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Phytochemical characterization of Izote (Yucca elephantipes) flowers

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Summary

Flowers of the Izote (Yucca elephantipes) are traditionally consumed in different dishes in the Mexican cuisine. Although the use of the flowers in Salvador, Guatemala and México is quite popular, there are no scientific reports of their physicochemical properties and phytochemical composition of petals, carpels and stamens. As part of our research program on characterization of edible wild plants, we have analysed the composition and content of phenolic compounds in methanol crude extracts of petals, carpels and stamens from Y. elephantipes. The petals exhibited eighteen phenolic compounds, including 4-coumaric acid, rutin, ferulic acid, 4-hydroxybenzoic acid, caffeic acid, quercetin 3-glucoside, trans-cinnamic acid, among others. The principal phenolic compound found in petals, carpels and stamens was 4-coumaric acid, with 1154.20, 526.19 and 484.50 µg/g, respectively. In addition, carpel and petals were found to be rich in fatty acids, including linoleic, oleic, and palmitic acid. The petals also contained the highest amount of total dietary fiber. Based on these results, the flowers of Y. elephantipes appear to be a good source of phenolic compounds. This information may be useful in identifying these types of flowers and contribute in future research related to their use in the food area.

Keywords: Izote; Phenolic compounds; Wild flower; Food composition; Functional food; Total dietary fiber.

Introduction

Edible flowers are becoming more popular, as evidenced by an increased number of edible flower cookbooks, culinary magazine articles and television shows. Consumers can purchase packaged flowers for use in meals, as a garnish, or as an ingredient in salads, soups, entrees, desserts and drinks. The combination of flowers with common vegetables or other foods could generate new and interesting flavours (CHAMBERS and KOPPEL, 2013; BENVENUTI et al., 2016). They improve the appearance, taste and aesthetic value of food, aspects that consumers appreciate, justifying the increasing trend of fresh top quality flowers' sales worldwide (KELLEY et al., 2001). However, consumers also demand foods with beneficial health properties, in addition to the nutrients they contain, looking for functional qualities such as phenolic compounds and unsaturated fatty acids (FERNANDES et al., 2017). However, some edible flowers may contain toxic and hazardous chemical compounds. So that, more investigations are needed in order to know either beneficial or adverse properties of these products. In the case of describing nutraceutical properties, these can offer additional values of edible plants biodiversity for feeding requirements. The biological activity of some edible flowers has been attributed to their antioxidant content, including polyphenols, vitamin C, vitamin E, beta-carotene and other

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important phytochemicals (BARROS et al., 2010). In addition, unsaturated fatty acids such as linoleic and linolenic, present in some edible flowers, which cannot be synthesized by humans and must be acquired through dietary intake. Unsaturated fatty acids synthesized by these types of flowers are now important dietary sources of these essential compounds.

Yucca elephantipes belongs to the Agavaceae family, and is native to Central America and México, where it grows at an altitude of 2000-3500 m a.s.l. It is also known as giant yucca, common yucca or Y. guatemalensis. Y. elephantipes is often used as an ornamental plant and tree fence, and generally, petals and carpels are separated from the flower to prepare specific dishes. Petals and carpels (stigma, style, ovules and ovaries) of the flowers are consumed in different dishes in México, including "chileatole" with egg, with meat and some other regional dishes. Its traditional use in ethnomedicine has been documented for many years, although little clinical evidence is available. In México and Central America the flowers are used as diuretics and for the treatment of kidney disease. However, it is important to highlight that no systematic studies have been conducted to determine the bioactive ingredients of Y. elephantipes flowers, which may play an important role in developing and making this edible flower popular on a global scale. Even though the flowers can only be consumed within a short annual blooming period during the summertime, they still play an important role in the diet of lowincome populations (SOTELO et al., 2007). All parts of the flower have a characteristic bitter taste, mainly due to their steroidal saponins content, particularly tigogenin and sarsasapogenin (GALVEZ, 1996). Unlike other flowers, the carpel and pistil carpel of Y. elephantipes can be consumed separately.

As part of our research program of characterization of edible wild plants and with the main goal to contribute to the knowledge on nutritional properties and phytochemical composition of *Y. elephantipes* flowers, the aim of this study was to evaluate the content of phenolic compounds, unsaturated fatty acids as well as other parameters in the different parts of the flower (petal, carpel and stamen).

