

RESEARCH ARTICLE

# Antioxidant properties of a thermoultrasonicated cactus pear [*Opuntia ficus indica* (L.) Mill (Cactaceae)] juice blend in comparison with traditional thermal processing

Alexis Ayala-Niño, Nelly del Socorro Cruz-Cansino, Esther Ramírez-Moreno, José Antonio Sánchez-Franco, José Alberto Ariza-Ortega, Zuli Guadalupe Calderón-Ramos, Luis Delgado-Olivares\*.

Centro de Investigación Interdisciplinario. Área Académica de Nutrición, Instituto de Ciencias de la Salud, Universidad Autónoma del Estado de Hidalgo. Circuito Ex hacienda La Concepción S/N, Carretera Pachuca-Actopan, San Agustín Tlaxiaca, Hidalgo, 42160, México

## ABSTRACT

Functional beverages market requires conservation methods where bioactive compounds could persist for their bioavailability. The aim of this work was determined the extraction yield, antioxidant capacity and bioaccessibility of a cactus pear (*Opuntia ficus-indica*) juice blended using different solvent to extraction after pasteurization (PAST) or thermoultrasonication (TUS) treatment. The best properties were exhibited by TUS juice, showing  $575.8 \pm 11.1$  mg GAE·L<sup>-1</sup> for total phenolic, and  $392.78 \pm 4.1$  mg BE·L<sup>-1</sup> for pigment content. Which resulted in a greater antioxidant activity bioaccessible (87% DPPH; 38% ABTS). The highest antioxidant activity was found in the TUS methanolic extraction, exhibiting higher bioaccessibility than the juice (78% ABTS; 179% DPPH). It can be concluded that thermoultrasonication and methanolic extraction of a cactus pear juice blend allows a higher amount of antioxidant compounds to be bioavailable.

**Keywords:** Antioxidants; Bioaccessibility; Blend Juice; Cactus Pear; *Opuntia ficus indica*; Thermoultrasound.

## INTRODUCTION

Cactus pear is the fruit of *Opuntia ficus-indica* (L.) Mill (Cactaceae), which is considered as a functional food (Piga, 2004), by the nutritional benefits on the health due to its high content in antioxidant compounds (Abdel-Hameed et al., 2014). The cactus pear fruit is cultivated to forage, or to grow the cochineal insect and as human consumption (Reyes-Agüero et al., 2005). Actuality the phytochemical and nutritional composition is well known (Feugang et al., 2006; Galati et al., 2003), and exist a special interest of their antioxidant compounds by its effect antiulcerogenic, neuroprotective, hepatoprotective, antiproliferative and anticlastogenic capacity (Kaur et al., 2012; Madrigal-Santillán et al., 2013).

The cactus pear is a seasonal fruit, which reaches its maturity only from July to October. The use of food technologies can increase cactus pear their shelf life (Cefola et al., 2014). In Mexico there are two varieties of

cactus pear of greater consumption, the purple and green pear, however, despite the higher content of antioxidants in purple pear, people prefer to consume green cactus pear. On the other hand, lack of a guaranteed market, the absence of collection spaces and proper storage for the conservation of the fruit as well as deficiency in the transportation infrastructure makes there is a post-harvest loss of about 60% (Sumaya-Martínez et al., 2011). To avoid these problems, has been proposed that this fruit can be processed as juice (Sáenz, 2000), where thermal and physical treatments are used in order to avoid spoilage by microorganisms and native enzymes, causing irreversible loss of fresh juice flavor, as well as undesirable browning reactions, loss of antioxidant content and its nutritional properties (Ikubor, 2003).

There are processing alternative methods for juices, using emerging technologies: high pressure, irradiation, electric fields, oscillating magnetic fields, no conventional heating, ultrasound (sonication), etc. Ultrasound has been described

### \*Corresponding author:

Luis Delgado Olivares, Centro de Investigación Interdisciplinario. Área Académica de Nutrición, Instituto de Ciencias de la Salud, Universidad Autónoma del Estado de Hidalgo. Circuito Ex hacienda La Concepción S/N, Carretera Pachuca-Actopan, San Agustín Tlaxiaca, Hidalgo, 42160, México. **Phone:** +52-771-717-2000, ext. 4312; **E-mail:** ldelgado@uaeh.edu.mx

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recently as a method able to preserve foods; this technology emits sound waves where the frequency is not perceived by the human. This emerging technology is low cost, eco-friendly and microbial decontamination efficient (Awad et al., 2012), without affecting the sensorial quality (Gómez-Lopez et al. 2018), in comparison with other technologies such as pasteurization. Alcantara-Zavala (2021), reported that the use of thermoultrasound (combination of ultrasound with controlled temperature) to conservation of pulque improves sensorial, physicochemical and microbiological properties, increasing the quality of the final product in comparison with pasteurization.

