# RESEARCH ARTICLE

# Physicochemical characterization and antioxidant activity of wild *Physalis* spp. genotypes

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# ABSTRACT

The center of origin and domestication of *Physalis* spp. is Mexico, where 70 wild species and only two cultivated species are found. Fruits of wild husk tomato are collected by farmers for home consumption or for sale in local markets, where prices are higher than for the cultivated husk tomato. Despite their economic and nutritional importance, they have been little studied. The objective of this study was to determine phytochemical properties, nutritional value, total contents of phenol, anthocyanins and flavonoids, and the antioxidant capacity of the fruits of five wild genotypes of *Physalis spp* (Quialana, Jalieza, Vigallo, Ejutla and Miahuatlán), collected in the corn harvest season in five localities of Oaxaca, Mexico. The genotypes Quialana and Miahuatlán had the best physical characteristics, but their antioxidant capacity was lower than that of the genotypes Jalieza, Vigallo and Ejutla, which did not exhibit physical characteristics that are attractive for consumers. Moreover, the high contents of phenols and flavonoids, as well as the antioxidant capacity of all the wild genotypes studied, showed that *Physalis* spp. is an option for more frequent inclusion in the population's diet. However, because of food globalization issues, production, commercialization and consumption of these wild species is disappearing.

Keywords: Nutritional value; Phenols; Vegetable; Wild husk tomato

# **INTRODUCTION**

Today, the population of Mexico has a pattern of consumption that is characterized largely by industrialized food and a gradual decrease of foods based on native regional plant species. This transition to food with high energy content has given rise to non-transmissible chronic diseases such as cardiovascular disease and type 2 diabetes mellitus. In Mexico, the diversity of plants that have potential use includes around 5000 species from different botanical families (Casas et al., 1994), most are herbaceous and wild (Caballero et al., 1998) with food and medicinal uses (Caballero et al., 2001) and are commercialized mainly in local and regional markets of southeastern Mexico. The use of a large number of native Mesoamerican plant species is pre-Hispanic and their consumption continues today in rural communities where there is a broad diversity of wild foods used in local and regional gastronomy, but they have been little studied. Moreover, the availability of these plants is limited since they are seasonal, usually gathered in backyard gardens and rainfed crops or bought in local and regional markets. Among these plant resources is the genus Physalis, of the Solanaceae family, that produces edible fruits commonly known as husk tomato, miltomato or tomatillo (Montes-Hernández and Aguirre, 1994). The existence of 124 species is known (The plant list, 2013); 70 are found in Mexico, considered its center of origin (D'arcy, 1991; Vargas-Ponce et al., 2011). In natural areas and traditions agrosystems, like those that are still conserved in the state of Oaxaca, wild husk tomato grows in association with corn or interspersed with corn, beans and squash. Their fruits are small, purple, green or yellow, and most are collected for home consumption. When rains are abundant, production and gathering of husk tomato in the corn agrosystem increases (Zamora-Tavares et al., 2015), and the surplus that is sold in regional markets at a price 100% higher than that of cultivated husk tomato (Montes-Hernández and Aguirre-Rivera, 1988) because the fruits of husk tomato is

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sweeter or less sour than cultivated husk tomato, a quality conferred by physiological and commercial maturation in the field. Also, it has been found that the fruit of several Physalis species contains secondary metabolites that have significative antioxidant potential (Chang et al., 2008; Qiu et al., 2008). Several studies have confirmed its therapeutic properties; it has been used in traditional medicine to treat diverse pathologies (Lan, et al., 2009; Soares et al., 2006). This potential is related to its high content of bioactive compounds, such as phenols and flavonoids, which heavily contribute to its antioxidant potential (Bergier et al., 2012). However, it is known that the content of these active compounds in vegetables varies according to the variety, maturation and agronomic factors as shown by the study carried out on tomato (Solanum lycopersicum) (Navarro-Gonzalez and Periago, 2016). For these reasons, it is important to study native plant species of Mesoamerica, such as the fruits of wild Physalis spp. This study had the objective of analyzing the physicochemical characteristics, and antioxidant capacity of five wild genotypes of Physalis spp. that grew in association with the cultivation of corn, in five localities of Oaxaca, Mexico.

