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Physicochemical quality, antioxidant capacity and nutritional value of edible flowers of some wild dahlia species

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ABSTRACT

Mexico is the centre of origin and domestication of the dahlia, which has the widest distribution worldwide as an ornamental plant; however, due to changes in eating habits in some sectors of the population (vegetarians and vegans), it has also been incorporated as an ingredient in the gastronomic field. The aim of this research was to determine the attributes of physicochemical quality, antioxidant capacity and nutritional value of ligulate flowers of some wild dahlia species. A completely randomized experimental design with five repetitions was used. Fresh weight, total titratable acidity (TTA), total soluble solids (TSS), ascorbic acid (AA), colour, total phenols (TP), total flavonoids (TFl), total anthocyanins (TAn), antioxidant capacity (AC) and proximate composition were evaluated. The highest fresh weight was observed in D. × hortorum, with similar values among its different flower colours. Likewise, D. merckii and D. coccinea stood out for their high TTA value; however, D. coccinea also had the highest AA content (0.77-0.9 mg g⁻¹ FW), TP (12.87-19.92 mg GAE g⁻¹ FW), TFl (6.78-11.32 mg QE g⁻¹ DW) and AC (47.28-60.43 mg AAEVC g⁻¹ FW). On the other hand, D. campanulata was notable for its high levels of moisture, fat and raw fibre; similarly, D. coccinea stood out for its high amounts of dry matter and total carbohydrates. The ligulate flowers of wild dahlias have a high concentration of bioactive compounds and a nutritional composition with beneficial contributions to health care, so they could be used for consumption as fresh products.

Key words: antioxidant capacity, floriphagia, functional food, non-traditional foods, plant genetic resources, post-harvest handling, vegans, vegetarians

Abbreviations:

TTA - total titratable acidity, TSS - total soluble solids, AA - ascorbic acid, AAEVC - Antioxidant Activity Equivalent to Vitamin C, TP - total phenols, TFl - total flavonoids, TAn - total anthocyanins, AC - antioxidant capacity, ABTS - free radical-scavenging ability determined by the use of a stable 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) radical cation, M – moisture; DM – dry matter, CP – crude protein, RF - raw fat, RFi - raw fibre, TC - total carbohydrates, A - ash



INTRODUCTION

Flowers and inflorescences have constituted an important gastronomic element for many centuries for the civilizations of diverse regions around the world because there are innumerable reports in the literature that indicate their consumption. Among the species that can be mentioned are the rosebush (Rosa spp.), dandelion (Taraxacum officinale) and squash (Cucurbita spp.) in Europe and pre-Hispanic Mexico, but in countries such as China and India consumption of a greater variety of species is also indicated (Sotelo et al., 2007; Rop et al., 2012; Fernandes et al., 2018) At present, the search for new culinary experiences (shapes, colours and flavours) that provide attractive appearance and high nutritional value of the dishes (Mlcek and Rop, 2011) has allowed us to resume and broaden this culinary tradition.

One of the reasons for the increase in the consumption of edible flowers is associated with the presence of biologically active compounds that have beneficial effects on human health (Lee et al., 2009; Huang et al., 2014; Benvenuti et al., 2016) because they are widely valued by consumers seeking a healthy lifestyle (Yang et al., 2014; Chen and Wei, 2017). Among these compounds are phenolic acids, flavonoids and anthocyanins, which have antioxidant properties and, by inhibiting free radicals that induce oxidative stress, reduce the risk of chronic degenerative diseases (diabetes, cognitive deterioration and some types of cancer) and cardiovascular diseases, among others (Zeng et al., 2014; Loizzo et al., 2016; Pires et al., 2018). Edible flowers also have a variable concentration of fat, protein, vitamins, sugars and minerals, which makes them even more attractive for consumption as part of salads, or as decoration for gourmet dishes (Rop et al., 2012; Lara-Cortés et al., 2014).

On the other hand, the most visible problems of post-harvest handling of edible flowers and inflorescences are related to their high perishability due to their lack of photoassimilate accumulation organs, high sensitivity to ambient temperature and mechanical damage (Zeng et al., 2014; Ahmad and Tahir, 2016), as a result of which floral structures tend to undergo changes not only in their external appearance (colour and turgor), but also in their nutritional composition (phenolic compounds, flavonoids, vitamins) and their antioxidant capacity (Kaisoon et al., 2011; Gebremedhin et al., 2013; Liu et al., 2016) Therefore, studies of the variation in these compounds are of paramount importance for determining its magnitude and the possibility of implementing post-harvest conservation methods that allow maintaining and preserving the quality of flowers and inflorescences for use as food (Mlcek and Rop, 2011; Rachkeeree et al., 2018).

The dahlia is a plant native to Mexico. It is represented by 37 species in the genus Dhalia that belongs to the Asteraceae family. They produce flowers with a great diversity of colours (red, purple, pink, white and variegated) (Ohno et al., 2013; Ahmad and Tahir, 2016), and that is why they are cultivated mainly for the production of cut flowers, potted plants and elements for the design and construction of gardens (Younis et al., 2014). The flowers of dahlias also have nutritional and functional properties that make them suitable for consumption as food (Lara-Cortés et al., 2013; Lara-Cortés et al., 2016). However, there are very few or no references pertaining to this topic. Generation of relevant information will allow to expand the study, exploitation and rescue of wild dahlia species, whose natural diversity space is increasingly reduced and threatened by anthropogenic activities. That is why the objective of this research was to determine the attributes of physicochemical quality, antioxidant capacity and nutritional value of ligulate flowers of some wild dahlia species.