In different studies this technique has been evaluated as an alternative to heat treatments to process purple and green cactus pear juices in achieving the microbial count reduction without affecting the juice quality and its antioxidant properties (Zafra-Rojas et al., 2013; Cruz-Cansino et al., 2015). This is important to ensure a high quality in the juice with bioactive compounds that can help to human health. For this reason, the present study had as objectives 1) compare the effect of thermoultrasound (TUS) and pasteurization (PAST) treatments on antioxidant properties of a mixed juice with two kinds of cactus pears (*O. ficus-indica*) fruits and 2) determinate the *in vitro* bioaccessibility of antioxidants before and after of its extraction with different solvents.

## MATERIAL AND METHODS

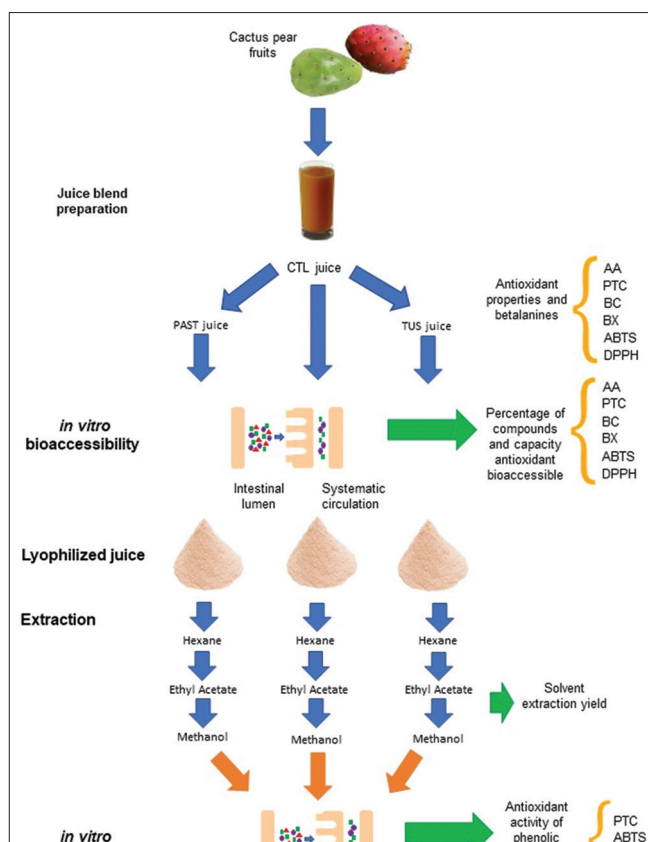
Fig. 1 describes the methodology used in this work.

### Plant material

Samples of two varieties of green and red cactus pears (*O. ficus-indica*) (ten kilograms of each cultivar Reyna and Rojo Pelon) were donated by Mexican Association CoMeNTuna growers of Actopan; Hidalgo, Mexico, to the Institute of Health Sciences laboratory. The samples were selected according to their standards of commercial maturity, then washed and the epicarp was removed manually. The samples correspond to the season from June to August 2018. They were grown in a semi-dry climate with a temperature ranging from 11 to 37 °C.

### Juice preparation and treatments

The juice elaboration and thermoultrasound treatment was according previous study by Cruz-Cansino et al. (2016), where processing conditions (amplitude and time) in which the optimal treatment conditions for juice were established based on complete inactivation of microorganisms, high ascorbic acid, phenolic content and antioxidant capacity. To obtain the juice, the fruits were homogenized with a Lab blender (38BL52 LBC10, Waring



**Fig 1.** Methodologic diagram. Cactus pear juice was prepared from green and red cactus pear, which was used as a control (CTL), later it was subjected to pasteurization (PAST) or thermoultrasound (TUS), and the antioxidant activity by ascorbic acid (AA), total phenolic content (TPC), betacyanin (BC), and betaxanthin (BX) and capacity (ABTS and DPPH) were determined. Subsequently, the bioaccessibility assay was carried out to determine the percentage of bioaccessible components and antioxidant capacity. The juices were lyophilized to subsequently carry out a consecutive extraction with three solvents (hexane, ethyl acetate and finally with methanol), and the extraction performance was determined. Finally, the bioaccessibility assay of the solvent fraction with the highest yield was carried out, determining the antioxidant activity and capacity.

