel	486.79b	23.88ab	2.26de
		Pulp	120.75f	4.37df	37.14a
<i>O. ficus indica</i>	Huatusco	Peel	425.58bc	20.13ab	0.56e
		Pulp	131.96ef	2.60ef	0e
<i>O. ficus indica</i>	Solferino	Peel	404.59bc	22.41ab	6.80cd
		Pulp	148.6ef	3.23df	4.62ce
<i>O. ficus indica</i>	Roja Villanueva	Peel	242.98ed	20.05ab	0e
		Pulp	118.39f	9.60ce	0e
<i>O. ficus indica</i>	Jade	Peel	374.88bc	20.05ab	14.85b
		Pulp	122.52f	6.69df	2.80ce
<i>O. ficus indica</i>	Copena CEII	Peel	322.99cd	20.74ab	0e
		Pulp	106.60f	8.34df	0e
<i>O. ficus indica</i>	Copena VI	Peel	425.59bc	23.96ab	16.59b
		Pulp	143.74ef	8.23df	8.04c
<i>O. megacantha</i>	Morada	Peel	389.03bc	17.76bc	0e
		Pulp	107.19f	6.00df	0e
<i>O. megacantha</i>	Plátano	Peel	400.82bc	21.43ab	15.83b
		Pulp	111.90f	6.19df	15.49b
<i>O. robusta/Larrey</i>	Tzaponopal Rojo	Peel	219.22ef	21.79ab	0e
		Pulp	161.43ef	11.21cd	0e
CV			16.59	22.1	34.88

\*Fresh weight; CV = Coefficient of variation. Means with the same letters in a column are not significantly different (Tukey,  $P \leq 0.05$ ).

chemicals and the pulp and peel colors of the different varieties studied; this behavior was also observed by SUMAYA-MARTÍNEZ et al. (2011), who point out that when investigating the fruit colors of three groups of prickly pear cultivars (purple, yellow, and white), no relationship with total phenol content was found. However, KHATABI et al. (2016) reported that the red prickly pear contains higher amounts of polyphenols than those of the yellow variety. Assessment of the phenolic profile of prickly pear has been limited. Phenolic composition of different parts of *O. ficus-indica* had been previously addressed (MENA et al., 2018; MOUSSA-AYOUB et al., 2014). MENA et al. (2018) identified the phytochemical profiles (41 compounds, mainly phenolics) of four botanical parts from six different prickly pear cultivars of *O. ficus-indica*.

Results obtained in the present study suggest that the prickly pear is an important source of phenolic compounds and can be used as a functional and nutraceutical food. Its consumption could help in the prevention of some chronic or degenerative diseases (SOTO-HERNÁNDEZ et al., 2017).

### Flavonoid content

The flavonoid concentrations in all varieties were lower than the phenolic compound content, probably due to the presence of procyanidins (condensed tannins) and phenolic acids (chlorogenic acid and ferulic acid), which also occurs in other fruits (MENA et al., 2018). It is important to highlight that procyanidin content has not been studied in prickly pear. Another explanation for the loss of flavonoids may be that during the ripening of fruits these metabolites are transformed into phenolic compounds (BARZ and HOESEL, 1977). No significant differences in flavonoid content were found among varieties (Tab. 3). In all varieties, higher concentrations of these metabolites were observed in the peel than in the pulp. According to BRIELMAN et al. (2006), some types of flavonoids are responsible for the white or yellow pigmentation of some plant tissues, which agrees with the results obtained in the present study. Few studies describe the presence of flavonoids in prickly pear fruits (MOUSSA-AYOUB et al., 2011; MENA et al., 2018). These metabolites are also important as they have antioxidant, anti-inflammatory, and anticancer properties (CROZIER et al., 2009). Furthermore, the results of this study can contribute knowledge of a food resource used ancestrally and still a part of the cultural identity of some states in the Mexican Republic, but with little recognized potential today.