MATERIAL AND METHODS

Plant material and location of the experiment

The experiment was carried out in 2016 in the fruit and multipurpose laboratory of the Departamento de Fitotecnia at the Universidad Autónoma Chapingo (UACh). The plant material consisted of inflorescences formed by ligulate and tubular flowers (flower head) of 4 wild species (D. brevis, D. merckii, D. coccinea and D. campanulata) and 1 cultivated species ($D. \times hortorum$) (control), which were harvested in August 2016 from the Experimental Agricultural Field "San Martin" (UACh) located at 19°29'23" N and 98°53'37" W, at an altitude of 2246 m and average annual temperature of 15.6°C. The inflorescences were harvested when their stamens were exposed in the first row of tubular flowers of the flower head, and their stem was 25 cm long. They were placed in glass containers (0.75 L) with tap water for a period of 5 days at room temperature ($20 \pm 1^{\circ}$ C). The ligules were separated from the flower head and classified according to colour; an SP62[®] sphere colorimeter (X-Rite, Michigan, USA) was used to obtain the parameters of brightness (L*), pitch angle (h°) and purity of colour (chroma, C*). On day 1, they were

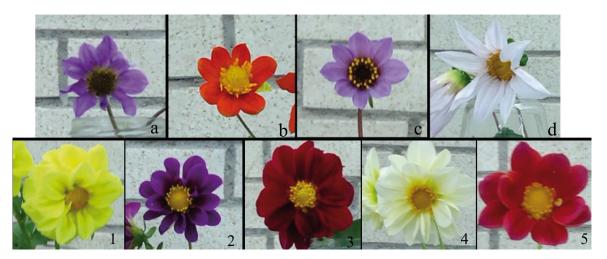


Figure 1. Colour of the ligulate flowers of wild dahlia species: a. D. brevis, b. D. coccinea, c. D. merckii, d. D. campanulata, and of the cultivated species of dahlia D. \times hortorum (1-5)

as follows: *D. brevis* – violet ligules L* 40.47, C* 33.21, h° 304.48; *D. merckii* – violet L* 50.93, C* 27.75, h° 306; *D. coccinea* – orange L* 35.85, C* 63.49, h° 52.66; *D. campanulata* – white with light purple hue L* 74.91, C* 5.37, h° 338.50), and the cultivated species *D.* × *hortorum* – cherry L* 10.46, C* 20.18, h° 15.91; yellow L* 80.28, C* 93.79, h° 93.79; white L* 81.52, C* 20.19, h° 95; purple L* 26.92, C* 45.09, h° 332.92; and red L* 24.55, C* 39.56, h° 25.22) (Fig. 1).

Crop management

The multiplication and obtaining of the vegetal material began in the month of March of 2016 with the planting of the tuberous roots obtained in the previous growth cycle into boxes with peat (peat moss) in a greenhouse; their buds sprouted after 15 days. Transplanting into a field was performed in April 2016, where the distance between plants and rows was 0.6 and 1 m, respectively. The supply of water and nutrients was carried out by means of a drip irrigation system 1-2 times per week, with an expenditure of 0.5-3.5 L per plant, depending on the ambient temperature, relative humidity and phenological stages of the crop. Fertilization was carried out based on the general NPK fertilizer ratio of 120-0-200, which was supplied in split doses to the transplants and before the beginning of flowering. Weed control and some crop management practices such as hilling were carried out manually.

Experimental design

A completely randomized experimental design with five repetitions was used, where the experimental unit consisted of a set of 3 containers (0.75 L) with 3 flowers with a 25 cm long stem. The evaluations were made on days 1, 3 and 5 (vase life).

Parameters evaluated

Fresh weight

The ligulate flowers were separated manually from the flower head and used to measure their weight with an EX125D analytical balance of the Explorer[®] Semi Micro Series (Ohaus, Parsippany, USA) with a maximum capacity and minimum reading of 51 and 0.01 g, respectively. The results are expressed in g.

Total titratable acidity (TTA) and total soluble solids (TSS)

The parameters TTA and TSS were obtained by macerating and obtaining juice from 1 g of ligulate flowers. For the determination of the percentage of citric acid (TTA), 5 mL of the juice were taken and neutralized with 0.1 N NaOH, using 1% phenolphthalein as indicator. TSS were quantified with a PAL-1[®] manual digital refractometer (Atago, Tokyo, Japan).