Commercial<sup>®</sup>, USA). Then, the juice obtained was placed in a conventional strainer to remove the seeds and finally mix the juices [Reyna and Rojo Pelon 4:6 (v: v)]. After the thermoultrasound (TUS) (80 % of amplitude for 20 min, at 45 °C) (VCX-1500, Sonics & Materials, Inc. Newtown, CT, USA) was applied to the juice. To corroborate the effect of the thermoultrasound treatment on the bioactive components of the juice, this was compared with a pasteurized juice (70 °C, 30 min) (PAST) and another juice without treatment, considered as a control sample (CTL).

### Antioxidant content and antioxidant activity in juice

The samples of juice were centrifuged at  $3400 \times g$  for 20 min and the supernatant was filtered with a pore size of  $0.22 \mu\text{m}$  (Millipore Millex<sup>™</sup> - GV PVDF). The filtrate was used to evaluate ascorbic acid (AA), which was expressed as milligram ascorbic acid per liter (mg

AA·L<sup>-1</sup>) (Dürüst et al., 1997), total phenolic content (TPC), the values are expressed as milligram gallic acid equivalent per liter (mg GAE·L<sup>-1</sup>) (Stintzing et al., 2005), betalains content was expressed as milligram betalains equivalent per liter (mg BE·L<sup>-1</sup>) (Castellar et al., 2003), while the values of antioxidant capacity by ABTS<sup>•+</sup> and DPPH<sup>•</sup>, were expressed as milligrams of Vitamin C Equivalents Antioxidant Capacity per liter (VCEAC·L<sup>-1</sup>) and micromoles of trolox equivalents per liter (μmol TE·L<sup>-1</sup>), respectively (Kuskoski et al., 2005; Kim et al., 2002).

### ***In vitro* intestinal bioaccessibility**

CTL, PAST and TUS juices were used to determine intestinal bioaccessibility, thus the *in vitro* digestion model followed by dialysis was performed according to the method described by Miller et al. (1981) with some modifications. Aliquots of 20 mL were used for *in vitro* digestion and after dialyzed during 6 h. Finally, the samples were used to evaluate ascorbic acid, total phenolic content, betalains and antioxidant capacity by ABTS and DPPH. The values obtained of bioactive compounds and antioxidant capacity before *in vitro* bioaccessibility were used as 100%, and percentage in samples after digestion was considered as an indicator of bioaccessibility in the small intestine.

### **Extractions with solvents and yields of extraction**

One liter of control, pasteurized and thermoultrasonicated juice were lyophilized (Labconco. FreeZone 6 Liter -50C Console Freeze Dryer USA), grounded and sieved at particle size at 500 micres. 5 g of powder were used to realize the extraction by maceration at room temperature (20 °C) subsequently in the following order: hexane, ethyl acetate (EtOAc) and methanol (MeOH). The extraction yield was determined according to Chougui et al. (2013). In each one of extractions with the different solvents were used to measure total phenolic content and antioxidant capacity by ABTS<sup>•+</sup> and DPPH<sup>•</sup>. The extract with the highest phenolic and antioxidant capacity content was used to measure *in vitro* bioaccessibility assay.

### **Statistical analysis**

The values were obtained by triplicate and expressed by standard deviation of the mean (SD). The data were analyzed by one-way analysis of variance (ANOVA) test, and differences among means compared using Tukey test with a level of significance of  $p < 0.05$ , using the SPSS<sup>®</sup> System for WIN<sup>™</sup> version 15.0.

## **RESULTS AND DISCUSSION**

### **Ascorbic acid**

The ascorbic acid (AA) has been studied in many food matrices e. g vegetables and fruits (Kapur et al., 2012),

