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ORIGINAL PAPER

Physicochemical, nutritional and antioxidant characterization of three vegetables (*Amaranthus hybridus* L., *Chenopodium berlandieri* L., *Portulaca oleracea* L.) as potential sources of phytochemicals and bioactive compounds

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Abstract

The aim of this work was the characterization of three endemic plant species [quelite cenizo and quintonil (*quelites*), and purslane], from Mexico, in terms of their physicochemical, nutritional and antioxidant properties. These species are highly used by rural populations in their local dishes. Nevertheless, scarce information exists about their nutritional and functional properties. The experimental included physicochemical properties, nutrimental composition, qualitative and quantitative analysis of antioxidants by HPLC. The results showed that purslane was the sample with the highest concentration of total carotenoids (2.85 mg/g DW), and with the highest antioxidant activity by DPPH and FRAP methods. Quelite cenizo and purslane displayed similar values for antioxidant activity by ABTS method, nevertheless, the former showed higher values for total phenolics (10.24 mg GAE/g DW) and flavonoids (17.57 mg QE/g DW) content than the latter. Finally, quintonil showed similar results for total chlorophyll (70.50 mg/g DW) and protein (3.65 g/100 g FW) to those found in quelite cenizo. The contents of Fe, Ca, and Mg were higher in quintonil, meanwhile the contents of P, K and Zn were higher in quelite cenizo. The analysis by HPLC also showed the presence of ferulic, and chlorogenic acids as the most abundant phenolic acids, meanwhile rutin and phloridzin were the main flavonoids in the *quelites*, displaying higher contents than in the purslane. Therefore, it can be assumed that these samples possess high nutritional quality and could be considered for their use in strategies to improve food security in rural populations.

Keywords *Amaranthus hybridus* L. · *Chenopodium berlandieri* L. · *Portulaca oleracea* L. · Antioxidant compounds · Food safety

Introduction

There are several crops of plants for human consumption with a good resistance to stressors, whose production cost is minimal, so they have a high potential to be commercialized and consumed in rural populations, contributing to improve the living conditions of the region. Among these crops, the Amaranthaceae and Portulacaceae families can be highlighted.

Portulaca oleracea (Purslane) is an edible plant growing wild with a cosmopolitan distribution, which is very adaptable to different environments. Purslane has been proposed in this research not only for its frequency in consumption but for the numerous phytochemicals and fatty acids such as α-Linolenic acid (omega-3), β-carotenes, α-tocopherols,

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flavonoids (quercetin) and phenolic alkaloids identified in their stems and leaves [1, 2] which promote antioxidant activity by decreasing H_2O_2, and provide neuroprotective effects against environmental neurotoxic agents [3]. On the other hand, the "*quelites*" studied in this work, [including *Chenopodium berlandieri* L. ("quelite cenizo") and *Amaranthus hybridus* L. ("quintonil")] refers to young stems, suckers or seedlings of edible plants growing wild; this term is only used in Mexico. The leaves of "*quelites*" have shown significant contents of protein and inorganic nutrients (Ca, Fe, Mg), and considerable amounts of fiber as well as the presence of terpenoids and flavonoids such as quercetin and kaempferol, which have been related with a high antioxidant capacity and properties against cytotoxic processes [4–7].

Some studies have been conducted about *Portulaca oleracea* L. and *Amaranthus hybridus* L. from other countries, however, as far as we know, there are no reports about *Chenopodium berlandieri* L. included in this work, which are endemic of rural areas of Mexico. In this way, the incorporation of any of these plants, within the diet, constitutes an economical, accessible and gastronomically acceptable source, with high nutritional potential, leading to the implementation of a sustainable diet in marginalized rural populations. Therefore, the aim of this research was to determine the nutritional composition, the content of the main bioactive compounds, the antioxidant activity by DPPH, ABTS and FRAP and the identification of major phenols and flavonoids in *Portulaca oleracea* L., *Amaranthus hybridus* L. and *Chenopodium berlandieri* L. by HPLC.