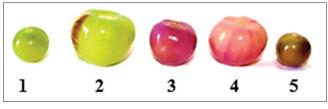
## **MATERIALS AND METHODS**

#### **Collection of fruits**

In September and October 2019, at the end of the maize crop (Zea mayz L.), in the localities of San Bartolomé Quialana, Tlacolula (1), Santa Cecilia Jalieza, Ocotlán (2), Santa María Vigallo, Zimatlán de Álvarez (3), Ejutla de Crespo (4), and Miahuatlán de Porfirio Díaz (5) (Fig. 1), in the state of Oaxaca, Mexico, between the coordinates 17° 1' 31" N and 96° 43' 11" W, at 1530 m altitude, 200 wilds Physalis spp. fruits were collected in each locality and taken to the food laboratory of the Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional Unidad Oaxaca, of the Instituto Politécnico Nacional (CIIDIR IPN Oaxaca), Mexico. The fruits were washed with water and disinfected with a 20 ppm solution of sodium hypochlorite, then dried at ambient temperature. The calix was eliminated and a random sample of 100 fruits per locality were taken for analysis and stored at room temperature ( $25 \pm 5 \,^{\circ}$ C). All the physicochemical, nutritional and quality determinations were done in triplicate and each locality was a treatment.

#### **Physicochemical parameters**

Fruits were weighed on a digital scale (OHAUS PIONEER<sup>®</sup> Corporation USA), and their equatorial and longitudinal diameters were measured with a digital vernier (Serie 500, MITUTOYO<sup>®</sup> USA). Three samples of 20 fruits were obtained at random from each treatment and the juice was extracted. It was determined the total soluble solids (TSS) with a digital refractometer (HANNA<sup>®</sup> HI 96801), acidity



**Fig 1.** Wild genotypes of *Physalis spp*, collected in different locations in Oaxaca, Mexico. 1: Quialana; 2: Jalieza; 3: Vigallo; 4: Ejutla and 5: Miahuatlán.

by titration with 0.1 mol/L NaOH using phenolphthalein and expressed as g/100 g anhydrous citric acid. pH was determined by direct reading with a potentiometer (OAKTON<sup>®</sup> PC 700). The flavor and ripeness indexes were calculated following Hernández-Suárez (2008a).

#### **Color determination**

Fruit color was determined with a colorimeter (HUNTERLAB'S<sup>®</sup> model Mini Scan); the results were transformed to hue values (\*h) by calculating the arc tangent of  $(b^*/a^*)$  Chroma (C) was calculated with the equation  $[(a^{*2} + b^{*2})^{1/2}]$  (McGuirre, 1992; McGuirre-Raymond, 1992), and the color index (CI) with the equation  $[(a^*x1000)/(L \times b^*)]$  (Francis 1975). L\*, a\*, b\*, chroma and hue were assessed by direct readings on the pericarp of three fruits.

#### **Bromatological analysis**

To determine moisture, ash, lipids, proteins and fiber, the techniques of AOAC (2000) were performed in triplicate. Nitrogen was obtained with the Kjeldahl technique, and protein concentration was calculated using the nitrogen factor 6.25. Lipid content was determined by extraction with petroleum ether, and total dietetic fiber was determined by incineration of the organic residue left after digestion with solutions of sulfuric acid and sodium hydroxide. The carbohydrates were determined by difference with the values of the parameters indicated above. Total energy was calculated in function of the results obtained for carbohydrates, proteins and lipids. The conversion factors used were based on metabolizable energy and analytical methods: 4 kcal/g for carbohydrates, 4 kcal/g for protein and 9 kcal/g for lipids (FAO, 2001).