Materials and methods

Standards and reagents

The solvents used for the extraction and liquid chromatography-mass spectrometry procedures were HPLC and MS grade, respectively and they were purchased from Sigma-Aldrich (St Louis, MO, USA). All the stock solutions, samples, solvents and reagents were filtered through 0.20 μ m PTFE membrane filters (Phenomenex, USA) before separation or injection in the instrument.

Samples

Whole flowers of *Y. elephantipes* were collected during summer (July-August) in 2014 and 2015 around the city of Xalapa in the state of Veracruz, México (18°N, 96°O, 1200 m a.s.l.), according to

recommendations by local consumers. The harvest indicator considered was that the panicle had more than 90% of flowers completely open. Immediately after harvest, the petals, carpels and stamens were manually separated, washed and dried in air oven at 60 °C until approximately 90% moisture was removed. The dried samples were then ground and stored in air-free bags at -40 °C until use. Samples from two years (2014 and 2015) were analysed in triplicate and separately, but no significant differences were found in the physicochemical and antioxidant properties of the two years evaluated (data not shown), so in this work the data were analysed as a single sample with six replicates.

Physicochemical properties

One hundred fresh flowers were randomly selected. The petals, carpels and stamens of the flowers were weighed with an accuracy of 0.001 g. The proportion mass of each flower part was calculated as a percentage. Titratable acidity and pH, as well as the moisture, ash, protein, fat, reducing sugars and soluble solid content was determined in the different parts of the flower (petals, carpels and stamens) following the methodology described by the Association of Official Analytical Chemists (AOAC, 2000). The vitamin C content was determined by using a second-order derivative spectrophotometric method. A calibration curve was generated from different standard ascorbic acid solutions, which were measured using a spectrophotometer (8453; Agilent Technologies, Santa Clara, CA, USA) at 250-350 nm (PFENDT et al., 2003). The colour measurement was made with a ColorFlex V1-72 colorimeter (Hunter Lab, Reston, VA, USA), using 0°/45° geometer specular excluded, spectral with a range of 400-700 nm, an spectral resolution < 3 nm, effective bandwidth of 10 nm equivalent triangular with a photometric range from 10 to 150%. The light source was a pulsed xenon lamp. The external colour of each part of the flower was analysed by measuring the L*, a* and b* parameters, and subsequently calculating the secondary hue angle (H°) and Chroma.

Phenolic compounds extractions and phytochemical analysis

Petals (0.5 kg) were extracted three times, each with 3 L of 85% methanol for 48 hours at room temperature, 150 rpm. The extracts were filtered through Whatman no. 1 filter paper under vacuum, and then they were concentrated in a rotary evaporator at 35 °C (R-210; Buchi, Flawil, Switzerland). These extracts were used to determine the phenolic content of Y. elephantipes. For the phytochemical analysis, extracts were prepared using an accelerated solvent extraction system (ASE 350, Thermo Scientific, USA). Briefly, 3.0 g of dry material was dispersed in 1.0 g of diatomaceous earth and placed in a 34 mL cell. The cell was filled up with MeOH up to a pressure of 1,500 psi and heated at 60 °C during 5 min. Then, the cell was washed off with 30% of cell volume. The extract was concentrated by rotary evaporation (Büchi RII, Switzerland). 10 mg of the crude extract was re-dissolved in 1.0 mL of MeOH with 0.1% of formic acid (Both MS grade, Sigma-Aldrich), filtered and placed in a 1.5 mL UPLC vial. Samples were analysed by triplicate.

The identification and quantitation of individual phenolic compounds was performed with an Ultra High Performance Liquid Chromatography (UPLC, Agilent 1290 series) coupled to a triple quadrupole mass spectrometer (MS-MS, Agilent 6460). The chromatographic analysis was carried out on a ZORBAX SB-C18 column (1.8 μ m, 2.1 × 50 mm) (Agilent Technologies) with the column oven set at 40 °C. The mobile phase consisted of (A) water containing 0.1% formic acid and (B) acetonitrile (90%) containing 0.1% formic acid. The gradient conditions of the mobile phase were: 0 min 1% B, 0.1-40 min linear gradient 1-40% B, 40.1-42 min linear gradient 40-90% B, 42.1-44 min isocratic 90% B isocratic, 44.1-46 min linear gradient 90-1% B, 46.1-47 min 1% B isocratic (total run time 47 min). The flow rate was 0.1 mL/min, and 1 µL of sample injection volume. A dynamic multiple reactions monitoring (dMRM) acquisition method was established. The MRM transitions for each compound were searched in public databases as Metlin and were corroborated experimentally in our laboratory (Tab. 1). The precursor and product ions were considered as qualifier ions and the product ion was considered as the quantifier ion. The ESI source was operated in positive and negative ionization modes with the desolvation temperature of 300 °C, the Cone gas (N₂) flow of 5 L/min, the nebulizer pressure of 45 psi, the sheath gas temperature of 250 °C, the sheath gas flow of 11 L/min, the capillary voltage (positive and negative) was 3,500 V and the nozzle voltage (positive and negative) of 500 V. The retention time of each compound was determined experimentally and the search window was set in maximum 1 min. The cell accelerator voltage was 7 V for each compound. For quantitation, a calibration curve for each phenolic compound with 10 concentration points (0.5, 1, 3, 5, 7, 9, 11, 13, 15 and 17 μ M) was prepared. Each concentration point was injected twice and the respective areas considered for the quadratic regressions (Tab. 1). The coefficient of determination (r^2) values were higher than 0.99 for each compound (Tab. 1). The data was obtained with the MassHunter Workstation Software version B.06.00 (Agilent Technologies) and the results were expressed as the average \pm standard deviation of $\mu g/g$ of sample (dry weight).