kiwi, *Emblca officinalis* fruits (Kvesitadze et al., 2001; Raghu et al., 2007), juice fruits and vegetable (bergamot, lemon, tomato) (María & Riccardo, 2020; Rekha et al., 2012; Adekunle et al., 2010) as well as sonicated (strawberry, purple cactus pear, grapefruit, kiwifruit) (Tiwari et al., 2009; Zafra-Rojas et al., 2013; Aadil et al., 2013; Wang et al., 2019) and thermoultrasonicated juices (apple, purple cactus pear; blackberry, quince, jackfruit) (Abid et al., 2014; Cruz-Cansino et al., 2015; Cervantes-Elizarrarás et al., 2017; Yıkmiş et al., 2019; Cruz-Cansino et al., 2021). And it has been reported that the thermoultrasonication in juices allow the ascorbic acid stability or sometimes a decrease, and it depends of food matrix, equipment, ultrasound application conditions and probe used (Aguilar et al., 2017). The ascorbic acid values (Table 1) in CTL juice were significantly high ( $p < 0.05$ ) (286.4 mg AA·L<sup>-1</sup>) compared to PAST and TUS juices (259.4 and 236.8 mg AA·L<sup>-1</sup>, respectively).

The loss of this compound during the process of thermoultrasound could be due to processes of oxidation in aerobic and anaerobic environments related with the production and use of hydroxyl radicals (Valdramidis et al., 2010). The severe physical conditions existing inside the cavitation bubbles collapse in a micro scale, causing several sonochemical reactions that happen at the same time or separately (Tiwari et al., 2009). For instance, when juice is sonicated, the empty spaces are filled with water vapor and gases dissolved in liquid medium, which may aid oxidation reactions promoted by interactions with free radicals (Korn et al., 2002).

Compared to a previous study in cactus pear juices, control and pasteurized purple cactus pear juice values were low (around 100 mg AA·L<sup>-1</sup>), while the thermoultrasonicated

**Table 1: Antioxidant content and antioxidant capacity**

Determination	CTL	PAST	TUS
Ascorbic Acid (mg AA·L <sup>-1</sup> )	286.4 ± 10.8 <sup>a</sup>	259.4 ± 10.8 <sup>b</sup>	236.8 ± 3.1 <sup>a</sup>
Total Phenolic Content (mg GAE·L <sup>-1</sup> )	411.5 ± 7.9 <sup>a</sup>	499.6 ± 23.1 <sup>b</sup>	575.8 ± 11.1 <sup>c</sup>
Betacyanin (mg BE·L <sup>-1</sup> )	371.51 ± 8.8 <sup>b</sup>	335.6 ± 4.6 <sup>a</sup>	392.78 ± 4.1 <sup>c</sup>
Betaxanthin (mg BE·L <sup>-1</sup> )	176.07 ± 1.2 <sup>b</sup>	164.52 ± 2.7 <sup>a</sup>	176.42 ± 0.4 <sup>b</sup>
ABTS (mg VCEAC·L <sup>-1</sup> )	30.5 ± 0.9 <sup>a</sup>	35.8 ± 0.0 <sup>b</sup>	39.5 ± 0.3 <sup>c</sup>
DPPH (μmol TE·L <sup>-1</sup> )	426.8 ± 29.9 <sup>a</sup>	434.3 ± 16.6 <sup>a</sup>	673.8 ± 41.6 <sup>b</sup>

Data were expressed as means ± standard deviation. Different letters between treatments indicate statistically significant differences ( $p < 0.05$ ) between means. CTL (Control); PAST (Pasteurization); TUS (Thermoultrasonication); mg AA·L<sup>-1</sup> (milligrams of Ascorbic Acid per liter); mg GAE·L<sup>-1</sup> (milligrams of Gallic Acid Equivalents per liter); mg BE·L<sup>-1</sup> (milligrams of betalains Equivalents per liter); mg VCEAC·L<sup>-1</sup> (milligrams of Vitamin C Equivalents Antioxidant Capacity per liter); μmol TE·L<sup>-1</sup> (micromoles of Trolox Equivalent per liter).



juice showed similar values ( $> 250 \text{ mg AA}\cdot\text{L}^{-1}$ ) (Cruz-Cansino et al., 2015). In other fruit juice matrices such as blackberry juice, values of 360.14, 291.16 and 248.10  $\text{mg AA}\cdot\text{L}^{-1}$  have been reported in control, thermoultrasonication and pasteurization, respectively (Cervantes-Elizarrás et al., 2017). While thermoultrasonicated jackfruit nectar the result was of 101.24  $\text{mg AA}\cdot\text{L}^{-1}$  in comparison with pasteurized nectar (52.40  $\text{mg AA}\cdot\text{L}^{-1}$ ) (Cruz-Cansino et al., 2021) and in blood fruit juice on different thermoultrasound conditions obtained a range of 212.3 to 397.2  $\text{mg AA}\cdot\text{L}^{-1}$ , while untreated sample had 312.3 and the pasteurized 181.3  $\text{mg AA}\cdot\text{L}^{-1}$  (Raju and Deka, 2018).