# Total phenols, Total flavonoids and Anthocyanin content

Phenols were determined following the Folin-Ciocalteu method (Singleton and Rossi, 1965). The total phenol content was determined as gallic acid equivalents (GAE, mg of gallic acid/g dry sample). Total flavonoid content was found following the method described by Chen et al. (2014). The results were expressed as mg of quercetin equivalents (QE) per g of dry sample (mg QE/g dry sample). Total anthocyanins were determined according to the procedure established by Abdel-Aal and Hucl (1999).

#### Determination of antioxidant activity

Antioxidant activity of the five husk tomato genotypes was determined using the technique of the radical 1,1- diphenyl-2-picrilhydrazil (DPPH) (Sigma, Aldrich), following Matthaus (2002). Antioxidant activity was expressed as the percentage of inhibition corresponding to the quantity of DPPH radical neutralized by the extract at a given concentration.

## Analysis of results

The data obtained were subjected to an analysis of variance and a comparison of means (Tukey,  $P \le 0.05$ ), using SAS<sup>®</sup> version 9.0 (SAS, 2002). Graphs were obtained with Microsoft Excel<sup>®</sup>.

# **RESULTS AND DISCUSSION**

# **Physicochemical parameters**

The physicochemical parameters analyzed showed significant differences (P < 0.05) among the five groups of wild Physalis spp. fruits (Table 1). Two sizes were found: small spherical globose with equatorial and longitudinal diameters smaller than 1.7 and 2.0 cm, weighing 6.72 g fruit<sup>-1</sup> and represented by the wild genotypes Quialana and Miahuatlán; and medium-sized fruits, oblong shaped with equatorial and longitudinal diameters larger than 3.4 and 2.6 cm and fruit heavier than 18.40 g fruit<sup>-1</sup>, represented by the genotypes Jalieza, Vigallo and Ejutla. Fruits of the two groups weighed less than the husk tomatoes (P. ixocarpa) Sero Gordo cultivated in northern Mexico, which weigh 50.0 to 62.5 g (Bock et al., 1995). The results of our study are similar to those reported by Vargas-Ponce et al. (2011) for milpa race *Physalis*; they also indicate that these small green and purple fruits can vary 1 to 2 cm in diameter. Magaña-Lira et al. (2011) also report that wild Physalis fruits are highly valued by inhabitants of rural communities and demand high prices in regional markets.

The five wild genotypes have pH between 3.78 and 4.38; the genotype Miahuatlán had the highest value (4.38) and can be consumed fresh, like the red tomato (*Solanum lycopersicum* L.),

which that has pH around 4.4 (Akbudak, 2010). However, husk tomato is mostly consumed cooked for salsas and diverse dishes of Mexican cuisine. Regarding the pH, it has been found that *P. pubescens* fruits have a pH of 3.6 (El Sheikha et al., 2010), *P. philadelphica*, a high-yielding variety, has pH in the range of 3.78-3.88 (Jiménez-Santana et al., 2012), for *P. peruviana* the pH reported is 3.7 (Puente et al., 2011) and 3.79 - 3.86 (Ramadan, 2011), and the pH of *P. ixocarpa* varies from 4 to 4.5 (Gonzalez-Mendoza et al., 2011).

Citric acid is the most abundant organic acid in red tomatoes (Solanum lycopersicum L.). In this study, we found that the genotypes Vigallo and Ejutla contain higher levels of citric acid than the other varieties analyzed (1.25 and 1.65 g/100 g, respectively). The studies of Sheikha et al. (2010) on the genus Physalis show that P. pubescens L. is consumed as tropical fruit juice with a titratable acidity of  $1.43 \pm 0.08\%$ . Total soluble solids (TSS) are closely related to fruit flavor and the plant's accumulation of sugars (Jiménez-Santana et al., 2012). In this respect, the five analyzed genotypes had a range of 4.2 to 7.0 °Brix; the wild genotype Quialana had the highest TSS content  $(7.0 \pm 0.1 \text{ °Brix})$ . This characteristic of the genotype, as well as its titratable acidity, is comparable with that of cherry tomatoes (Solanum lycopersicum L.), which when harvested red ripe, has 6.07 °Brix and 0.67 g/100 g citric acid (Raffo et al., 2002). With the TSS data, we obtained the values of the flavor and ripeness index. If the flavor index is below 0.7, the tomato is considered to have an unpleasant taste (Navez et al., 1999), and the genotypes obtained a flavor index above 0.7 and even higher than the mean value reported for diverse red tomatoes (Solanum lycopersum L. var. ceraciforme), which was 0.85 (Hernández-Suárez et al., 2008b). Another parameter related to the flavor index is ripeness, which is usually a better predictor of acid taste intensity than TSS and acidity values alone. Acidity tends to decrease with fruit maturation, while TSS increase (Raffo et al., 2002). Average ripeness of tomatoes (Solanum lycopersicum L.) analyzed by Hernández et al. (2008) and Hernández et al. (2007b) was 9.4 ± 1.9, indicating that tomato ripeness is adequate for consumption. For