Ascorbic acid (AA)

The assay was carried out by the method described by Jagota and Dani (1982), with slight modifications. A 1 g sample was mixed with 5 mL of 10% trichloroacetic acid, homogenized and agitated vigorously for 5 minutes in the cold (ice), after which it was centrifuged for 5 minutes at 17,000 rpm at 5°C. Subsequently, a 400 μ L aliquot of the supernatant of each sample was taken and 2.2 mL of double-distilled? water and 400 μ L of 10% (v/v) Folin-Ciocalteu reagent were added to it. The mixture was stirred vigorously for 15 seconds and allowed to stand for 10 minutes; it was subsequently

used to obtain the absorbance reading ($\lambda = 760$ nm) using a Genesys 10-STM spectrophotometer (Thermo scientific, Florida, USA). The concentration was expressed in mg 100 g⁻¹ FW after calibration of a standard curve for ascorbic acid.

Colour

Its determination was made at the equatorial zone of the internal side of the ligulate flower by means of an SP62[®] sphere colorimeter (X-Rite, Michigan, USA) to obtain the parameters of brightness (L*), angle of tone (h°) and the purity of the colour (chroma, C*).

Phenolic compounds (TP)

The determination of TP was carried out by the Folin-Ciocalteu method described by Waterman and Mole (1994), for which a 0.2 g sample was taken and mixed with 5 mL of 70% absolute ethyl alcohol (v/v); from this extract 50 μ L was taken, to which 7.950 mL of distilled water and 500 µL of the Folin-Ciocalteu reagent (2N) were added; the mixture was stirred and left to stand for 8 minutes. Subsequently, 1.5 mL of 20% (w:v) Na₂CO₃ was added, stirred and allowed to stand for 2 h under conditions of total darkness at room temperature $(22 \pm 1^{\circ}C)$. The absorbance value ($\lambda = 760$ nm) of the mixture was obtained with a Genesys 10-STM spectrophotometer (Thermo Scientific, Florida, USA). The concentration of total phenols was expressed as mg of gallic acid g⁻¹ FW.

Total flavonoids (TFl)

The TFl were determined according to the method described by Loizzo et al. (2016), with slight modifications. A 1 g sample was taken and 5 mL of methanol was added; the mixture was homogenized and kept at room temperature until the methanol evaporated and the extract was dry. After taking 0.01 g of the dried extract, 5 mL of distilled water were added to it and stirred for 20 minutes. From that solution, 650 µl was taken and 75 μ L of NaNO₂ at 5% (v/v) was added, giving it 6 minutes of agitation, followed by 6 minutes at rest. Next, 150 µL of AlCl, was added to a concentration of 10%, giving it 4 minutes of agitation, followed by 5 minutes at rest; then 500 µL of NaOH (1M) and 1150 µL of distilled water were added. Next, the absorbance reading ($\lambda = 510$ nm) was taken with a Genesys 10-STM spectrophotometer (Thermo scientific, Florida, USA). The concentration of total flavonoids was expressed as mg of quercetin g⁻¹ of dry weight (DW).

Total anthocyanins (TAn)

The TAn concentration was determined with the differential pH method described by Giusti and Wrolstad (2011), for which a 0.2 g sample was taken and homogenized with 10 mL of 80% (v/v) absolute ethyl alcohol. Subsequently, 800 μ L of the extract was taken separately, 3.2 mL of a buffer solution at pH = 1 (0.025 M KCl) and 3.2 mL of a buffer solution at pH = 4.5 (0.4 M CH₃CO₂Na 3H₂O) were added to another sample. The absorbance readings of each mixture was taken at $\lambda = 510$ and 700 nm in a Genesys 10-STM spectrophotometer (Thermo Scientific, Florida, USA). The total absorbance (At) of the sample was determined from the formula:

At = $\left[(A510 - A710) \text{ pH} = 1.0 \right] - \left[(A510 - A700) \text{ pH} = 4.5 \right].$

The concentration of total anthocyanins was calculated according to the formula:

$$TAn = \left[\frac{Ta \ x \ MW \ x \ DF \ x \ 1000}{\varepsilon \ x \ 1}\right]$$

where AT is total absorbance, MW = molecular weight of the molecular standard (cyanidin-3glucoside) (449.2 g mol⁻¹), DF = dilution factor (1), \mathcal{E} = is the molar absorptivity coefficient from cyanidin-3-glucoside (26,900). The results are expressed in mg of cyanidin-3-glucoside per 100 g⁻¹ FW.

Antioxidant capacity (AC)

The determination of AC was carried out with the method employing ABTS [2,2-azino-bis (3-ethylbenzothiazolin-6-sulfonic acid)], modified by Ozgen et al. (2006), for which the radical ABTS⁺⁺ was formed after the reaction of ABTS (7 mM) with potassium persulfate (2.45 mM, final concentration) incubated at room temperature and in the dark for 16 hours. After the radical ABTS⁺⁺ was formed, it was diluted with PBS (phosphate buffer solution) (pH 7.4) until a value of absorbance from 0.7 ± 0.1 to 734 nm (maximum absorption length). For the assay, 3.98 mL of the ABTS⁺⁺ solution and 20 µL of the sample extract were mixed, and allowed to stand for 2 hours; the absorbance reading was taken at $\lambda = 734$ nm. The results are expressed in AAEVC (Antioxidant Activity Equivalent to Vitamin C).

Proximate composition

The standard AOAC methods were used to determine moisture, dry matter, crude protein, raw fat, raw fibre, total carbohydrates and ash (AOAC, 1990).