### Ascorbic acid content

Significant differences ( $P \leq 0.05$ ) were found in ascorbic acid content in the peel of the Plátano, Jade, and Copena VI varieties as compared to the remaining varieties (Tab. 3). In the peel of the Roja Villanueva, Morada, Copena CEII, Tzaponopal Rojo, and Huatusco varieties, no ascorbic acid was detected. Despite to the unstable structure of this metabolite in the presence of oxygen (DE ANCOS et al., 2009), its possible degradation can be ruled out because all the samples were processed simultaneously; therefore, the difference may be due to genetic factors. LATOCHA et al. (2011) point out that genotype is one of the most important factors in the identification of fruits with high ascorbic acid content. Alteña Roja pulp presented the highest ascorbic acid content ( $37.14 \text{ mg AAE } 100 \text{ g}^{-1} \text{ FW}$ ), which is similar to that reported by CORRAL-AGUAYO et al. (2008) ( $40 \text{ mg AAE } 100 \text{ g}^{-1} \text{ FW}$ ) but inferior to that reported by KUTI (2004) in *O. ficus-indica* fruits ( $45.8 \text{ mg AAE } 100 \text{ g}^{-1} \text{ FW}$ ). FIGUEROA et al. (2010) reported lower levels for the Mango (*O. albicarpa*) and Cacalote (*O. cochinera*) cultivars ( $5.31 \text{ mg AAE } 100 \text{ g}^{-1} \text{ FW}$  and  $25 \text{ mg AAE } 100 \text{ g}^{-1} \text{ FW}$ , respectively).

Values found in the present study support the idea that the content of this metabolite in some prickly pear varieties could contribute

to antioxidant activity in consumers' diets (ALBANO et al., 2015). Differences in ascorbic acid content reported by other authors could be attributable to crop conditions, taking into account the tolerance of prickly pear species to extreme climatic conditions, because when *Opuntia* species (CAM plants) are grown under limiting soil nutrient and water conditions, their chemical composition may change (SUMAYA-MARTÍNEZ et al., 2011).

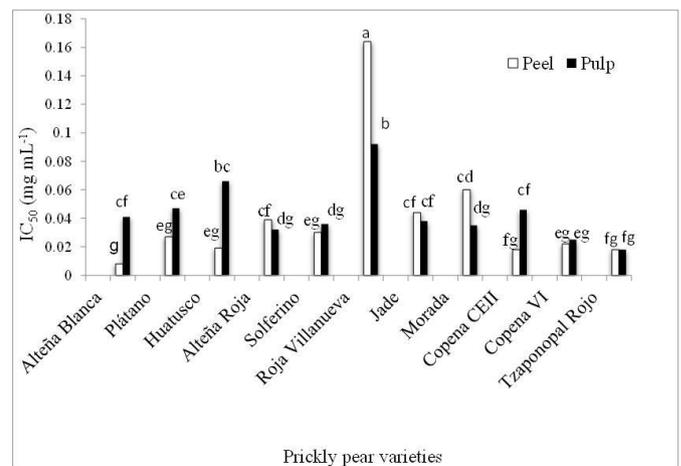
### Antioxidant activity

Alteña Blanca peel presented the highest antioxidant activity (lowest concentration of sample required to inhibit 50% of the DPPH radical concentration) (Fig. 1). Roja Villanueva peel had the lowest antioxidant activity. In addition, some significant differences ( $P \leq 0.05$ ) were found between the  $IC_{50}$  of the pulp and peel of this variety compared to that of the same tissues of the other varieties (Fig. 1). However, other methods that more completely reflect antioxidant capacity were not evaluated.

Among the advantages of cultivating *Opuntia* fruits are the presence of antioxidant compounds and the high content of stable pigments (betalains) in the pulp and peel of some varieties (Tzaponopal Rojo, Copena CEII, and Copena VI), which are important to the food industry, and their low water requirements. These advantages make them an option for agriculture in arid and semiarid regions of the country.

BRAT et al. (2007) mention that the antioxidant activity of fruits and vegetables is not only associated with phenolic compounds but is also attributed to the content of other metabolites such as vitamin C, carotenoids, and sulfur compounds; therefore, the antioxidant activity found in some varieties in the present study may be due to the possible synergistic effect of a different set of phytochemicals.

ÁVILA-NAVA et al. (2014) used the DPPH method to evaluate the antioxidant activity of *O. ficus-indica* fruits. Healthy individuals first consumed an antioxidant-poor diet and then consumed  $300 \text{ g}$  of *O. ficus-indica*. At the end of the study, a significant increase in antioxidant activity in plasma and blood (20% and 5%, respectively) resulted.



**Fig. 1:** Antioxidant activity in the fruit of 11 varieties and species of prickly pear (*Opuntia* sp.) harvested at the Universidad Autónoma Chapingo Experimental Station, Chapingo, Mexico. Different letters in the bars are significantly different (Tukey,  $P \leq 0.05$ ).

### Color parameters

In most varieties, significant differences between pulp and peel lightness were observed (Tab. 4). Alteña Blanca pulp (lightest color) presented the greatest  $L$ , followed in descending order by the

**Tab. 4:** Color parameters of the fruit of 11 varieties and species of prickly pear (*Opuntia* sp.) harvested at the Universidad Autónoma Chapingo Experimental Station, Chapingo, Mexico.