Materials and methods

Chemicals

2,2′-Diphenyl-1-picrylhydrazyl (DPPH), 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), Trolox (6-hydroxy-2,5,8- tetramethylchroman-2-carboxylic acid), TPTZ (2,4,6-Tris (2-pyridyl)-1,3,5-triazine), ferric chloride, Folin–Ciocalteau reagent, α-amylase, protease, and amyloglucosidase were purchased from Sigma-Aldrich (St. Louis Missouri, USA). Sodium hydroxide, sodium carbonate, aluminum chloride, acetone, ascorbic acid, sodium acetate anhydrous, glacial acetic acid, hydrochloric acid, sulfuric acid, potassium sulfate, copper sulfate pentahydrate, boric acid, sodium phosphate monobasic, sodium phosphate dibasic, sodium arsenate heptahydrate, sodium bicarbonate, ammonium molybdate tetrahydrate, sodium sulfate anhydrous and potassium sodium tartrate, which were purchased from J.T. Baker (Mallinckrodt, USA); gallic acid, quercetin, methanol and ethanol were purchased from Meyer Chemical (Tlahuac, Mexico), and potassium persulfate was acquired from Reasol laboratories (State of Mexico, Mexico). Trichloroacetic acid was used from Macron Fine Chemicals (Pennsylvania, USA). Sodium nitrite and petroleum ether purchased from Monterrey Chemical Products (Monterrey, Mexico); meta-phosphoric acid purchased from Merck (Danstadt, Germany) was used. Distilled water was used in the performance of all the experiments, and all the reagents used were of analytical grade.

Plant material

The samples of *Portulaca oleracea* L. (Purslane), *Amaranthus hybridus* L. (Quintonil) and *Chenopodium berlandieri* L. (Quelite cenizo) were collected from Tulancingo de Bravo, Hidalgo, Mexico (latitude: 20°4′53″N, longitude: 98°22′2″W). The roots were removed and the aerial parts (leaves) were used. All the samples were collected during the month of February 2017. The samples free of rot and pests were cleaned, washed, rinsed, and were allowed to drain 1 h at room temperature (25 °C) before the freezing process. The samples were stored at − 76 °C in an ultra freezer (Thermo-Scientific, 703, Outside, USA) per 24 h before its subsequent lyophilization (Labconco, 7948000, Missouri, USA).

Samples preparation

The samples used for the determination of antioxidant compounds and antioxidant activity were prepared as follows: 0.1 g of lyophilized sample was mixed in 10 mL of solvent and sonicated per 15 min at a frequency of 40 kHz at 25 °C (Ultrasonicator LSS, 32V118A, China), followed by storage at 4 °C per 24 h and centrifuged at 16,500×*g* per 10 min at 5 °C (Centrifuge Thermo-Scientific, ST 16R, Germany). The supernatant was used for the subsequent analyses.

Physicochemical properties

The total soluble solids (TSS), pH and titratable acidity (TA) were determined according to the AOAC [8] (942.15). A digital refractometer (Atago-Palette, PR-101, Tokyo, Japan) was used for the TSS determination, the results were expressed in ºBrix. The pH measurements were made with a digital pH meter (Hanna Instruments, HI 2211, Woonsocket, RI, USA). The results of the TA were reported as citric acid percent (%).

Color

Color measurements were conducted in a Minolta colorimeter (Minolta, CM-508d, Osaka, Japan). The color parameters L*, a*, b* were obtained. The values a* (green–red) and b* (yellow–blue) were used to calculate the Hue angle

(h°) and the chroma value (*C*). For the calculation of h° and *C*, the following formulas were used:

$$h^\circ = \tan^{-1}\left(b^*/a^*\right)$$

$$C = \sqrt{a^{*2} + b^{*2}}$$

Chlorophyll content

Chlorophyll *a, b*, and total (Chl *a*; Chl *b*; Chl t) was determined according to Witham et al. [9] with slight modifications. The solvent used for the sample preparation was 80% acetone. The absorbance of the sample was spectrophotometrically measured at 645 and 663 nm (UV–Vis spectrophotometer, Jenway, 6715, USA). The results were reported in mg of chlorophyll per g of dry weight (mg/g DW).

Total carotenoids

The red (RC = capsanthin and capsorubin) and yellow (YC = β-carotene, β-cryptoxanthin, zeaxanthin) isochromatic fractions of the total carotenoids were evaluated according to Hornero-Mendez and Minguez-Mosquera [10]. The solvent used for the sample preparation was acetone. The absorbance of the samples was read spectrophotometrically at 472 and 508 nm respectively. Measurements of red and yellow carotenoids were expressed as mg/g DW.

Total phenols

The total phenols content was determined by the Folin–Ciocalteu method described by Singleton and Rossi [11]. The solvent used for the sample preparation was 80% ethanol. Briefly, 0.5 mL of the sample was mixed with 0.5 mL of 50% Folin–Ciocalteu reagent in water. The mixture was left to stand for 7 min, after which 1.5 mL of 7.5% sodium carbonate was added, and this mixture was left to react in the darkness for 60 min. The absorbance was measured at 725 nm using a UV–Vis spectrophotometer (Jenway, 6715, USA). The results were expressed as mg of gallic acid equivalents (mg GAE)/g DW.