Statistical analysis

For the analysis of the data, analyses of variance (ANOVA) and the Tukey multiple comparison of means ($p \le 0.05$) were performed, for which the statistical program SAS version 9.0 was used.

RESULTS AND DISCUSSION

Physicochemical quality

The results of fresh weight and titratable acidity are shown in Table 1. On day 1, after cutting the flowers, it was observed that the ligulate cherry, yellow, and white flowers of D. × hortorum had the highest values of fresh weight, with 1.6, 1.54 and 1.75 g, respectively; however, these values were not statistically significantly different from what was shown by the red ligules (1.29 g) of the same species. On day 3, the cherry and white ligules stood out the most, with a tendency to increase the weight values, while on the fifth day this behaviour was reversed and those with greater weights were the cherry, yellow, and white ligules. Similar results have been reported in several other species such as Iris versicolor (Ahmad and Tahir, 2016), Gerbera jamesonii (Babarabie et al., 2016), Lilium spp., and Rosa hybrid (Ren et al., 2017).

The accumulation of citric acid on day 1 was higher for *D. brevis*, *D. merckii* and *D. coccinea* (1.4, 1.2 and 1.3 %, respectively), however, on day 3, it decreased to 0.7 and 0.9% (*D. merckii* and *D. coccinea*), increasing again on day 5 to 2.7 and 3.1%, while the lowest values were present in *D.* × hortorum in the yellow, white and cherry ligules. In this regard, Ahmad and Tahir (2016) indicate that cut flowers have a temporary variation in their fresh weight due to the opening of the floral bud; however, this fresh weight decreases as translocation of nutrients from other organs occurs (leaves and stems), which concludes with senescence (Ren et al., 2017), since, according to Lara-Cortés et al. (2016) and Azuma et al. (2019), dahlia inflorescences do not show sensitivity to the presence of external ethylene.

On the other hand, Gebremedhin et al. (2013) and Azuma et al. (2019) indicate that the low concentration of soluble carbohydrates that can be used as cellular respiration substrate prevent maintaining the integrity of the membrane, cell wall and synthesis of pigments, which makes it difficult to maintain life in a vase of cut flowers. In this sense, the TSS concentration fluctuated between 0.06 and 0.24 °Brix, with the highest values in the ligulate flowers (cherry, yellow and white) of D. × hortorum, in which the trend was similar to that observed in fresh weight and TTA, where the wild species did not show significant values (Tab. 2). Similar results had been reported in Gerbera jamesonii flowers (Babarabie et al., 2016) and in the 'Red Sky' and 'Blizzard' varieties of rosebush (Gebremedhin et al., 2013).

Among the nutritional components present in the ligulate flowers of dahlia, there was ascorbic acid, which was higher in *D. coccinea*, with values of 0.9, 0.77 and 0.89 mg g⁻¹ FW, and the lowest in *D. brevis* and *D. merckii*, with 0.31 and 0.300 mg g⁻¹ FW on day 1; however, on days 3 and 5 the lowest contents were in *D. merckii* (0.19 and 0.25 mg g⁻¹ FW, respectively) and *D.* × *hortorum* with purple ligules (0.20 and 0.21 mg g⁻¹ FW, respectively). These results exceed 0.5 µg of ascorbic acid g⁻¹ of ligule indicated by Lara-Cortés et al. (2014) for different varieties and colours of dahlia, in which

Species		Fresh weight (g)			Titratable acidity (% citric acid)			
	Day 1	Day 3	Day 5	Day 1	Day 3	Day 5		
D. brevis	0.23 d*	0.31 e	0.09 d	1.4 a	0.6 b-d	2.9 a		
D. merckii	0.17 d	0.22 e	0.10 d	1.2 a	0.7 ab	2.7 a		
D. coccinea	0.19 d	0.26 e	0.08 d	1.3 a	0.9 a	3.1 a		
D. campanulata	0.67 cd	0.85 d	0.41 cd	0.6 bc	0.4 d	1.4 b		
$D. \times hortorum$								
Cherry	1.6 a	2.01 ab	1.98 a	0.7 bc	0.5 b-d	0.6 c		
Yellow	1.54 a	1.76 bc	1.51 ab	0.4 c	0.5 b-d	0.7 bc		
White	1.75 a	2.28 a	1.78 a	0.6 bc	0.4 d	1.1 bc		
Purple	0.85 bc	1.01 d	0.64 cd	0.8 b	0.7 a-c	0.8 bc		
Red	1.29 ab	1.65 c	0.99 bc	0.6 bc	0.5 cd	0.8 bc		

Table 1. Changes in mean values of fresh weight and titratable acidity of ligulate flowers of some wild dahlia species

*Values with the same letter within a column are not statistically different according to the Tukey test ($p \le 0.05$)