Table 1: Physalchemical parameters of the fruits of five wild genotypes of Physalis spp. collected in Oaxaca, Mexico.

Parameters	Wild genotypes of <i>Physalis</i> spp.					
	Quialana	Jalieza	Vigallo	Ejutla	Miahuatlán	
Weight (g•fruit <sup>-1</sup> )	6.72±3.26 b	16.04±2.85a	16.58±5.07a	20.74±2.51a	5.13±0.81b	
ED (cm)	2.05±0.045 b	3.34±0.37a	3.23±0.41a	3.49±0.23a	2.07±0.10b	
LD (cm)	1.97±0.41 bc	2.68±0.13a	2.60±0.24ab	2.71±0.24a	1.79±0.14c	
pH	4.11±0.04 b	4.02±0.9b	3.84±0.08c	3.78±0.01c	4.38±0.01a	
Acidity (gA•CA100gFF <sup>-1</sup> )	0.83±0.20 b	0.72±0.08b	1.25±0.09a	1.65±0.20a	0.83±0.17b	
TSS (ºBrix)	7.00±0.10 a	4.53±0.12d	5.37±0.06c	5.90±0.00b	4.16±0.58e	
Flavor index	1.26±0.09 bc	1.04±0.06dc	1.47±0.08b	1.84±0.18a	1.09±0.11dc	
Madurity index	8.82±2.23 a	6.33±0.56ab	4.29±0.28b	3.60±0.45b	5.18±1.08b	

Mean values with the same letters in each row are not statistically different (Tukey test; P <0.05); AC: Citric acid; TS: Total soluble solids; ED: Equatorial diameter; LD: Longitudinal diameter. ± SD: Standard Deviation.

*P. ixocarpa* spp., there are no data previously reported for the ripeness index. However, in this study we found that the genotype Quialana has a higher ripeness index ( $8.8 \pm 2.2$ ), related to its TSS content, which was also higher.

#### Color

Color analysis is frequently an important consideration to determine the effectiveness of a postharvest treatment in vegetables. Consumers are easily influenced by preconceived ideas of how a fruit or vegetable should look when sold in the market (McGuirre-Raymond, 1992). In the case of husk tomato, like chili peppers, the green color is a quality parameter for preparing salsas in Mexican cuisine (Bock et al., 1995). Of the color parameters (Table 2), a positive a\* component indicates a red-purple hue and a negative a\* indicates a blue-green hue, while a positive b\* component indicates yellow and a negative b\* indicates blue (McGuirre-Raymond, 1992). The fruit of the genotype Ejutla is less green than the other genotypes; Jalieza and Quialana have an intense green color, and Vigallo is purple. Moreover, if we take the parameters a\* and b\* together, we find that the fruit of the genotype Miahuatlán is purple, relative to the other fruits analyzed, possibly indicating higher anthocyanin concentrations (Chen et al., 2014). The L\* component indicates luminosity on a scale of 100 (white) to 0 (black). In general, the genotype Ejutla had a lower the L\* component than the rest of the analyzed husk tomato. No significant differences were found in hue or chroma. For these reasons, the genotype Ejutla is clearly different from the other varieties, with a color that is possibly less attractive for the consumers.