Total flavonoids

The flavonoids were determined according to Rosales et al. [12], using the same solvents described above for the samples preparation. Briefly, 2 mL of the sample was mixed with 1.5 mL of Sodium nitrite and 2 mL of water, this mixture was left to stand for 5 min in the darkness. Aluminum trichloride (1.5 mL) and 1 mL of sodium hydroxide were added to the mixture, and it was left to stand per 20 min more. The absorbance was read at 415 nm using a UV–Vis spectrophotometer (Jenway, 6715, USA). The results were reported as mg of quercetin equivalents (mg QE)/g DW.

Ascorbic acid

The content of ascorbic acid was determined according to Dürüst et al. [13]. The solvent used for the sample preparation was 10% trichloroacetic acid in water. Briefly, 0.2 mL of sample was mixed with Folin–Ciocalteu reagent diluted 1:10 in water. The mixture was left to stand for 10 min at 4 °C. The absorbance was measured at 760 nm (Jenway, 6715, USA). The results were expressed as mg of ascorbic acid (mg AA)/g DW.

Evaluation of antioxidant activity

The antioxidant activity was evaluated in the samples using the DPPH, ABTS and FRAP methods. The solvent used for the samples preparation was 80% ethanol. The results were expressed as μM of Trolox equivalents (μM TE)/g DW.

The DPPH (2,2′-diphenyl-1-picrylhydrazyl) method, was performed according to Brand-Williams et al. [14]. Briefly, 0.3 mL of sample were mixed with 2.7 mL of DPPH (6×10^{-5} M). This mixture was left to stand in the darkness for 1 h at 4 °C, the absorbance was read at 517 nm (A_{517}).

The ABTS [2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)] method was performed according to Re et al. [15]. Briefly, the ABTS radical was produced by mixing a solution of 7 mM ABTS with 10 mL of 2.45 mM potassium persulfate in complete darkness during 16 h at constant stirring. The ABTS radical was diluted in 80% ethanol for the samples until an absorbance value of 0.7 ± 0.02 at 734 nm was achieved. It was taken 3.9 mL of ABTS radical diluted and mixed with 0.1 mL of sample, and let to stand for 6 min in the darkness. The absorbance of this mixture was measured at 734 nm.

The analysis of the ferric reducing antioxidant power (FRAP), was performed according to Benzie and Strain [16]. Briefly, the FRAP reagent was prepared mixing acetate buffer (0.3 M; pH 3.6), TPTZ (2,4,6-Tris (2-pyridyl)-1,3,5-triazine) (10 mM) and iron chloride hexahydrate (20 mM) in a 10:1:1 ratio. Then, 2.25 mL of FRAP reagent was mixed with 0.3 mL of sample and incubated for 30 min at room temperature in the darkness. The absorbance was read at 593 nm.

Nutritional composition

The official methods of the AOAC [8] were used for the analysis of moisture (925.09), fat (983.23), protein (950.48), ash (930.05) and crude fiber (985.29, 993.21); total carbohydrates were calculated by difference. The energy was

determined according to Chahdoura et al. [17], using the following equation:

$$Energy,\ \frac{\text{kJ}}{100\ \text{g FW}} = \left[(4)\left(g_{protein} + g_{carbohydrates}\right) + (2)\left(g_{fiber}\right) + (9)(g_{fat})\right][4.184]$$

The results for moisture, fat, protein, carbohydrates, ash and crude fiber were expressed in g/100 g FW, and for energy in kJ/100 g FW.

Dietary fiber: soluble fiber and insoluble fiber

Soluble fiber (SDF) and insoluble fiber (IDF) were determined according to the enzymatic-gravimetric method (993.19 and 991.42) of the AOAC [8]. The enzymes used were obtained from the dietary fiber test kit (Sigma-Aldrich, Buchs, Switzerland). The results were expressed in g/100 g FW.

Mineral composition (macro and micro-elements)

The analysis of minerals was determined in lyophilized samples according to the AOAC [8]. The macro and micro-elements were determined after $HClO_4/HNO_3$ digestion. The analysis was performed by photocolorimetry by molybdovanadate reduction (P), photocolorimetry with Azomethine-H (B), flame emission spectrophotometry (K) and atomic absorption spectrophotometry (Ca, Mg, Fe, Cu, Zn, and Mn) (inductively coupled plasma atomic emission spectroscopy analyzer, GBC, 932AA, USA). The results were expressed in mg/100 g DW.