Species	Total soluble solids (°Brix)			Ascorbic acid (mg g ⁻¹ FW)			
	Day 1	Day 3	Day 5	Day 1	Day 3	Day 5	
D. brevis	0.10 b	0.12 b	0.06 b	0.31 ef	0.26 e	0.27 f	
D. merckii	0.10 b	0.10 b	0.08 ab	0.30 f	0.19 f	0.25 fg	
D. coccinea	0.14 ab	0.16 ab	0.12 ab	0.90 a	0.77 a	0.89 a	
D. campanulata	0.10 b	0.16 ab	0.10 ab	0.40 d	0.36 d	0.39 de	
D. × hortorum							
Cherry	0.20 a	0.16 ab	0.10 ab	0.61 b	0.42 b	0.49 b	
Yellow	0.20 a	0.24 a	0.14 a	0.43 d	0.37 cd	0.42cd	
White	0.14 ab	0.24 a	0.10 ab	0.34 e	0.33 d	0.36 e	
Purple	0.18 a	0.10 b	0.06 b	0.22 g	0.20 f	0.21 g	
Red	0.20 a	0.18 ab	0.10 ab	0.48 c	0.41 bc	0.47 bc	

Table 2. Total soluble solids and ascorbic acid contents in ligulate flowers of some wild dahlia species

*Values with the same letter within a column are not statistically different according to the Tukey test ($p \le 0.05$)

the variation is determined by the balance between biosynthesis and degradation, as well as between reduction and oxidation (Huang et al., 2014), which is part of the role played by this vitamin as a watersoluble antioxidant and the conditions in which the plant or organ develops post-harvest (Liu et al., 2016).

The colour of the floral structures constitutes one of the most important characteristics with respect to the post-harvest handling of cut flowers (Zhang et al., 2015). In this work, the colour brilliance (L^*) was greater for D. × hortorum with yellow flowers (80.28, 84.49 and 87.52) and white flowers (81.52, 82.47 and 89.11) (Tab. 3), while those of cherry colour showed the lowest brightness (10.46, 22.29 and 16.12), which is associated with the absorption of a greater range of wavelengths of light, given its darker appearance, since, according to Arellano et al. (2015), low values of the L* component contribute to the visualization of darker colours and a high concentration of secondary pigments (anthocyanins and carotenoids) (Chen and Wei, 2017).

In the evaluation period, the yellow flowers showed greater purity of colour (C*) in D. × *hortorum* (93.79, 90.47 and 94.24), followed by D. *coccinea* (63.49, 70.64 and 55.61), with lower values observed in D. *campanulata* (Tab. 3). Younis et al. (2014), when analysing varieties with different colours, had found that the clear varieties (white) showed the highest L* values and the lowest C* values, which agrees with the results obtained in this research. On the other hand, higher values of C* are usually more attractive to the human eye (Yang et al., 2014).

D. campanulata had ligulate flowers with the highest colour tonality (h°), with values of 338.50, 345.46 and 350.74, followed by $D. \times hortorum$ with purple flowers (332.92, 333.63 and 318.92), and the lowest values in D. × hortorum with red flowers (25.22, 33.04 and 32.34). The variability with respect to colour tonality can be explained by the same colour differences in each species analysed as a result of the concentration of secondary pigments, as indicated for flowers of 10 species of Rhododendron (Liu et al., 2016). Likewise, it is important to point out that colour is one of the most important factors in the selection and use of flowers as food (Lee et al., 2009; Lara-Cortés et al., 2016) and its conservation is influenced by a great diversity of factors, among them the genotype and the management conditions postharvest (Ren et al., 2017). However, a low concentration of carbohydrates can accelerate the degradation of pigments and contribute to discoloration in cut flowers (Li et al., 2015; Azuma et al., 2019).

D. coccinea showed statistically the highest values of TP (15.29, 12.87, 19.92 mg GAE g⁻¹ FW); however, on day 5, cherry and yellow ligules (D. × hortorum) were similar, with 7.29 and 7.82 mg GAE g⁻¹ FW, respectively (Tab 4). A similar behaviour had been observed by Ahmad and Tahir (2016) in flowers of the genus *Iris* (*I. germanica* and *I. kashmiriana*), which the authors associate with failures in the redistribution of these compounds between organs, as well as the increase in their synthesis as a mechanism of defence to delay senescence, given the ability of these compounds to inhibit the activity of free radicals (Zeng et al., 2014).

Species	Day 1	Day 3	Day 5
-		Brightness (L*)	
D. brevis	40.47 d*	55.86 d	51.18 c
D. merckii	50.93 c	60.64 c	54.44 c
D. coccinea	35.85 e	46.07 e	42.42 d
D. campanulata	74.91 b	76.47 b	73.29 b
D. × hortorum			
Cherry	10.46 g	22.29 g	16.12 f
Yellow	80.28 a	84.49 a	87.52 a
White	81.52 a	82.47 a	89.11 a
Purple	26.92 f	27.11 f	29.15 e
Red	24.55 f	27.12 f	29.46 e
		Purity (C*)	
D. brevis	33.21 d	28.27 d	19.70 e
D. merckii	27.75 d	18.31 e	16.86 e
D. coccinea	63.49 b	70.64 b	55.61 b
D. campanulata	5.37 f	5.92 f	5.05 f
D. × hortorum			
Cherry	20.18 e	42.55 c	35.34 d
Yellow	93.79 a	90.47 a	94.24 a
White	20.19 e	15.96 e	16.76 e
Purple	45.09 c	40.94 c	44.86 c
Red	39.56 c	41.09 c	48.49 c
		Tonality (h°)	
D. brevis	304.48 c	314.05 c	307.21 c
D. merckii	306.00 c	315.30 c	310.37 с
D. coccinea	52.66 e	53.95 e	61.28 f
D. campanulata	338.50 a	345.46 a	350.74 a
D. × hortorum			
Cherry	15.91 g	20.56 g	22.05 h
Yellow	93.79 d	96.72 d	94.72 e
White	95.00 d	97.85 d	100.07 d
Purple	332.92 b	333.63 b	318.92 b
Red	25.22 f	33.04 f	32.34 g