### Total phenolic content

The value of total phenolic (IPC) content in CTL was  $411 \pm 7.9 \text{ mg GAE}\cdot\text{L}^{-1}$ . After thermoultrasonication, juice showed higher ( $p < 0.05$ ) values ( $575.8 \pm 11.1 \text{ mg GAE}\cdot\text{L}^{-1}$ ) than PAST ( $499.6 \pm 23.1 \text{ mg GAE}\cdot\text{L}^{-1}$ ) and CTL juices ( $411.5 \pm 7.9$ ) (Table 1). The increase of phenolic content is attributed to cavitation effects and collapse bubbles that implode which break the vacuoles or membrane of vegetable cells allowing release of these compounds that were linked to the polysaccharides of the cell wall (Cheng et al., 2007; Jabbar et al., 2015), as well as hydroxyl radicals (OH) generated sonochemically bind to the aromatic ring (Bath et al., 2011). The increase of these compounds in thermoultrasonicated juice has been reported in several studies on different fruits e. g. star, quince, and blueberry (Nayak, 2018; Yıkımsı, 2019; Wu, 2021). However, the results were different in thermosonicated mango juice since the phenolic content was low in comparison with control sample with treatments at 25 °C and 45 °C, indicating that these compounds are thermosensibles (Dars, 2019). Therefore, the variability of result can depend on the food matrix and on the conditions of treatments applied.

### Betalains content

Respect to betalains, the TUS juice had significantly high ( $p < 0.05$ ) values in betacyanins ( $392.78 \pm 4.1 \text{ mg BE}\cdot\text{L}^{-1}$ ) compared with CTL and PAST samples ( $335.6 \pm 4.6$  and  $371.51 \pm 8.8 \text{ mg BE}\cdot\text{L}^{-1}$  respectively) (Table 1). Regarding betaxanthins, TUS and CTL juices showed similar ( $p > 0.05$ ) values ( $176.42 \pm 0.4$  and  $176.01 \pm 1.2 \text{ mg BE}\cdot\text{L}^{-1}$ , respectively) and significantly higher ( $p < 0.05$ ) than the PAST ( $164.52 \pm 2.7 \text{ mg BE}\cdot\text{L}^{-1}$ ). The high betalains content in TUS is due to the combination of ultrasound and moderated temperature application preventing the degradation of these compounds (Zenker et al., 2003). On the other hand, the temperature applied in PAST juice degraded these molecules since they are sensitive to high heat treatment causing hydrolysis (Fernández-López et al., 2007). In a previous study on purple cactus pear juice different behaviour was obtained, the control and pasteurized juice had similar values of betacyanins ( $291.52 \pm 38.92$  and  $272.43 \pm 15.34 \text{ mg}$

$\text{BE}\cdot\text{L}^{-1}$ , respectively), while thermoultrasonication at 80% amplitudes by 15 and 25 min had  $217.86 \pm 55.71$  and  $237.54 \pm 35.16 \text{ mg BE}\cdot\text{L}^{-1}$ , respectively, and in betaxanthins there were no significant differences between the samples (Cruz-Cansino et al., 2015). In other food matrix such as clear red pitaya betacyanins content reported similar values between thermoultrasonicated and untreated juice ( $149.0 \pm 0.07$  and  $140.7 \pm 0.10 \text{ mg BE}\cdot\text{L}^{-1}$ , respectively) (Liao et al., 2020). The same matrix in other study found a decrease ( $p < 0.05$ ) of 130.9  $\text{mg BE}\cdot\text{L}^{-1}$  in TUS and 136.1  $\text{mg BE}\cdot\text{L}^{-1}$  in thermal processing (Zhu et al., 2021).