Identification and quantification of phenols and flavonoids by HPLC

Phenols and flavonoids were determined according to Aguiñiga-Sánchez et al. [18]. The content and type of the metabolites in the extract were identified by high-performance liquid chromatography (HPLC-DAD, Agilent Technologies 1100, USA). For the flavonoids, the analyses were performed on a Hypersil ODS (125 × 40 mm) Hewlett Packard Column with a gradient of (A) H_2O at pH 2.5 with TFA (trifluoroacetic acid) and (B) ACN (acetonitrile) and the following parameters: flow, 1 mL/min; temperature, 30 °C; variable injection volume; and analysis time, 25 min. The standards used were rutin, phloridzin, myricetin, quercetin, naringenin, apigenin, phloretin, and galangin. For phenolic acid, a nucleosil column (125 × 4.0 mm) from Macherey-Nagel was used with a gradient of (A) H_2O at pH 2.5 with TFA (trifluoroacetic acid) and (B) ACN (acetonitrile). Other experimental parameters included: flow, 1 mL/min; temperature, 30 °C; variable injection volume; and analysis time, 25 min. Caffeic, gallic, chlorogenic, vanillic, *p*-hydroxybenzoic, *p*-coumaric, ferulic and syringic acids were used as the standards. The results were expressed in µg/g DW.

Statistical analysis

All results were reported as mean + SD, each with three replicates (n = 3). The software JMP.5.0.1 (A Business Unit of SAS, Statistics Analysis System, v. 9.0) was used for statistical analyses. Comparisons among treatment means were tested for significance, using analysis of variance (ANOVA) and the Tukey test at a level of significance $P < 0.05$.

Results and discussion

Physicochemical properties

Table 1 shows the results about physicochemical properties. Quelite cenizo showed a TSS content significantly higher than those found in the other vegetables ($P < 0.05$). Regarding pH and titratable acidity, purslane displayed the lowest values, meanwhile quelite cenizo and quintonil showed the highest values. The highest value of TA was found in quelite cenizo, and the lowest values were observed in purslane and quintonil, without significant differences between them. The results found in these parameters are commonly used to characterize the quality and to provide an idea of the shelf life of these plant materials. For example, the higher TSS values found in quelite cenizo and quintonil can be related to a lower availability of water to carry on rotting processes.

Color

Table 1 shows L*, a*, b*, *C* and h° values, for each sample. The L* (luminosity) and b* [from blueness (−) to yellowness (+)] values were positive in all samples, meanwhile, a* [from greenness (−) to redness (+)] value presented negative results. The green coloration is due to the presence of chlorophyll in the leaves of the vegetables. The *C* value saturation (color purity) was 18.71–24.87 for the samples. The variations in chromatic coordinates were expected since the analyzed samples were of different species. The obtained data can be related to the quality and freshness of the samples, and they are commonly suggested as a quality index in vegetables [19].

Table 1 Physicochemical properties in *P. oleracea* L., *C. berlandieri* L., and *A. hybridus* L. samples

Physicochemical properties	Sample		
	Purslane	Quelite cenizo	Quintonil
TSS (°Brix)	4.73 ± 0.21^{c}	8.13 ± 0.06^{a}	5.80 ± 0.40^{b}
pH	4.29 ± 0.16^{b}	6.33 ± 0.02^{a}	6.40 ± 0.26^{a}
TA (% citric acid or g citric acid/100 mL)	0.24 ± 0.02^{b}	0.65 ± 0.02^{a}	0.26 ± 0.02^{b}
Color			
*L**	44.64 ± 0.49^{a}	36.85 ± 1.80^{b}	37.95 ± 1.81^{b}
*a**	-7.59 ± 1.82^{a}	-10.27 ± 1.09^{a}	-7.56 ± 0.23^{a}
*b**	19.44 ± 1.68^{a}	22.65 ± 2.66^{a}	17.09 ± 3.08^{a}
C	20.89 ± 2.23^{a}	24.87 ± 2.87^{a}	18.71 ± 2.92^{a}
h (°)	111.11 ± 3.01^{a}	114.41 ± 0.25^{a}	114.18 ± 3.02^{a}

TSS (°Brix) total soluble solids, *TA* (%) titratable acidity, *C* chroma value, °*h* hue angle expressed in degrees

The observed data express values of the mean ± SD ($n = 3$). The values with the same letter within each row are the same according to the Tukey test at $p < 0.05$

Table 2 Antioxidant compounds in *P. oleracea* L., *C. berlandieri* L. and *A. hybridus* L. samples

Antioxidant compounds	Sample		
	Purslane	Quelite cenizo	Quintonil
Carotenoids			
RC	1.39 ± 0.07^{a}	0.23 ± 0.01^{b}	0.14 ± 0.02^{b}
YC	1.45 ± 0.04^{a}	1.11 ± 0.02^{b}	0.55 ± 0.01^{c}
TC	2.85 ± 0.03^{a}	1.34 ± 0.01^{b}	0.70 ± 0.01^{c}
Chlorophyll			
Chl *a*	23.22 ± 1.04^{b}	19.40 ± 0.11^{c}	25.27 ± 0.90^{a}
Chl *b*	43.00 ± 0.13^{b}	46.83 ± 0.90^{a}	45.22 ± 0.80^{a}
Chl t	66.22 ± 0.39^{b}	66.23 ± 0.78^{b}	70.49 ± 1.05^{a}
TPC	10.06 ± 0.26^{a}	10.24 ± 0.31^{a}	7.35 ± 0.28^{b}
TFC	8.97 ± 0.18^{c}	17.57 ± 0.14^{a}	15.29 ± 0.10^{b}
AA	1.88 ± 0.03^{a}	0.03 ± 0.01^{b}	0.12 ± 0.07^{b}