Table 3. Colour components of ligulate flowers of some wild dahlia species

*Values with the same letter within a column are not statistically different according to the Tukey test ($p \le 0.05$)

The TP observed in *D. coccinea* was lower than that observed by Navarro-González et al. (2015) in orange flowers of *Tagetes erecta* (26.63 mg GAE g^{-1} FW). Orange flowers of *Tropaeolum majus* are reported with a higher concentration of TP (907.05 \pm 10.36 mg GAE g^{-1} FW) than the red and yellow flowers (799.35 \pm 10.77 and 442.02 \pm 5.09 mg GAE g^{-1} FW, respectively) (Arellano et al., 2015). For their part, Lara-Cortés et al. (2014) indicate values of 127.5 mg GAE g^{-1} FW for dahlia with purple flowers, in addition to gallic acid and caffeic acid as the compounds responsible for this concentration, because species such as *D. mignon* show higher concentrations of chlorogenic acid (Pires et al., 2018). Likewise, mean values between 2.53 and 5.11 g of gallic acid per kg of fresh weight are reported by Rop et al. (2012) for 12 species with edible flowers, among them, *Dianthus caryophyllus*, *Tagetes patula*, *Rosa odorata*, *Fuchsia* × *hybrida* and *Viola* × *wittrockiana*.

During the evaluation period, the highest concentration of TFl was observed in the wild species *D. coccinea* (8.15, 6.78 and 11.32 mg QE g⁻¹ DW), followed by *D.* × *hortorum* with cherry and yellow flowers (Tab 4). These results are comparable to those reported in edible flowers of

Species		Total phenols (mg GAE g ⁻¹ FW)			Total flavonoids (mg QE g ⁻¹ FW)			
	Day 1	Day 3	Day 5	Day 1	Day 3	Day 5		
D. brevis	11.91 c*	7.26 c	10.23 f	1.82 cd	1.23 ef	1.72 d		
D. merckii	10.60 de	7.02 c	8.03 g	1.03 d	0.69 f	0.90 e		
D. coccinea	15.29 a	12.87 a	19.92 a	8.15 a	6.78 a	11.32 a		
D. campanulata	1.66 g	1.25 d	2.55 h	1.02 d	0.76 f	1.47 de		
$D. \times hortorum$								
Cherry	12.06 bc	10.77 b	11.70 cd	7.29 a	5.18 b	7.17 b		
Yellow	9.81 e	9.93 b	12.36 c	7.82 a	6.23 a	7.52 b		
White	6.98 f	7.08 c	10.36 ef	1.90 cd	1.81 de	2.04 d		
Purple	13.11 b	12.02 a	13.94 b	2.00 cd	1.94 d	2.08 d		
Red	11.41 cd	7.10 c	11.02 de	4.21 b	4.12 c	4.55 c		

 Table 4. Changes in the concentration of total phenols and total flavonoids in ligulate flowers of some wild dahlia species

*Values with the same letter within a column are not statistically different according to the Tukey test ($p \le 0.05$)

Capparis spinosa and Hedysarum coronarium (5.8 and 8.0 mg QE g⁻¹ of dry extract, respectively), but lower than those of Sambucus nigra (23.6 mg QE g⁻¹ dry extract) indicated by Loizzo et al. (2016), who also found that routine and quercetin were the main flavonoids. The higher concentration of flavonoids in D. coccinea, whose flowers are orange, confirms that the colour orange is related to the presence of flavonoids (Garzón et al., 2015), as does the yellow colour of the flowers of different edible plants (Liu et al., 2016). Similarly, it is important to note that D. coccinea showed a significant increase in the TFl value at the end of the evaluation, which may be associated with the presence of specific flavonoids (quercetin, naringenin, butein and apigenin) in the dahlia petals (Lara-Cortés et al., 2014; Pires et al., 2018), given their ability to eliminate reactive

oxygen species as a response to the beginning of the senescence process, as described by Zhao et al. (2012) in flowers of three cultivars of *Paeonia lactiflora*.