Species	Variety	Tissue*	(L)	(Hue)	(Chroma)
<i>Opuntia</i> sp.	Alteña Blanca	Peel	38.23b	100.96a	17.90eg
		Pulp	45.83a	88.87b	12.03hj
<i>Opuntia</i> sp.	Alteña Roja	Peel	23.51e	9.91hj	10.39jl
		Pulp	18.73gh	21.25e	27.43a
<i>O. ficus indica</i>	Huatusco	Peel	25.82ed	52.57c	10.54il
		Pulp	36.11b	54.14c	14.74fh
<i>O. ficus indica</i>	Solferino	Peel	22.83ef	18.52ef	11.49hk
		Pulp	18.62gi	13.12fi	27.39a
<i>O. ficus indica</i>	Roja Villanueva	Peel	19.81fg	4.73 j	14.21gi
		Pulp	19.67fh	15.75eg	23.35bc
<i>O. ficus indica</i>	Jade	Peel	19.71fh	13.52 fi	7.89 km
		Pulp	15.29ik	9.01hj	20.38ce
<i>O. ficus indica</i>	Copena CEII	Peel	19.31gh	14.02fh	6.32m
		Pulp	14.68jk	8.54 hj	18.47df
<i>O. ficus indica</i>	Copena VI	Peel	19.32gh	12.01gi	6.97 lm
		Pulp	16.31hj	7.98ij	21.70cd
<i>O. megacantha</i>	Morada	Peel	19.39fh	12.21gi	8.24km
		Pulp	15.52ik	13.27fi	23.12bc
<i>O. megacantha</i>	Plátano	Peel	29.25cd	54.91c	14.52gh
		Pulp	31.08c	43.08d	25.56ab
<i>O. robusta/Larreyi</i>	Tzaponopal Rojo	Peel	19.31gh	12.31gi	6.95 lm
		Pulp	12.60k	11.75gi	25.56ab
CV			5.79	8.09	9.02

\*Fresh weight; Lightness (L); Tone (Hue); Purity (Chroma); CV = Coefficient of variation. Means with the same letters in a column are not significantly different (Tukey,  $P \leq 0.05$ ).

Huatusco and Plátano varieties. In the exact same order, the above varieties recorded the lowest total betalain content and are thus considered to be low-pigmented varieties. The Tzaponopal Rojo variety had the darkest color pulp and also recorded the lowest L. The pulp of this variety recorded the highest total betalain content, whereas its IC<sub>50</sub> was the lowest.

Alteña Blanca had the clearest peel color since it presented the greatest L; however, the peel of this variety also had the lowest IC<sub>50</sub>, (i.e., the highest antioxidant activity per unit weight). This could indicate that phytochemicals other than pigments were involved and contributed to the antioxidant activity. Copena CEII, Copena VI, and Tzaponopal Rojo presented the lowest peel L; these varieties consistently have dark colors (purple-red) and in this analysis showed low hue values (tone angle).

Most varieties exhibited lower hue values in the pulp than in the peel (i.e., the pulp was redder than the peel). The peel and pulp showed similar hue values only in some varieties (Huatusco, Morada, and Tzaponopal Rojo). Pulp hue values followed a trend very similar to that described for peel; the lowest hue values (less than 9°) were observed in the pulp of the Copena CEII and Copena VI varieties, which consistently had the highest total betalain content.

Peel hue values could be divided into four distinct groups: (a) the Alteña Blanca variety, which had a high value (greater than 100°) and consistently presented the lowest total betalain content in the peel; (b) the Plátano and Huatusco varieties, with medium values (52°-55°) and relatively low total betalain content; (c) most other