RC red carotenoids, *YC* yellow carotenoids, *TC* total carotenoids. The total carotenoids result from the sum of red and yellow carotenoids. Results were expressed as mg/g of dry weight (DW). *Chl a* chlorophyll *a, Chl b* chlorophyll *b, Chl t* total chlorophyll. Results were expressed as mg/g of dry weight (DW). *TPC* total phenols content. Results were expressed as mg gallic acid equivalents (mg GAE)/g of dry weight (DW). *TFC* total flavonoids content. Results were expressed as mg quercetin equivalents (mg QE)/g of dry weight (DW). *AA* ascorbic acid. Results were expressed as mg of ascorbic acid (mg AA)/g of dry weight (DW)

The data express values of the mean ± SD ($n = 3$). The values with the same letter within each row are the same according to the Tukey test at $p < 0.05$

Chlorophyll content

The results about chlorophyll content in green leaves are presented in Table 2. Significant differences were observed in the chlorophyll *a* content between quelite cenizo, quintonil, and purslane. The total chlorophyll content was higher in quintonil. Lower contents of chlorophyll have been reported in *Amaranthus hypochondriacus, Amaranthus tricolor* [20], *Chenopodium quinoa* [21], and *Portulaca oleracea* [22] in comparison with our findings.

These results are interesting from the point of view of the nutritional and antioxidant evaluation since it has been reported that the presence of chlorophyll in green leaf plants reduces reactive oxygen species. Also, all the chlorophyll compounds act through similar anti-carcinogenic mechanisms, with variable degrees of strength [23].

Total carotenoids, total phenols, total flavonoids and ascorbic acid content

Purslane showed significantly higher total carotenoids content and lower total chlorophyll content when it was compared to quintonil (Table 2). These results are probably because the chlorophyll is degraded by seasonal periods, and due to an increase in the carotenoids biosynthesis by mechanisms of adaptation reported in plants [24]. Similar carotenoids contents have been reported for *Portulaca oleracea* [25], *Chenopodium album* [6] and *Amaranthus* spp. [26].

Regarding the TPC, purslane and quelite cenizo showed the highest values, without statistically significant differences between them (Table 2). TPC values are dependent on the samples collection location, plant growth stages, and plant age. TPC is increased in developing stages of growth and decrease during maturity [27]. Some studies have reported lower TPC contents for *Chenopodium* spp. [7], *Portulaca oleracea* L. [25], and *Amaranthus hybridus* L. [28].

On the other hand, quelite cenizo showed the highest concentration of total flavonoids content (TFC), followed by quintonil (Table 2). These results are in accordance

with those reported by other researchers [6, 20] which have reported that quelites are characterized by presenting a wide variety of flavonoids in their leaves. As it was explained above, the environmental and physiological factors influence on the phenolic compounds and flavonoids composition in plants. According to Alves et al. [29], the TPC and TFC values may vary between species and within species; where the main factors, are the genetic variability and the edaphoclimatic conditions.

With respect to the ascorbic acid (AA) concentration, purslane was the sample showing the highest values (Table 2); meanwhile quelite cenizo and quintonil are negligible sources of ascorbic acid when compared to other sources of this compound. Uddin et al. [27], found similar values in different growing stages of purslane, meanwhile, Funke [30] and Gqaza et al. [31], reported higher values in *Amaranthus* spp. and in *Chenopodium album* L. in comparison with the species studied in this work; these results suggest that the contents of ascorbic acid and others antioxidant compounds in the quelites are due to the environmental and physiological factors in the plants collected [32, 33]. However, despite the fact that the *Amaranthus* and *Chenopodium* genera showed lower AA contents in this study, if a comparison is made with the recommended daily intake (RDI) of ascorbic acid, which is 60 mg/day [34], a portion of *Amaranthus hybridus* L. (9 mg AA/100 g FW) or *Chenopodium berlandieri* L. (2.1 mg AA/100 g FW), would be equivalent to 15% and 3.5% of the RDI respectively. These values, if combined within a correct diet, represent an alternative, less expensive and more accessible source of ascorbic acid.