Fatty acid profile

The fatty acid profile was determined in the oil extracted from the petals, carpel and stamens with hexane HPLC grade using a Soxhlet system and converting the oil into methyl esters by the addition of BF₃, in accordance with the method described by LOPEZ et al. (2001). The hexane extract obtained from the esterification process was analysed in a GCD Plus gas chromatograph coupled to a mass spectrometer, Hewlett-Packard model 1800 B, with the following parameters: the initial temperature of both the injector and the detector was 250 °C; the temperature was adjusted as follows: initial temperature of 80 °C for 5 min, then later elevated by 30 °C/min reaching to 250 °C. A carbowax column with a length of 30 m, a diameter of 0.25 mm and a film thickness of 0.25 um was used, with helium as the carrier gas at a flow of 1 mL/min. The mass spectra were obtained by means electron ionization at 70 eV. The identification was performed by comparison of the mass spectra obtained for each compound with a database (HP Chemstation-NIST 05 Mass Spectral search program, version 2.0d). In addition to the comparison with authentic commercial standards (FAME mix, C8:C22, Catalogue No. 18920-1AMP, Sigma-Aldrich) analysed under the same conditions.

Statistical analysis

The analysis of the sample (2014 and 2015) was performed in triplicate. Variables responses were transformed to square root and analysed with a unifactorial multivariate design (MANOVA, GLM) testing by each part of the flower and verified that the assumptions of normality and homogeneity of variances were met. Within each test, comparisons among the each part of the flower were performed by a *post-hoc* Tukey analysis with α 0.05 and average and standard error was back transformed from square root. All statistical analyses were performed using STATISTICA 7.0[®] Statsoft, Inc. 1984-2004.

Results and discussion

Physicochemical properties

The average weight of one flower of *Y. elephantipes* was 0.39-0.45 g. The part that contributed most to the mass of the flowers was the car-