#### **Bromatological analysis**

The P. ixocarpa genotype "Sero Gordo" cultivated in northern Mexico has been reported to have a nutrient content of 0.9 % protein, 1.51% fat, 0.4% fiber and 4.36% carbohydrates (excluding dietetic fiber). In our study, we found that the fat content is below that reported previously (Table 3). In fiber content, the genotype Miahuatlán had a significantly higher (P < 0.05) content than the other four genotypes analyzed. Because of the health problems associated with food intake and non-transmissible diseases, it is important to determine caloric (energy) content to be able to make pertinent recommendations concerning its incorporation into human diets. The genotypes Quialana and Jalieza are significantly lower in caloric content per 100 g of produce. Bock et al. (1995) mention an average of 20.58 kcal/100 g in husk tomato samples (P. ixocarpa Brot.) collected in northern Mexico. The Mexican equivalent food system (Perez., et al., 2014) suggests a ration of vegetables equivalent to 25 kcal, which is equal to consuming five husk tomato. In this sense, consumption of 100 g of husk tomato would be equivalent to one ration of vegetables.

Total phenols and flavonoids and antioxidant capacity Plant phenolic contents contribute widely to their antioxidant potential, exhibited in the relation between phenolic compounds and elimination of free radicals (Zovko et al., 2010). In this study, we found significant variations in phenolic compounds among the five husk tomato genotypes analyzed (Fig. 2). The highest content of phenolic compounds was found in the genotype Vigallo  $(27.93 \pm 0.9 \text{ mg GA/g dry matter, DM})$ . The genotypes Ejutla and Miahuatlán had lower contents (8.67  $\pm$  0.16 and 7.88 ± 0.84 mg GA/g DM, respectively). González-Mendoza et al. (2011) evaluated the content of phenolic compounds in six varieties of P. ixocarpa; they found a maximum value of 9.65 mg GA/g DM. Also, the highest content of flavonoids (Fig. 2) was found in the genotype Vigallo (19.97  $\pm$  0.56 mg QE/g DM), which was significantly different from that of the other husk tomato. Jalieza was the genotype with the lowest flavonoid content  $(10.13 \pm 0.6 \text{ mg QE/g DM})$ . The content of anthocyanins in the five evaluated genotypes did not have a definite trend but was significantly higher in the fruits of the genotypes Miahuatlán and Vigallo, coinciding with intense purple coloring of the fruits (Fig. 2).

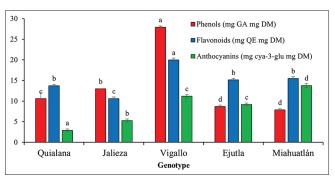


Fig 2. Content of phenols, flavonoids and anthocyanins in the fruit of five wild genotypes of *Physalis spp.* AG: Acid Galic; DM: Dry matter; QE: Quercetine equivalents. The different letters indicate significant differences for  $P \le 0.05$  based on Tukey test.

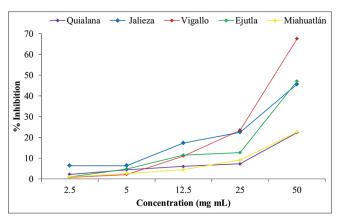


Fig 3. Antioxidant activity of the fruits of five wild genotypes of *Physalis* spp.

Table 2: Color parameters of five wild genotypes of <i>Phyaslis</i> spp. collected in Oaxaca, Mexico.

Parameters		Wild genotypes of <i>Physalis</i> spp.					
	Quialana	Jalieza	Vigallo	Ejutla	Miahuatlán		
a*	-2.99±6.01ab	-10.71±4.54b	-4.25±5.83b	11.07±1.51a	-8.08±6.66b		
b*	21.19±5.84a	26.53±5.43a	17.41±7.15a	-1.28±2.03b	29.10±4.23a		
L	58.46±6.85a	54.71±5.56a	50.88±6.87a	42.47±3.49b	50.28±2.76a		
Matiz	-0.72±0.91	-1.17±0.14	0.23±1.27	-0.83±0.53	-1.27±0.23		
Croma	26.71±2.01	25.43±0.95	22.06±8.00	18.48±2.84	29.35±2.90		

Mean values with the same letters in each row are not statistically different (Tukey test; P <0.05). ± SD: Standard Deviation.