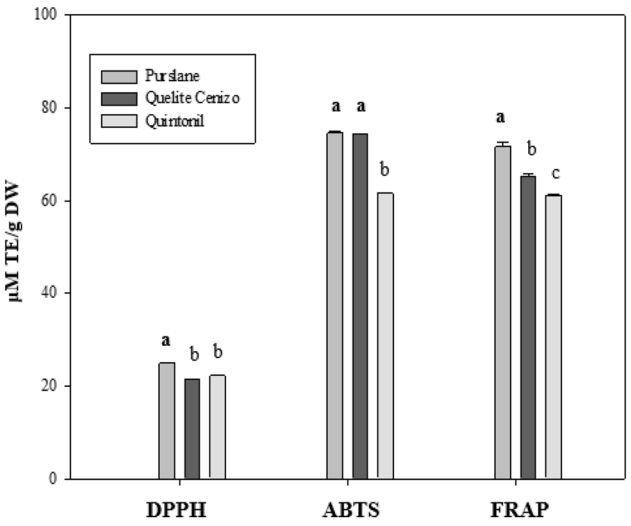

Fig. 1 Antioxidant properties (DPPH, ABTS and FRAP) in *P. oleracea* L., *C. berlandieri* L., and *A. hybridus* L. samples. The results were expressed as µM of Trolox equivalents/g of dry weight (DW). The values with different letters mean statistically significant difference according to the Tukey test ($p \leq 0.05$)

Antioxidant activity

Antioxidant activity values were dependent on the assay performed (Fig. 1). DPPH assay showed values ranged from 21.42 to 24.85 µM TE/g DW. Meanwhile, ABTS showed values ranged from 61.61 to 74.60 µM TE/g DW. Results about the ABTS assay displayed higher values than those obtained with the DPPH for all the samples studied, this is in accordance with Floegel et al. [35] and Piscopo et al. [36], which have reported that the ABTS assay is better for a variety of food matrixes, particularly fruits, vegetables, and oil. Regarding the FRAP assay, results were ranged from 61.17 to 71.57 µM TE/g DW. Purslane showed the highest antioxidant activity among the three assays performed, and it was significantly different from the other samples, except for ABTS, where it was not significantly different from quelite cenizo.

According to our findings, the samples with the highest TPC, showed the highest antioxidant activity, independently of the assay performed, either DPPH or ABTS, although they did not present significant correlation according to Pearson's test (data not shown). Additionally, no correlation was found between other antioxidant compounds (total carotenoids, chlorophyll, and ascorbic acid) and the DPPH, ABTS, or FRAP assays.

Nutritional composition

The nutritional composition of samples is listed in Table 3. The moisture ranged between 90.65% for quelite cenizo to 92.66% for purslane. This result was the expected, since purslane is a plant with succulent leaves, presenting big vacuoles containing higher amounts of water [37, 38] when compared with the other samples studied. With respect to fat, the purslane showed the highest results, which are in the range reported by Oliveira et al. [39] for different varieties of *Portulaca oleracea* leaves. Meanwhile, quintonil and quelite cenizo displayed the highest protein content, which was expected since the leaves of the genus *Chenopodium* and *Amaranthus* have been recognized as important sources of the vegetal protein [6, 40]. Regard to the ash content, quelite cenizo and quintonil showed also the highest values. The *quelites* showed the highest crude fiber content, being able to be considered as a good source of fiber. Purslane and quelite cenizo showed the highest total carbohydrates content. The energy values were not higher than 112 kJ/100 g FW, which were found in quelite cenizo, whose main supply of energy was protein.

Table 3 Nutritional composition in *P. oleracea* L., *C. berlandieri* L. and *A. hybridus* L. samples

Nutritional parameters	Sample		
	Purslane	Quelite cenizo	Quintonil
Moisture	92.66 ± 0.10^{a}	90.65 ± 0.10^{c}	91.43 ± 0.10^{b}
Fat	0.32 ± 0.03^{a}	0.20 ± 0.02^{b}	0.15 ± 0.01^{c}
Protein	1.81 ± 0.03^{c}	3.45 ± 0.05^{b}	3.65 ± 0.03^{a}
Ash	1.91 ± 0.01^{c}	2.34 ± 0.03^{a}	2.21 ± 0.03^{b}
Crude fiber	0.98 ± 0.03^{b}	1.15 ± 0.01^{a}	1.18 ± 0.02^{a}
Available carbohydrates	2.32 ± 0.05^{a}	2.21 ± 0.20^{a}	1.38 ± 0.10^{b}
Energy	89.39 ± 0.09^{c}	111.80 ± 1.00^{a}	99.96 ± 1.12^{b}
Dietary fiber			
Insoluble dietary fiber	2.34 ± 0.21^{a}	2.36 ± 0.08^{a}	2.41 ± 0.05^{a}
Soluble dietary fiber	0.68 ± 0.25^{a}	0.81 ± 0.08^{a}	0.58 ± 0.11^{a}
Total dietary fiber	3.02 ± 0.08^{a}	3.17 ± 0.10^{a}	2.99 ± 0.07^{a}

The data express values of the mean ± SD ($n = 3$). In each row, different letters mean statistically significant difference according to the Tukey test ($p \leq 0.05$). Results were expressed as kJ/100 g of fresh weight (FW) for the energy and g/100 g of FW for the other parameters

Dietary fiber: soluble fiber and insoluble fiber

The soluble, insoluble and total dietary fiber content was evaluated in the aerial parts (Table 3). The results showed no statistically significant differences among samples.