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		MRM tra	unsitions	Chromatog	raphic data	Mass s	pectrometer con	ditions		Quantification	
#	Compound name	Precursor ion	Product ion	Retention time (min)	Retention window (± min)	Fragmentor voltaje (V)	Collision energy (V)	Polarity	Range (µM)	Regression type	r ^{.2}
1	Shikimic acid ¹	173.1	111.1	1.7	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
5	Gallic acid ²	169	125.2	3.8	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
3	Protocatechuic acid ¹	153	109.1	6.4	0.25	100	10	Negative	0.5 - 17	Quadratic	0.99
4	4-Hydroxybenzoic acid ¹	137.1	92.8	9.11	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
5	Gentisic acid ¹	153	109	9.2	0.25	100	10	Negative	0.5 - 17	Quadratic	0.99
9	(-)-Epigallocatechin ³	305.1	125	10.44	0.5	140	20	Negative	0.5 - 17	Quadratic	0.99
7	4-Hydroxyphenylacetic acid ⁴	107.1	77	10.52	0.5	140	20	Positive	0.5 - 17	Quadratic	0.99
8	(+)-Catechin ²	291	138.9	11.2	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
6	Vanillic acid ¹	169	93.03	11.81	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
10	Scopolin ⁵	355.1	193	11.87	0.5	100	20	Positive	1 - 17	Quadratic	0.99
11	Chlorogenic acid ¹	355.1	163.03	12.12	0.3	100	10	Positive	0.5 - 17	Quadratic	0.99
12	Caffeic acid ¹	181.04	163.03	12.24	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
13	Procyanidin B2 ¹	577.1	425.1	13.69	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
14	Kuromanin ¹	449	286.9	14.24	0.5	140	30	Positive	1 - 17	Quadratic	0.99
15	(-)-Epicatechin ²	291	138.8	14.48	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
16	Vanillin ¹	153	124.9	14.99	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
17	Mangiferin ²	423	302.8	15.18	0.5	100	10	Positive	1 - 17	Quadratic	0.99
18	Keracyanin ²	595.2	287.1	15.2	0.5	100	20	Positive	1 - 17	Quadratic	0.99
19	4-Coumaric acid ¹	165.05	147.04	16.22	0.25	100	10	Positive	0.5 - 17	Quadratic	0.99
20	(-)-Gallocatechin gallate ²	458.9	139	16.57	0.5	80	20	Positive	0.5 - 17	Quadratic	0.99
21	Umbelliferone ¹	163	107	17.1	0.5	100	30	Positive	1 - 17	Quadratic	0.99
22	Quercetin-3,4-di-0-glucoside ¹	627	302.9	17.87	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
23	Scopoletin ¹	193	133	18.44	0.5	100	10	Positive	1 - 17	Quadratic	0.99
24	3-Coumaric acid ¹	165.05	147.04	18.7	0.25	100	10	Positive	0.5 - 17	Quadratic	0.99
25	Ferulic acid ¹	195.1	145.02	19.18	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
26	Sinapic acid ¹	225.1	207.1	19.57	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
27	Epicatechin gallate ³	443.1	123	19.91	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
28	Myricitrin ¹	465	318.9	20.25	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
29	Ellagic acid ¹	301	145	20.36	0.5	100	40	Negative	0.5 - 17	Quadratic	0.99
30	Quercetin-3-D-galactoside ²	465	302.9	20.6	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99

		MRM tra	nsitions	Chromatog	raphic data	Mass s	pectrometer con	ditions		Quantification	
#	Compound name	Precursor ion	Product ion	Retention time (min)	Retention window (± min)	Fragmentor voltaje (V)	Collision energy (V)	Polarity	Range (µM)	Regression type	1 ^{.2}
31	Rutin ²	611	302.9	20.65	0.5	100	10	Positive	0.5 - 17	Quadratic	66.0
32	Quercetin-3-glucoside ²	465	303	21.19	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
33	Luteolin-7-0-glucoside ¹	449	287	21.4	0.6	100	10	Positive	0.5 - 17	Quadratic	0.99
34	<i>p</i> -Anisic acid ⁴	153.1	109	22.21	0.5	120	5	Positive	0.5 - 17	Quadratic	0.99
35	Penta- O -galloyl- β -D-glucose ²	771.1	153	22.27	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
36	2,4-dimethoxy-6-methylbenzoic acid ²	197	179	23.43	0.5	80	5	Positive	1 - 17	Quadratic	0.99
37	Kaempferol-3-0-glucoside ¹	449	286.9	23.64	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
38	Naringin ¹	273	153	23.64	0.5	120	10	Positive	0.5 - 17	Quadratic	0.99
39	Quercitrin ¹	449.1	303.1	23.71	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
40	Hesperidin ¹	609.1	301.1	24.55	0.5	100	20	Negative	0.5 - 17	Quadratic	0.99
41	Myricetin ¹	317	179	24.64	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
42	Rosmarinic acid ¹	361.1	163	24.85	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
43	Phloridzin ¹	435	272.9	25.21	5.0	100	10	Negative	0.5 - 17	Quadratic	0.99
44	Trans-resveratrol ²	229.1	135	25.86	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
45	Trans-cinnamic acid ¹	149.1	131	28.5	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
46	Cirsimarin ⁵	477	314.9	29.4	5.0	100	10	Positive	0.5 - 17	Quadratic	0.99
47	Quercetin ²	302.9	153.1	29.7	0.5	100	35	Positive	1 - 17	Quadratic	0.99
48	Luteolin ¹	287.1	153	30.09	0.3	100	30	Positive	1 - 17	Quadratic	0.99
49	Psoralen ¹	187	131.1	31.4	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
50	Angelicin ²	187	131.1	32.74	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
51	Naringenin ¹	271	151	32.83	0.5	100	10	Negative	0.5 - 17	Quadratic	0.99
52	Apigenin ¹	271	153	34.1	0.5	100	30	Positive	1 - 17	Quadratic	0.99
53	Kaempferol ¹	287.1	153	34.79	0.3	100	30	Positive	0.5 - 17	Quadratic	0.99
54	Hesperetin ¹	303.1	177.1	34.8	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
55	Citropten ⁴	207.06	192.04	36.11	5.0	100	20	Positive	0.5 - 17	Quadratic	0.99
56	Nordihydroguaiaretic acid ¹	303	193.1	42.36	0.5	100	10	Positive	0.5 - 17	Quadratic	0.99
57	Chrysin ¹	255.1	153	43.13	0.5	100	40	Positive	1 - 17	Quadratic	0.99
58	Kaempferide ¹	301	258.2	43.5	0.5	100	20	Positive	0.5 - 17	Quadratic	0.99
59	Emodin ¹	269	225	44.2	0.5	150	20	Negative	0.5 - 15	Quadratic	0.99
60	Chrysophanol ¹	255.1	153	45.3	0.2	100	40	Positive	0.5 - 17	Quadratic	0.99