### Antioxidant capacity by ABTS and DPPH

Assays of antioxidant capacity by ABTS and DPPH have been applied extensively in different matrices e. g. soil, oil, coffee, vegetables, leaves, seeds, fruits and juices (Panuccio et al., 2019; Giuffrè et al., 2018; Choi and Koh 2017; Embuscado, 2015; Sandoval-Gallegos et al., 2021; Ortega-Ortega et al., 2017; Becker et al., 2019; Zapata et al., 2017). Two action mechanisms to blend juice analysis in the present study were used, the results are shown in table 1. The TUS sample had significantly ( $p < 0.05$ ) higher values of antioxidant activity measured by ABTS mechanism ( $39.5 \pm 0.3 \text{ mg VCEAC}\cdot\text{L}^{-1}$ ) than CTL ( $30.5 \pm 0.9 \text{ mg VCEAC}\cdot\text{L}^{-1}$ ) and PAST juice ( $35.8 \pm 0.1 \text{ mg VCEAC}\cdot\text{L}^{-1}$ ). The results of DPPH behavior was similar to ABTS analysis in TUS juice, significantly high ( $p < 0.05$ ) values of  $673.8 \pm 41.6 \mu\text{mol TE}\cdot\text{L}^{-1}$ , while similar results ( $p > 0.05$ ) were found between CTL and PAST samples ( $426.8 \pm 29.9$  and  $434.3 \pm 16.6 \mu\text{mol TE}\cdot\text{L}^{-1}$ , respectively). Studies by thermoultrasound in juices reported similar behaviour, in thermoultrasonicated blackberry juice presented high antioxidant capacity by ABTS and DPPH ( $1118.20 \pm 96.8$  and  $13964.96 \pm 570.1 \mu\text{mol TE}\cdot\text{L}^{-1}$ , respectively) in comparison with pasteurized juice ( $941.22 \pm 31.8$  and  $11935.04 \pm 425.0 \mu\text{mol TE}\cdot\text{L}^{-1}$ , respectively) (Cervantes-Elizarrás et al. 2017).

It has been reported that bioactive compounds such as ascorbic acid, phenol, anthocyanins and betalains are related with the antioxidant activity by ABTS and DPPH (Flores-Mancha et al., 2021; Zafra-Rojas et al., 2016; Sawicki et al., 2016). This behavior is due to the deliverance of antioxidants of the food matrix during the treatment of thermoultrasound (Cervantes-Elizarrás et al. 2017).

The results obtained showed that the conditions of TUS treatment could be applied as a potential option to conventional technology due to the preservation of antioxidant properties in cactus pear juice blend.

### In vitro intestinal bioaccessibility in juices

The fig. 2 shows the percentages of *in vitro* intestinal bioaccessibility of antioxidant compounds *In vitro* of the

samples. TUS and PAST juices had a high ascorbic acid bioaccessibility (56 %), in comparison with CTL juice (41 %), this because during pasteurization and ultrasonication, these treatments allows the release of the compounds of cellular structures causing a high bioavailability (Herrero and Romero, 2006). Meanwhile, control juice remained the highest concentration in percentage of phenolic compounds after the digestion process (83.17 %), followed by TUS (54.66 %), while pasteurized juice had the lowest concentration (50.99 %). The decrease of phenolic compounds in TUS and PAST samples could be due to the degradation of these compounds during the processing for the formation of new hydrogen nets, which can catch these compounds (Anese et al., 2013).

After digestion process, the betacyanins and betaxanthins in the three juices remained only around of 24 – 36 % respectively, and had no statistical difference between treatments, because the pH conditions used probably affected the bioactive compounds (Herbach and Carle, 2006).

The antioxidant activity was also affected by the *in vitro*, digestion process because there exists a positive correlation positive between antioxidant activity and antioxidant molecules. Even so, the TUS juice showed the highest antioxidant activity bioaccessible (87% of ABTS and 38% of DPPH) compared with the other samples (Fig. 2).

### Extraction yield

Extraction with three different solvents was performed and the yield obtained is presented in table 2. It can be perceived that methanolic extraction of all the treatments (TUS, PAST and CTL) had the highest extraction yield over 800 g·kg<sup>-1</sup> compared with other solvents extraction ( $p < 0.05$ ). This means that methanol extraction had the most of the compounds, and it is due to the polar characteristic of this solvent (Aderogba et al., 2010).

### Total phenols and antioxidant capacity in extracts

The antioxidant capacity of extractions with three different solvents, hexane, ethyl acetate, and methanol was evaluated. In all samples, the total phenolic content and antioxidant capacity increased with the more polar solvent (Table 3), because the antioxidant compounds generally tend to be more polar. TUS methanolic extract showed the highest value ( $p < 0.05$ ) in comparison with other extracts. The high values in TUS samples is due to the deliverance of antioxidant compounds during sonication (Zafra-Rojas et al., 2013) which is related to the antioxidant activity obtained in the TUS juice (Table 1).