According to the ADA [41], the recommended daily intakes (RDI) of fiber, should range from 14 g per each 1000 kcal, or 25 g for adult women and 38 g for adult men to show protective effects against cardiovascular diseases. Purslane provides 12% and 8%, quelite cenizo 12.7% and 8.3% and quintonil provides 12% and 8% out of the RDI of total dietary fiber for adult women and men respectively.

The soluble dietary fiber contents ranged from 0.58 g/100 g FW for quintonil to 0.81 g/100 g FW for quelite cenizo, without statistically significant differences among them. These results are comparable with other important sources of soluble dietary fiber as kiwi fruit, mango and plums [42], nevertheless, these fruits are less accessible for some rural populations, mainly due to their relatively higher costs. It is well known that insoluble fiber has hypolipidemic effects and cardioprotective benefits associated with its consumption [43], therefore, it can be assumed that the samples studied in this work represent inexpensive sources of this component.

On the other hand, insoluble fiber fractions ranged from 2.34 g/100 g FW for purslane to 2.41 g/100 g FW for quintonil, without statistically significant differences among the samples. As it has been reported by Dhingra et al. [42], most of the plant materials contain a higher proportion of insoluble dietary fiber when compared with the soluble fraction. It has been demonstrated that insoluble dietary fiber retards the utilization and absorption of carbohydrates, and helps to control the postprandial serum glucose level [44]. Thus, the potential ability of these vegetables in lowering postprandial serum glucose level and reducing calories level would be useful as important insoluble dietary fiber sources in functional food applications.

Mineral composition

The mineral composition is shown in Table 4. Quintonil showed the highest contents of iron, calcium, and magnesium when compared with the other species. On the other hand, quelite cenizo showed the highest content of phosphorous, potassium and zinc. However, the purslane showed the highest concentration of copper, manganese, and boron among the three species studied. These results agree with the literature, which reports the presence of calcium, potassium, magnesium and other minerals in leaves of different species of *quelites* such as *Chenopodium* spp. and *Amaranthus* spp., and in leaves of *P. oleracea* [45]. The evidence reports that minerals and trace elements play an important role in the regulation of certain body functions. Macro and micro-elements are found in the structure of the teeth (Ca and P) and bones (Ca, Mg, Mn, P and B), whereas most micro-elements (Cu, Fe, Mn, Mg and Zn) play a vital role as a structural part in many enzymes. Also, microelements have key roles in the formation of the erythrocyte cells (Fe), and macro minerals such as Ca and K have a high potential

Table 4 Mineral composition of *P. oleracea* L., *C. berlandieri* L. and *A. hybridus* L.

	Microelements (mg/100 g DW)					Macroelements (mg/100 g DW)			
	Fe	Cu	Mn	Zn	B	P	Ca	Mg	K
P	14.05 ± 0.004^{b}	1.93 ± 0.003^{a}	5.15 ± 0.030^{a}	5.79 ± 0.010^{b}	6.20 ± 0.010^{a}	360 ± 0.010^{b}	900 ± 0.010^{c}	300 ± 0.001^{b}	6920 ± 0.030^{b}
QC	10.21 ± 0.010^{c}	1.60 ± 0.003^{b}	5.01 ± 0.040^{b}	6.72 ± 0.030^{a}	6.02 ± 0.020^{b}	480 ± 0.020^{a}	960 ± 0.004^{b}	280 ± 0.004^{c}	8420 ± 0.001^{a}
Q	65.57 ± 0.005^{a}	1.27 ± 0.010^{c}	4.55 ± 0.020^{c}	4.64 ± 0.030^{c}	5.42 ± 0.010^{c}	240 ± 0.010^{c}	1980 ± 0.020^{a}	740 ± 0.030^{a}	4610 ± 0.003^{c}

P purslane, *QC* quelite cenizo, *Q* quintonil

The data express values of the mean ± SD ($n = 3$). In each column, different letters mean statistically significant difference according to the Tukey test ($p \leq 0.05$). Results were expressed as mg/100 g of dry weight (DW) for the microelements and macroelements

to control blood pressure. The minerals are also involved in immune (Ca, Mg, Cu and Zn), and brain (Mn) systems [46]. Thus, the analyzed samples are important sources of macro and micro-elements contributing in diverse functionalities and potentials in the body's metabolism and homeostasis.