Tab. 1 continued: Analytical conditions by UPLC-MS-MS for identification and quantification of phenolic compounds.

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The cell accelerator voltage was 7 V for each compound. Compounds purchased from ¹Extrasynthese (Lyon, France), ²Sigma Aldrich (St. Louis, USA) ³Cayman Chemical Company (Michigan, USA), kindly donated by ⁴Dr. Thor Armason (University of Ottawa), isolated ⁵in house and kindly donated by ⁶Dr. Sergio Peraza Sánchez (Scientific Research Center of Yucatan).

pels (78.4 g/100 g), followed by the petals (13.4 g/100 g) and stamens (7.4 g/100 g).

Tab. 2 shows the results of the proximate analysis and the physicochemical properties of the different parts of the flower. Water was the main constituent of the flowers. The initial moisture content of the whole flower, expressed as a percentage of the fresh weight, was about 70 g/100 g. The moisture content decreased after drying, being for petals, carpels and dry stamens of 5.36 g/100 g, 8.36 g/100 g and 7.67 g/100 g, respectively. These moisture values prevent the deterioration and spoilage of the samples by microorganisms and chemical reactions (BELL, 2008). Total dietary fiber content in the petals, carpel and stamens was 19.23, 15.28 and 14.34 g/100 g, respectively. These values were similar to observed by other edible flowers (NAVARRO-GONZALEZ et al., 2015) and that previously reported for some species of algae which have been used to supplement the diet (RUPÉREZ and SAURA-CALIXTO, 2001), indicating that this flower may be considered as a good source of fiber. In addition, it has been reported that these flowers contain saponins (GALVEZ, 1996) which may contribute to the reduction of cholesterol (SAN JOSE et al., 2016) and explain its traditional use for hypercholesterolemia. However, these should be used with caution since it has been reported that they are not absorbed in the intestine and therefore decrease the absorption of iron and zinc (HON et al., 1988). The fat content varied from 12.40 to 19.88 g/100 g, being carpels the ones with the highest concentration. These values are higher than those reported for other edible flowers (BARROS et al., 2010). Stamens showed a higher content of reducing sugars of around 12.20 g/100 g, possibly because they contain pollen grains with a higher concentration of simple sugars and pollen to favor pollination.

The edible petals had a bright white colour (L* = 57.56, a* = 6.07, b* = 26.98), while the carpels (L* = 54.64, a* = 2.62, b* = 26.08) and stamens (L* = 39.77, a* = 8.42, b* = 22.60) presented a typical light green colour.

Tab. 2:	Proximal	analysis	and	physicochemical	properties	of	the	different
	parts of th	ne Izote (Үиссы	a elephantipes) flo	owers.			