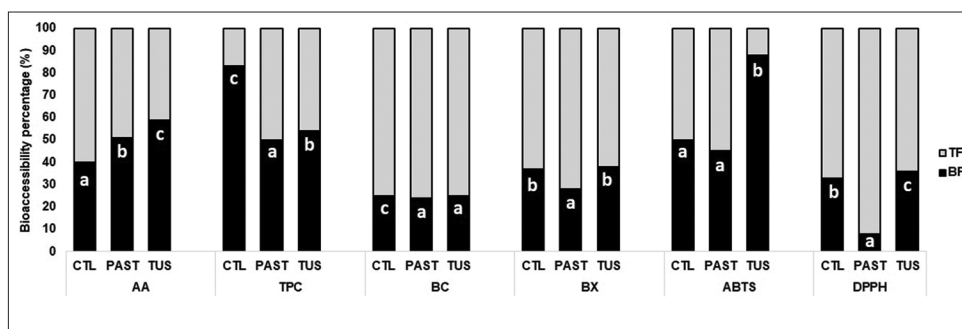
### *In vitro* Intestinal bioaccessibility of antioxidants extraction

As it has been mentioned, methanolic extraction reported the highest yield in the obtention of compounds and antioxidant activity, therefore, the bioaccessibility study was measured in this sample. In fig. 3, it can be observed that all determinations had more than 67 % of bioaccessibility. This could be because the methanolic extraction had less concentration of complex carbohydrates that could interact with the antioxidants and interfere with its bioavailability (Palafox-Carlos et al., 2011). In general, the antioxidant capacity measured by ABTS, PAST and TUS treatment samples showed a decrease of its antioxidant activity bioaccessible in comparison with the control, while DPPH, all the samples had more than 100 % of bioaccessibility. Neither ascorbic acid and betalains could be

**Table 2: Solvent extraction yield in cactus pear juice blended.**

Treatment	Hexane (g·Kg <sup>-1</sup> ).	Ethyl Acetate (g·Kg <sup>-1</sup> ).	Methanol (g·Kg <sup>-1</sup> ).
CTL	0.18 ± 0.0 <sup>a</sup>	0.56 ± 0.2 <sup>b</sup>	894.7 ± 28.0 <sup>c</sup>
PAST	0.24 ± 0.0 <sup>a</sup>	0.78 ± 0.2 <sup>b</sup>	866.1 ± 32.0 <sup>c</sup>
TUS	0.48 ± 0.0 <sup>a</sup>	0.82 ± 0.4 <sup>b</sup>	832.1 ± 26.0 <sup>c</sup>

Data were expressed as means ± standard deviation. Different letters between treatments indicate statistically significant differences ( $p < 0.05$ ) between means. CTL (Control); PAST (Pasteurization); TUS (Thermoultrasonication).

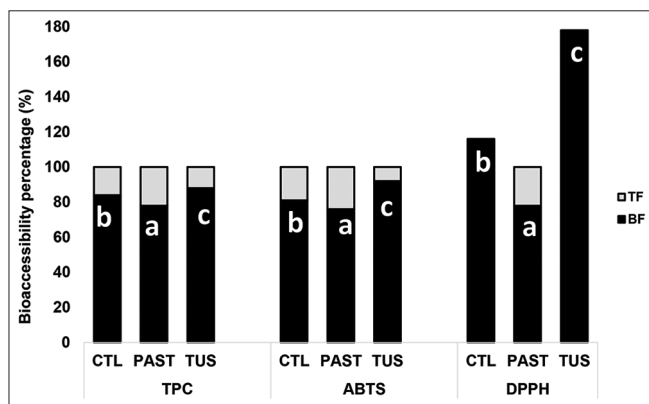


**Fig 2.** Percentage of compounds and antioxidant capacity bioaccessible in cactus pear juice blend. Data are expressed as means ± standard deviations. Different letters between treatments indicate statistically significant differences from the same determination ( $p < 0.05$ ) between means. The values obtained of antioxidant activity and capacity, before *in vitro* bioaccessibility were used as 100 % and percentage in each sample after bioaccessibility assay was considered as an indicator of bioaccessibility in the small intestine (100% is presented in the table 1 data). CTL = Control; PAST = Pasteurization; TUS = Thermoultrasonicated; AA = Ascorbic Acid; TPC = Total Phenolics Content; BC = Betacyanins; BX = Betaxanthins; TF = Total Fraction; BF = Bioaccessible Fraction.