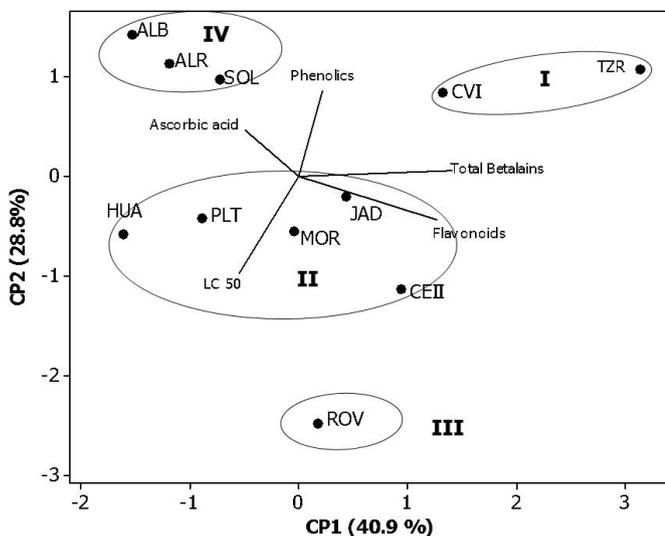
varieties, with low values (18°-20°) and relatively high total betalain content; and (d) the Roja Villanueva, which had a very low value (less than 5°) and consistently presented the highest total betalain content. Except for the Alteña Blanca variety, chroma values (color purity) were higher in the pulp than in the peel. The chroma in descending order was as follows: Alteña Roja > Solferino > Plátano > Tzaponopal Rojo > Roja Villanueva > Morada > Copena VI > Jade > Copena CEII > Huatusco > Alteña Blanca. The greater the chroma, the more vibrant and attractive the colors. Results indicate that the pulp in varieties with higher chroma values has more attractive color than that in varieties with lower chroma values.

The highest chroma values in peel were exhibited by the yellow varieties (Alteña Blanca and Plátano), meaning that these had more defined and brighter colors; however, they showed the lowest total betalain content, which indicates that different pigments are possibly responsible for their colors. The Roja Villanueva variety also exhibited a relatively high chroma value, which, in this case, may be associated with high total betalain content. Neither in the peel nor in the pulp was there any clear association between color purity (chroma) and pigment content or antioxidant activity.

In the international trade of prickly pear, major markets demand colorful red and yellow fruits, the brighter the better. The typical colors of the fruits are due to the presence of certain pigments (betalains, anthocyanins, and carotenes), which have antioxidant capacity. Therefore, it is important to analyze color and antioxidant content to identify phylogenetic materials with high nutraceutical

potential. According to the results of the present study, peels of the Roja Villanueva, Copena CEII, Copena VI, and Tzaponopal Rojo varieties could be outstanding raw material for the food industry because of their high betalain content.

Principal component analysis was used to group the varieties by type of antioxidant components (JOLLIFFE, 2005). The principal components (PC 1 and PC 2) explained 69.6% of data variability (40.9% and 28.8%, respectively) (Fig. 2). Fig. 2 depicts the makeup of the pulp of four groups of prickly pear fruit varieties: Group I (Copena VI and Tzaponopal Rojo varieties) had a high betalain content and the highest IC<sub>50</sub>; Group II (Jade, Morada, Copena CEII, Plátano, and Huatusco varieties) presented an intermediate total phenol content and LC<sub>50</sub>; Group III (Roja Villanueva variety) had the lowest IC<sub>50</sub>; and Group IV (Alteña Blanca, Alteña Roja, and Solferino varieties) had the highest phenol and vitamin C concentrations, respectively. Finally, it is recommended that research on the nutraceutical quality of other genotypes be intensified, aiming to increase their demand, generate new spaces for commercialization, and contribute to conservation of the country's cultural identity.



**Fig. 2:** Scatterplot of the principal component analysis of the pulp of 11 varieties of prickly pear by antioxidant metabolite type. Alteña Blanca (ALB), Alteña Roja (ALR), Copena CEII (CEII), Copena VI (CVI), Huatusco (HUA), Jade (JAD), Tzaponopal Rojo (TZR), Morada (MOR), Plátano (PLT), Roja Villanueva (ROV), and Solferino (SOL).

## Conclusions

Total betalain concentration was the highest in the pulp and peel of red-purple fruits, and *L* and *hue* values were the lowest in darker prickly pears; by contrast, betacyanin content was lowest in the pulp and peel of white and orange prickly pears and highest in red-purple fruits, while betaxanthin content was the greatest in white prickly pears. The peel of white prickly pears presented the greatest amount of phenolic compounds and flavonoids, as well as the highest antioxidant activity. The peel of Alteña Blanca, considered to have low marketing potential, had the highest antioxidant activity, an attribute that could help increase its marketing strength. Currently, the Tzaponopal Rojo variety is not considered an important source of pigments; however, its pulp was found to have the highest betalain content. The Alteña Roja variety had the highest ascorbic acid content in its pulp. Of all the varieties and species studied, the pulp of the Tzaponopal Rojo variety of the *O. robusta* var. *larreyi* species had the best nutraceutical attributes, as it exhibited the highest betalain

and flavonoid content, as well as the highest antioxidant activity (IC<sub>50</sub>); however, no ascorbic acid was detected in it. In conclusion, the extraction of pigments from the pulp and peel of the Tzaponopal Rojo variety could be useful mainly in the food industry.

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