Property	Petal	Carpel	Stamens
Moisture (g/100 g)	5.36 ± 0.38^a	8.36 ± 0.25^{b}	$7.67\pm0.39^{\rm b}$
a _w (T=25 °C)	0.449 ± 0.02^{a}	$0.610\pm0.01^{\rm b}$	$0.660 \pm 0.01^{\circ}$
Total ash (g/100 g)	$1.60\pm0.04^{\rm a}$	$3.19\pm0.99^{\rm c}$	$2.69\pm0.01^{\rm b}$
Total dietary fiber (g/100 g)	$19.23 \pm 1.15^{\mathrm{b}}$	$15.28\pm1.25^{\rm b}$	$14.34\pm2.50^{\rm a}$
Proteins (g/100 g)	0.31 ± 0.05^a	$0.37\pm0.04^{\rm a}$	0.26 ± 0.09^a
рН	6.44 ± 0.34^{b}	$4.60\pm0.20^{\rm a}$	$4.73\pm0.08^{\rm a}$
Fat (g/100 g)	$17.55 \pm 0.36^{\circ}$	$19.88\pm0.29^{\rm b}$	$12.40\pm0.80^{\rm a}$
Titratable acidity (meq citric acid/100 g)	0.31 ± 0.05^{a}	0.40 ± 0.07^{a}	0.52 ± 0.07^{b}
Reducing sugar (g/100 g)	9.77 ± 1.20^{a}	$9.27 \pm 1.50^{\mathrm{a}}$	12.20 ± 2.30^{b}
Colour			
L*	$57.56 \pm 1.61^{\mathrm{b}}$	$54.64 \pm 1.12^{\mathrm{b}}$	$39.77\pm0.70^{\rm a}$
<i>a</i> *	6.07 ± 0.23^{b}	$2.62\pm0.20^{\rm a}$	$8.42\pm0.26^{\rm c}$
<i>b</i> *	26.98 ± 0.34^{b}	26.05 ± 0.23^b	22.60 ± 0.65^a
Hue	$77.28\pm0.60^{\rm b}$	$84.25\pm0.47^{\rm c}$	$69.56\pm0.17^{\rm a}$
Chroma	27.59 ± 1.29^{a}	$26.18 \pm 0.90^{\rm a}$	$24.12\pm0.70^{\rm a}$
Vitamin C (mg/100 g)	$151.05 \pm 3.20^{\circ}$	65.60 ± 1.90^{a}	126.00 ± 2.05^{b}

Results are expressed as the mean $(n=6) \pm SD$. Values with different letters within the same row indicate significant differences by Tukey's test (p < 0.05).

All samples exhibited a lower protein concentration (0.37 g/100 g) than that previously reported for the flowers of *Y. filifera* (1.62 g/100 g) (SOTELO et al., 2007). Unlike other edible flowers such as *Rosa micrantha*, protein was the least abundant nutritional component in all of the examined parts of the flower, whereas the total dietary fiber was the most abundant component. The results revealed that petals (151.05 mg/100 g) and stamens (126.00 mg/100 g) contain about twice the vitamin C concentration than carpels (65 mg/100 g), but all three samples contain more than the recommended minimum daily value of the Food and Drug Administration of 45-90 mg/100g (FAO, 2018). These values are consistent with data reported for other edible flowers (LARA-CORTÉS et al., 2014).

According to these data, petals and stamens can be considered excellent vitamin C resources. Vitamin C is an antioxidant and anticarcinogenic compound and their lack causes scurvy (GROSSO et al., 2013). Our research represents an initial contribution to the study of this edible flower necessary to consider its use in traditional Mexican and Central American food and to encourage its use in international cuisine.

Phenolic compounds analysis

Phenolic compounds are the main active ingredients in edible flowers (HE et al., 2015; LI et al., 2014). The interest in edible flowers is probably continuously increasing due to their potential health effects that are related with their chemical composition (FERNANDES et al., 2017). In order to determine the compounds that may be responsible for the traditional therapeutic effects, we investigated the chemical constituents of the petals, carpel and stamens (Tab. 3). For this, we used an analytical method by UPLC-MS-MS that includes a large number of well-known phenolic compounds which have been widely described in edible and medicinal plants (Tab. 1) and that we have partially reported previously (JIMENEZ et al., 2018; LASCURAIN et al., 2018; REYES-RAMÍREZ et al., 2018).

Twenty compounds were identified and quantified based on their retention time and mass spectrometry data compared with authentic standards in the different parts of the flower (Fig. 1). From the MeOH extract, the most abundant compound identified in petals, carpel and stamens was 4-coumaric acid with 1154.21, 526.19 and 484.50 µg/g, respectively. Along with 4-coumaric acid, in petals it was identified other seventeen compounds including rutin (348.78 µg/g), ferulic acid (72.83 µg/g), 4-hydroxybenzoic acid (60.66 µg/g), caffeic acid (67.71 µg/g), quercetin 3-glucoside (37.40 µg/g), Kaempferol (25.81 µg/g), trans-cinnamic acid (23.40 µg/g), among others. The carpels exhibited ten compounds besides 4-coumaric acid (526.19 $\mu g/g$), among them vanillic acid (75.34 $\mu g/g$), 4-hydroxybenzoic acid (45.81 µg/g) and Trans-cinnamic acid (28.00 µg/g). Stamens presented fifteen compounds more besides 4-coumaric acid being vanillic acid (110.21 μ g/g) the second most abundant. 4-coumaric acid is a member of the phenolic compounds and it has been well reported its antioxidant effect, it has been used as natural antioxidant because it inhibits lipid peroxidation, decreases low density lipoprotein peroxidation and plays an important role in immune regulation in human (KILIC and YESILOGLU, 2013). In addition, phenolic compounds are known to be capable of electron delocalization, which is the cause of their free radical scavenging activity (RICE-EVANS et al., 1996). Therefore, these compounds may contribute to the antioxidant activity and therapeutic effects of this flower in the Mexican traditional medicine. It has been reported that the bioactivity of some edible flowers is strongly correlated with the phenolic compounds composition (KOIKE et al., 2015). The radical scavenging activity of the phenolic compounds mainly depends on the number and position of hydroxyl groups in the molecules. The phenolic compounds identified in Y. elephatipes petals were similar to that reported in other edible flowers (HE et al., 2015), but the concentration was specific for