**Table 3: Antioxidant properties of cactus pear juice solvent extraction.**

Determination	Extraction	CTL	PAST	TUS
Total Phenolic (mg GAE·L <sup>-1</sup> )	Hexane	25.63 ± 0.56 <sup>a</sup>	23.81 ± 0.89 <sup>a</sup>	33.51 ± 2.51 <sup>b</sup>
	Ethyl Acetate	221.53 ± 5.61 <sup>c</sup>	231.65 ± 11.37 <sup>c</sup>	242.92 ± 9.87 <sup>c</sup>
	Methanol	571.53 ± 6.37 <sup>d</sup>	543.87 ± 16.35 <sup>d</sup>	630.26 ± 23.12 <sup>e</sup>
ABTS (mg VCEAC·L <sup>-1</sup> )	Hexane	4.97 ± 0.33 <sup>b</sup>	1.45 ± 0.17 <sup>a</sup>	5.69 ± 1.44 <sup>ab</sup>
	Ethyl Acetate	7.10 ± 0.23 <sup>a<sup>b</sup></sup>	9.83 ± 0.69 <sup>b</sup>	14.71 ± 4.05 <sup>c</sup>
	Methanol	31.14 ± 0.83 <sup>d</sup>	43.11 ± 0.96 <sup>e</sup>	50.79 ± 1.82 <sup>f</sup>
DPPH (mmol TE·L <sup>-1</sup> )	Hexane	16.33 ± 0.9 <sup>a</sup>	62.21 ± 2.86 <sup>b</sup>	71.29 ± 4.31 <sup>b</sup>
	Ethyl Acetate	283.16 ± 25.51 <sup>c</sup>	360.12 ± 29.76 <sup>d</sup>	269.21 ± 21.99 <sup>c</sup>
	Methanol	392.02 ± 19.67 <sup>de</sup>	442.22 ± 26.23 <sup>e</sup>	622.62 ± 30.17 <sup>f</sup>

Data are expressed as means ± standard deviation. Different letters between treatments indicate significant statistically differences ( $p < 0.05$ ) between means ±. CTL (control); PAST (pasteurization); TUS (Thermoultrasonication); mg GAE·L<sup>-1</sup> (milligrams of Gallic Acid Equivalents per liter); mg VCEAC·L<sup>-1</sup> (milligrams of Vitamin C Equivalents Antioxidant Capacity per liter); µmol TE·L<sup>-1</sup> (micromoles of Trolox Equivalent per liter).



**Fig 3.** Percentage of antioxidant activity (by phenols) and capacity bioaccessible in cactus pear juice blend of methanolic extract. Data are expressed as means ± standard deviations. Different letters between treatments indicate significant statistically differences from the same determination ( $p < 0.05$ ) between means. The values obtained of antioxidant activity and capacity, before *in vitro* bioaccessibility were used as 100 % and percentage in each sample after bioaccessibility assay was considered as an indicator of bioaccessibility in the small intestine (100% is presented in the table 3). CTL = Control; PAST = Pasteurization; TUS = Thermoultrasonication; TPC = Total Phenolic Content, TF = Total Fraction; BF = Bioavailable Fraction.

identified in the bioaccessible fractions, which can suggest the use of more specific techniques for their identification during *in vitro* bioaccessibility determination. Even though, with these results, it can be observed that a methanolic extraction is related to extracts with a high intestinal absorption of antioxidants.

## CONCLUSIONS

From the data in this study, unconventional technology known as thermoultrasound, has the property to release bioactive compounds with antioxidant capacity in cactus pear juice blend, being these compounds more bioaccessible than those in no treated and pasteurized juice. Methanol had a higher extraction yield than hexane and acetate ethyl,

having as well, a high antioxidant capacity, suggesting that antioxidant compounds in cactus pear juice are polar. Also, these compounds are more bioaccessible due to solvent extraction and thermoultrasound, this because the decrease in fiber amount, and the disruption of biological cell walls that release bioactive compounds.

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## CONFLICT OF INTEREST DECLARATION

The authors have no conflict of interest to declare.

## Author's contributions

Alexis Ayala-Niño and José Antonio Sánchez-Franco performed the experiments and data analysis; Nelly del Socorro Cruz-Cansino, Esther Ramírez-Moreno and Luis Delgado-Olivares conceived, designed the experiments, data analysis and wrote the majority of the paper and managed the authors; José Alberto Ariza-Ortega and Zuli Guadalupe Calderón-Ramos wrote key sections of the paper. All authors have read and agreed to the published version of the manuscript.

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