As can be seen in Table 5, the cultivated species ($D. \times hortorum$) of cherry colour showed the highest concentration of TAn (14.74, 12.03 and 11.97 mg of cyanidin-3-glucoside 100 g⁻¹ FW). These results were lower than those reported by Garzón et al. (2015) in orange and red flowers of *Tropaeolum majus* (108.8 and 168.0 mg of cyanidin-3-glucoside 100 g⁻¹ FW, respectively); however, they are similar to those obtained by Benvenuti et al. (2016) in red flowers of *Dianthus* × *barbatus*, *Pelargonium peltatum* and *Viola* × *wittrockiana* (13.3, 12.5 and 12.4 mg cyanidin-3-glucoside 100 g⁻¹ FW, respectively). Lara-Cortés et

Table 5. Changes in the concentration of total anthocyanins and antioxidant capacity in ligulate flowers of som	e wild
species of dahlia	

Species	Total anthocyanins (mg of cyanidin-3-glucoside100 g ⁻¹ FW)			Antioxidant capacity (mg AAEVC g ⁻¹ FW)			
	Day 1	Day 3	Day 5	Day 1	Day 3	Day 5	
D. brevis	2.24 d*	1.87 d	2.11 d	16.10 e	13.72 d	15.01 e	
D. merckii	1.66 e	1.53 d	1.78 de	15.46 e	13.50 d	15.54 e	
D. coccinea	1.31 f	1.17 e	1.52 e	57.32 a	47.28 a	60.43 a	
D. campanulata	0.26 g	0.24 f	0.43 f	7.24 f	6.82 e	11.79 f	
D. × hortorum							
Cherry	14.74 a	12.03 a	11.97 a	22.97 с	18.00 c	21.89 d	
Yellow	0.24 g	0.11 f	0.10 fg	20.86 cd	18.24 c	22.77 cd	
White	0.09 g	0.07 f	0.06 g	15.11 e	13.60 d	15.60 e	
Purple	5.17 b	4.94 b	5.02 b	19.97 d	18.59 c	23.08 c	
Red	4.01 c	3.66 c	4.02 c	34.99 b	22.75 b	39.68 b	

*Values with the same letter within a column are not statistically different according to the Tukey test ($p \le 0.05$)

al. (2014) found the highest concentration of total anthocyanins in purple dahlia flowers (257.5 mg 100 g⁻¹ FW), while the white and yellow flowers had the lowest content, which is consistent with the results obtained in this research, where the flowers of more intense colour registered higher concentrations of anthocyanins than the white or clear flowers. In this sense, Yamaguchi et al. (1999) indicate a higher concentration of anthocyanins in pink, orange, red and purple colours, in contrast to yellows and whites, where they were not detected, according to these authors, the concentration of anthocyanins determined the colour intensity of the flowers. The concentration of anthocyanins can be affected by different factors such as stress due to environmental conditions, genetic aspects, variety and agronomic management (Lee et al., 2009). In addition, the concentration of total anthocyanins varies throughout the flowering stages (Lara-Cortés et al., 2013).

The wild species D. coccinea stands out for its high AC (57.32, 47.28 and 60.43 mg AAEVC g⁻¹ FW), followed by the cultivated species D_{\cdot} × hortorum with red flowers (34.99, 22.75 and 39.68 mg AAEVC g⁻¹ FW) (Tab. 5). The behaviour observed in D. coccinea (orange flowers), agrees with the results obtained by Benvenuti et al. (2016), who found greater antioxidant activity in the flowers of Tagetes erecta (70.42 mmol $FeSO_4$ 100 g⁻¹ FW), followed by the red flowers of Fuchsia hybrida, Dianthus × barbatus, Viola × wittrockiana and Pelargonium peltatum (47.52, 38.67, 36.55 and 34.78 mmol FeSO, 100 g⁻¹ FW, respectively). Similarly, Arellano et al. (2015) reported the highest antioxidant capacity in orange flowers of T. majus (DPPH: 3928.226 µM TE g⁻¹ DW, ABTS: 622.92 μ M TE g⁻¹ dry sample) in relation to red and yellow flowers.

On the other hand, the lowest values of AC are for *D. campanulata* (7.24, 6.82 and 11.79 mg AAEVC g⁻¹ FW), results that are similar to those reported by Rop et al. (2012) in edible flowers of 12 plant species (4.21 to 6.96 g VEAC kg⁻¹ FW). In this sense, the colour of the flowers is related to AC in such a way that intense colours (red and blue) are good indicators of this parameter, unlike the white colour that is associated with low values (Benvenuti et al., 2016). Besides, AC can be influenced differently by the phytochemicals present in flowers, so that a high antioxidant capacity can be mainly due to flavonoids or carotenoids, instead of anthocyanins (Chen and Wei, 2017).

Nutritional value

The data resulting from the analysis of the proximate composition are shown in Table 6. Among the wild species, with the exception of D. campanulata (93.4%), the moisture content did not exceed 90%, where D. coccinea was the lowest in moisture but the highest in DM - a result that confirms the correlation between M and DM. It was a behaviour that agrees with that obtained by Lara-Cortés et al. (2014) in dahlia flowers (88 to 92%), as well as with the data obtained in edible flowers of T. majus, Tagetes erecta and Spilanthes oleracea (89.3, 83.4 and 81.7%, respectively) (Navarro-González et al., 2015), in which it is important to point out that the accumulation of DM is associated with the nutritional contribution during the growth and development of the crop, so it varies depending on time.