#	Compound	Petals	Carpels	Stamens
1	Gallic acid	2.91 ± 0.02		$2.37 \pm 0.02*$
2	Protocatechuic acid	13.37 ± 0.15	2.94 ± 0.03	4.83 ± 0.12
3	Gentisic acid	$1.92 \pm 0.1^*$	5.53 ± 0.05	4.86 ± 0.12
4	4-hydroxybenzoic acid	60.66 ± 0.85^{b}	45.81 ± 0.79^{a}	62.74 ± 1.94
5	Epigallocatechin			8.16 ± 0.15
6	(+)-Catechin			57.94 ± 0.57
7	Vanillic acid	19.73 ± 0.24^{a}	75.34 ± 0.95^{b}	$110.21 \pm 1.64^{\circ}$
8	Chlorogenic acid	8.18 ± 0.15^a	$3.58 \pm 0.1*$	35.76 ± 0.71
9	Caffeic acid	67.71 ± 0.70^{b}	$2.44 \pm 0.09*$	3.95 ± 0.44^{a}
10	4-Coumaric acid	1154.21 ± 25.00	$526.19 \pm 15.97^{\circ}$	484.50 ± 6.10
11	Quercetin 3,4-di-O-glucoside	9.55 ± 0.29^{a}		19.69 ±0.39 ^b
12	Ferulic acid	72.83 ± 0.59°	26.14 ± 0.31^{a}	41.62 ± 0.34^{b}
13	3-Coumaric acid	5.73 ± 0.40^{a}		
14	Rutin	$348.78 \pm 6.87^{\circ}$	12.20 ± 0.16^{a}	32.22 ± 0.49^{b}
15	Quercetin 3-D-galactoside	19.71 ± 0.59^{a}		
16	Quercetin 3-glucoside	37.40 ± 0.57^{a}		
17	Kaempferol 3-O-glucoside	9.41 ± 0.13^{a}		
18	Trans-cinnamic acid	23.40 ± 0.33^{a}	28.00 ± 0.07^{b}	$9.55 \pm 0.04^{\circ}$
19	Quercetin	9.54 ± 0.15^{a}		
20	Kaempferol	25.81 ± 0.36^{b}		11.35 ± 0.15^{a}

Tab. 3: Phenolic compounds (µg/g of dry sample) present in methanol extract from the dried petals, carpels and stamens of Izote (Yucca elephantipes) flower.

*Identification and quantitation was confirmed by Ultra High Performance Liquid and dynamic multiple reaction monitoring (dMRM), using authentic standard for retention time mapping and calibration curve for validation in the analysis. --- No detected. Values with different letters within the same row indicate significant differences by Tukey's test (p < 0.05).

this flower, so these results suggest that the flower can be considered a good source of 4-coumaric acid.

results suggest that *Y. elephantipes* flowers are good options for consume of unsaturated fatty acids according to the recommendations of World Health Organization (WHO, 2008).