Parameters		wild genotypes of <i>Physalis</i> spp.					
	Quialana	Jalieza	Vigallo	Ejutla	Miahuatlán		
Humidity (%)	90.84±1.6ab	93.23±0.32a	91.02±1.30ab	89.92±0.44b	86.80±1.18c		
Ash (%)	0.10±0.01b	0.04±0.00b	0.05±0.01b	0.05±0.01b	0.26±0.16a		
Carbohydrates (%)	4.91±0.73bc	4.18±0.25c	5.88±0.83ab	6.89±0.36a	6.98±0.63a		
Total fiber (%)	2.74±0.64b	1.54±0.04c	1.72±0.31c	1.96±0.03bc	4.27±0.42a		
Protein (%)	0.88±0.15b	0.71±0.04b	0.96±0.11a	0.83±0.03b	1.18±0.12a		
Fat (%)	0.53±0.09a	0.30±0.01b	0.38±0.04ab	0.36±0.01ab	0.50±0.12a		
Caloric value (kcal)	27.95±4.28bc	22.28±1.22c	30.78±4.11abc	34.04±1.70ab	37.16±3.99a		

Mean values with the same letters in each row are not statistically different (Tukey test; P <0.05). ± SD: Standard Deviation.

DPPH is a fast, sensitive method of evaluating antioxidant capacity of plant extracts. It is based on decoloring of DPPH in presence of antioxidants in the analyzed samples. According to Shahidi and Wanasundara (1992) antioxidant power is due mainly to the redox nature of phenolic compounds. The results we obtained confirm this (Fig. 3). The total contents of phenols and flavonoids was significantly (P < 0.05) higher in the genotype Vigallo and consequently, at a concentration of 50 mg/mL, its activity in trapping free radicals is high (67.5 % inhibition), relative to the other genotypes.

Diverse epidemiological studies indicate a beneficial effect of consuming red tomatoes (Solanum lycopersicum L.) on dyslipidemias (Blum et al., 2006; Tsitsimpikou et al., 2014), chronic low-grade inflammation associated with obesity (Huang et al., 2007; Palozza et al., 2011), endothelial dysfunction caused by cardiovascular risk factors and levels of glucose and insulin (Tsitsimpikou, et al., 2014). This protective effect is due to antioxidant contents that could contribute to inhibit an oxidative process (Sies and Stahl, 1998). For Physalis ixocarpa, no detailed information exists on the quality and content of bioactive compounds in the different genotypes even when demand is growing in the United States and Canada, making it the fifth most important vegetable of Mexico. This study, then, is a good approximation for future research and for revaluating consumption of husk tomato for new uses in Mexican cuisine.

# **CONCLUSIONS**

The fruits of five wild *Physalis* spp. genotypes showed broad heterogeneity in the studied parameters, indicating

the wealth and diversity of this species. Because of their physicochemical characteristics, such as total soluble solids and acidity, the genotypes Quialana and Miahuatlán are attractive for fresh consumption and suitable for processing in new functional foods. Nutritionally, the genotypes Vigallo, Ejutla and Miahuatlán had higher caloric and carbohydrate contents, as well as red to purple colors, while the Vigallo genotype reached the highest values of the bioactive compounds phenols and flavonoids and of antioxidant capacity. It is important to study and revalue the wild *Physalis* spp. fruits, which can be an option that is beneficial to human health and nutrition.

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## Author's contributions

Gabino Alberto Martinez-Gutierrez and Isidro Morales conducted the experiments and wrote the first manuscript, Aleyda Pérez-Herrera designed the experiments and participated in the laboratory work and the data analysis, Aleyda Pérez-Herrera, Marco Antonio Sánchez-Medina and Cirenio-Escamirosa Tinoco, review the manuscript, and were responsible for the final review of the manuscript. All authors read and approved the final manuscript.

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