HPLC analyses

The phenolic acids and flavonoids identified by HPLC are listed in Table 5. The highest content of phenolic acids was found in quintonil, according to the total phenols index (TPI), meanwhile, as the total flavonoids index (TFI) indicates, the highest content of flavonoids was found in quelite cenizo. The analysis by HPLC revealed the presence of higher concentrations of gallic, caffeic, chlorogenic, vanillic and syringic acids in quelite cenizo. Meanwhile, quintonil showed higher contents of *p*-hydroxybenzoic and ferulic acids. On the other hand, the purslane presented the highest content of *p*-coumaric acid. It was also investigated the presence of flavonoids, finding rutin, phloridzin, myricetin, quercetin and phloretin in quelite cenizo, meanwhile quintonil showed only the presence of rutin and phloridzin. On the other hand, the purslane showed the presence of apigenin and myricetin. Naringenin and galangin were also analyzed, nevertheless, they were not found in the samples. Previous investigations about the content of phenolic acids and flavonoids by HPLC showed that some species of *Chenopodium* spp. have a considerable amount of those compounds [47]. It is well known that the identified compounds displayed varieties of biological activities, suggesting *quelites* as foods rich in antioxidant compounds. According to Sicari et al. [2], the main compounds found in *Portulaca oleracea* L., are *p*-coumaric acid and quercetin, but also, apigenin was identified.

The chromatograms showed other peaks, which could correspond to other phenolics and flavonoids, which were not identified due to their standards were not available, but contributing to the total concentrations given in "Total carotenoids, total phenols, total flavonoids and ascorbic acid content", for this reason, the TPC and TFC values in Table 2 differ from TPI and TFI showed in Table 5.

Conclusion

This study may conclude that leaves of purslane, quelite cenizo and quintonil are rich sources of phytochemicals and nutritional compounds. The chlorophyll, carotenoids, phenols, and flavonoids present in the plants studied here, are alternative sources of nutrients and bioactive compounds for human nutrition. Likewise, the protein content in quelite cenizo and quintonil suggest this variety of food as an accessible, economical and highly beneficial alternative

Table 5 Phenolic acids and flavonoids content in *P. oleracea* L., *C. berlandieri* L., and *A. hybridus* L.

Phenolic acids (µg/g DW)										
	Caffeic	Gallic	Chlorogenic	Vanillic	*p*-hydroxybenzoic	*p*-coumaric	Ferulic	Syringic	TPI
P	36.59 ± 0.07^{c}	31.68 ± 0.08^{c}	ND	38.08 ± 0.04^{c}	ND	60.78 ± 0.10^{a}	106.53 ± 0.50^{c}	ND	273.65
QC	130.71 ± 0.05^{a}	45.04 ± 0.06^{a}	270.82 ± 0.07^{a}	45.23 ± 0.06^{a}	84.31 ± 0.10^{b}	ND	157.54 ± 0.07^{b}	36.45 ± 0.20	770.10
Q	94.03 ± 0.03^{b}	40.68 ± 0.01^{b}	263.77 ± 0.05^{b}	42.12 ± 0.03^{b}	138.14 ± 0.40^{a}	5.49 ± 0.02^{b}	316.95 ± 0.40^{a}	ND	901.19
Flavonoids (µg/g DW)									
	Rutin	Phloridzin	Myricetin	Quercetin	Naringenin	Phloretin	Galangin	Apigenin	TFI
P	ND	ND	39.63 ± 0.03^{b}	ND	ND	ND	ND	56.51 ± 0.10	96.14
QC	2683.14 ± 0.50^{a}	1279.70 ± 0.03^{a}	1819.29 ± 0.06^{a}	135.13 ± 0.01	ND	602.81 ± 0.05	ND	ND	6520.07
Q	7.69 ± 0.10^{b}	377.96 ± 0.05^{b}	ND	ND	ND	ND	ND	ND	385.65

P purslane, *QC* quelite cenizo, *Q* quintonil, *ND* not detected, *TPI* total phenols index (sum of the concentration of individual phenols, µg/g DW), *TFI* total flavonoids index (sum of the concentration of individual flavonoids, µg/g DW)

The data express values of the mean ± SD ($n = 3$). In each column, different letters mean statistically significant difference according to the Tukey test ($p \leq 0.05$). Results were expressed as µg/g of dry weight (DW)

in marginalized rural areas, where the consumption of these wild crops, resistant to environmental conditions, can be used as important sources of antioxidants in a correct diet, and at the same time promote a strategy for food security.

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