Fatty acid profile

Fifteen, thirteen and ten fatty acids were identified in the oil extracted from petals, carpel and stamens, respectively. The fatty acids varied from C12: 0 to C23: 0 and their individual relative area (%) is shown in Tab. 4. The three oil extracts presented high concentration of unsaturated fatty acids corresponding to 9,12-octadecadienoic (linoleic) and 9-octadecenoic (oleic) acids, representing more than 70% in petals and carpel and about 50% in stamens. According with the recommendations of European Food Safety Authority (EFSA), the daily intake of saturated fatty acids should be as lowest as possible (EFSA, 2010). All samples also contained hexadecanoic (Palmitic) acid, but in lower proportions. Petals presented the higher number of the fatty acids, corresponding to linoleic acid with a value of 36.07%, oleic acid with a value of 35.17% and palmitic acid with a value of 18.78%, while carpel exhibited a higher ratio of linoleic (38.40%) and linolenic (5.20%) acids, both of them considered essential fatty acids, suggesting that these samples could be used to supplement foods with low levels of these compounds. The results obtained were similar to those reported in Malva silvestris flowers (BARROS et al., 2009). The saturated/unsaturated fatty ratio was 0.318, 0.276 and 0.630 for petal, carpel and stamens, respectively and they are comparable to those found in the oil of some varieties of pomegranate as well (AMRI et al., 2017). All samples showed a similar fatty acid profile considering the presence of linoleic (omega-6) and oleic (omega-9) acids in a higher proportion and linolenic acid (omega-3) in a lower proportion. These

Conclusions

We found that petals, carpels and stamens of the *Y. elephantipes* flowers have different chemical and nutritional composition which provides information to consumers to make decisions about their consumption. Our study revealed that petals contain a greater number of phenolic compounds, but in lower concentration compared to carpels and stamens. 4-Coumaric acid was the compound found in greater concentration in carpels followed by stamens and finally in petals. Carpels also had the highest concentration of free fatty acids, specifically linoleic and linolenic acid. In this regard, *Y. elephantipes* flowers could potentially be a valuable natural source of phenolic compounds, with possible applications in functional foods. Their physicochemical properties, combined with their chemical composition, suggest their suitability as a good source of fiber for human consumption, which may be used in many kinds of foods, beverage and supplements.

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Fig. 1: Chromatograms of the methanolic extract of petals (A), carpels (B) and stamens (C) of Izote (*Yucca elephantipes*) flowers. The number above peaks indicates the compound indicated in Tab. 3.

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#	RT	Compound		Relative area (%)	
			Petals	Carpel	Stamens
1	9.04	Dodecanoic acid	0.13 ± 0.01^{a}	-	-
2	11.31	Tetradecanoic acid	0.40 ± 0.12^{a}	0.54 ± 0.10^{a}	-
3	12.57	Pentadecanoic acid	0.54 ± 0.01^{a}	0.52 ± 0.05^{a}	-
4	13.72	9-Hexadecenoic acid	1.26 ± 0.03^{b}	0.13 ± 0.03^{a}	$2.80 \pm 0.04^{\circ}$
5	14.10	Hexadecanoic acid	18.78 ± 0.22^{b}	16.10 ± 0.21^{a}	27.60 ±0.25°
6	15.90	Heptadecanoic acid	$0.65 \pm 0.01^{\circ}$	1.75 ± 0.01^{b}	-
7	17.66	9,12-Octadecadienoic acid	36.07 ± 1.50^{b}	38.40 ± 1.70^{b}	22.15 ± 0.89^{a}
8	17.80	9-Octadecenoic acid	35.17 ± 0.98^{b}	33.70 ± 1.10 ^b	27.90 ± 0.01^{a}
9	17.83	9,12,15-Octadecatrienoic acid	1.88 ± 0.09^{b}	$5.20 \pm 0.11^{\circ}$	0.50 ± 0.08^{a}
10	18.28	Octadecanoic acid	1.99 ± 0.09^{a}	2.10 ± 0.07^{b}	$5.60 \pm 0.05^{\circ}$
11	24.41	11-Eicosaenoic acid	$0.59 \pm 0.02^{\circ}$	0.80 ± 0.01^{b}	$2.30 \pm 0.02^{\circ}$
12	25.49	Eicosanoic acid	0.50 ± 0.01^{b}	0.26 ± 0.10^{a}	2.30 ± 0.10^{a}
13	36.45	4,7,10,13,16-Docosapentaenoic acid	0.73 ± 0.09^{b}	0.25 ± 0.10^{a}	$5.30 \pm 0.08^{\circ}$
14	38.52	Docosanoic acid	0.63 ± 0.09^{b}	0.45 ± 0.09^{a}	$3.20 \pm 0.15^{\circ}$
15	48.65	Tricosanoic acid	0.50 ± 0.07^{a}	-	-

Tab. 4: Relative area (%) of free fatty acids in different parts of Izote (Yucca elephantipes) flowers.

The fatty acids were identified by their mass spectra and retention times according to the Wiley and NIST mass spectral databases. ",-" means not detectable. RT: Retention time. Values with different letters within the same row indicate significant differences by Tukey's test (p < 0.05).

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