Species	(%)							
	М	DM	СР	RF	RFi	TC	А	
D. brevis	88.8 h*	11.2 b	12.7 d	3.3 a	11.6 e	65.1 d	7.3 b	
D. merckii	89.5 g	10.5 c	14.8 b	3.4 a	11.3 ef	64.0 f	6.5 c	
D. coccinea	85.9 i	14.1 a	13.5 c	2.6 b	9.8 g	68.6 a	5.5 g	
D. campanulata	93.4 a	6.6 i	13.7 c	3.4 a	14.3 a	59.9 g	8.7 a	
D. × hortorum								
Cherry	89.84 f	10.1 d	14.3 b	1.5 e	11.2 ef	67.0 b	5.9 f	
Yellow	91.0 c	8.9 g	16.2 a	2.0 d	10.9 f	64.3 ef	6.6 c	
White	90.4 e	9.6 e	13.5 c	2.2 c	12.1 d	65.8 c	6.4 d	
Purple	90.5 d	9.5 f	11.9 e	2.2 cd	12.6 c	67.0 b	6.2 e	
Red	91.4 b	8.5 h	13.5 c	2.3 c	13.8 b	64.7 de	5.8 f	

Note: M: moisture; DM: dry matter; CP: crude protein; RF: raw fat; RFi: raw fibre; TC: total carbohydrates; A: ash. Data is expressed on dry weight basis. *Means with the same letter within columns are not statistically different according to the Tukey test ($p \le 0.05$)

The CP content was higher in D. \times hortorum yellow flowers (16.2%) than in the purple flowers of the same species (11.9%). In contrast, Lara-Cortés et al. (2014) report for dahlia flowers CP values between 0.8-4.0%, however, they are similar to those in edible flowers of Agave salmiana and Aloe vera (16.4 \pm 3 and 16.4 \pm 2 g 100 g⁻¹ DW, respectively) reported by Lara-Cortés et al. (2013). On the other hand, D. brevis, D. merckii and D. campanulata were the most outstanding species with respect to the RF content (3.3, 3.4 and 3.4%, respectively), while the lowest RF percentage was for the cherry D. × hortorum (1.5%). These results were slightly higher than those reported by Lara-Cortés et al. (2014) in dahlia flowers (0.5-1.6%). Because the main component of flowers is water, the fat content in them is minimal (Fernandes et al., 2018), so they are considered a low-calorie food (with a low calorific value) (López-Cervantes et al., 2018), recommended for consumption by people with obesity (Loizzo et al., 2016; Rachkeeree et al., 2018).

The RFi content fluctuated between 9.8 and 14.3%, in which *D. campanulata* stands out, followed by *D.* × *hortorum* with red flowers (13.8%), while the lowest percentage was in *D. coccinea*. Similar values are reported by Lara-Cortés et al. (2013) in *A. salmiana*, *Euphorbia radians*, *Arbutus xalapensis* and *A. vera* (12.7, 12.6, 10.4, and 13.8 g 100 g⁻¹ DW, respectively), so the dahlia species with a high RFi content could be a good alternative source of fibre to reduce the level of cholesterol (Rachkeeree et al., 2018) and help bowel evacuation (Fernandes et al., 2018).

The highest TC concentration was in *D. coccinea* (68.6%), followed by *D.* × *hortorum* with cherry and purple flowers (67.0% each), while the lowest value was presented by *D. campanulata* (59.9%). In this regard, López-Cervantes et al. (2018) indicate a TC concentration of 60.4-68.6% in *A. vera* flowers, which indicates that TC are the main nutrients, representing more than 50% of total dry weight.

With respect to the presence of ash, it was the highest in *D. campanulata* (8.7%), followed by *D. brevis* (7.3%), which exceeded that in the cultivated species D. × *hortorum* with yellow flowers (6.6%) by 32.2% and 10.3%, respectively, while the lowest content was presented by *D. coccinea* (5.5%). These results are similar to those indicated by Lara-Cortés et al. (2013) in the inflorescences of *A. salmiana*, *Erythrina americana*, *E. radians*, *Hibiscus sabdariffa*, *A. xalapensis* and *A. vera* (5.8,

9.6, 9.4, 6.5, 6.9 and 8.6 g 100 g¹ DW, respectively), with variations that could be associated with the nutritional status of the soil, plant species and organ, and the phenological stage at harvest (Sotelo et al., 2007; Juárez-Rosete et al., 2019).

CONCLUSIONS

The results of the study are the basis for formulating the following conclusions:

- 1. In the present study, the ligulate flowers of the wild species *D. coccinea* were the most outstanding with regard to the concentrations of ascorbic acid, TP, TFI and antioxidant capacity; however, *D.* × *hortorum* presented the highest values of fresh weight, TSS, colour parameters and total anthocyanins.
- Among the analysed wild species of dahlia, D. coccinea had the highest values of dry matter and total carbohydrates, whereas D. campanulata stood out for the highest fat content and crude fibre.
- 3. Intake of bioactive compounds through the incorporation of the ligulate flowers of wild species of dahlia into the diet can be an important alternative for the diversification in their use, as well as contribution to their conservation as plant genetic resources.

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AUTHOR CONTRIBUTIONS

E.A.R.E., M.R.G.M., J.M.M.M. and M.T.B.C.L. – designed the experiments and performed analytical measurements; E.A.R.E, O.C.A and M.T.M.D – collected data and performed analyses; M.T.M.D and O.C.A. – analysed the data and wrote the paper.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

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