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NUTRITIONAL VALUE, BIOACTIVE COMPOUNDS AND CAPACITY ANTIOXIDANT IN EDIBLE FLOWERS OF DAHLIA

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ABSTRACT

The aim of this research was to evaluate the nutritional value, bioactive compounds and capacity antioxidant in ligulate flowers of dahlia with different colour. The statistical analysis reveals that the petals of the pink and purple colour have more protein and raw fibre (18.54 and 19.25, 16.54 and 15.81%, respectively) and similar raw fat values between the analysed colours. Likewise, the flowers with the highest content of vitamin C, phenols and total flavonoids were observed in the pink, purple and cherry colours, where the latter also stands out for its content of total anthocyanins (3.2 mg cyanidin-3-glucoside 100 g^{-1}). However, the yellow ones showed higher antioxidant capacity (68.82 mg AAEVC 100 g⁻¹), which coincides with that found with the concentration of total carotenoids (0.64 μ g β -carotene g⁻¹). For the consumers who choose to change their eating habits and seek to include ingredients of vegetable origin (vegetarian and vegan) with low caloric content and a sufficient supply of protein and raw fibre, the ligulate flowers of dahlia can be considered an important alternative.

Key words: *Dahlia* × *hortorum*, floriphagia, functional foods, vegan, vegetarian

INTRODUCTION

In Latin America the consumption of flowers and inflorescences dates from pre-Hispanic times and remains part of the traditional cuisine of many populations settled mainly in rural areas [Lara-Cortes et al. 2014], among them, pumpkin (Cucurbita pepo L.) and sunflower (Helianthus annuus L.) [Navarro et al. 2015, Costa-Silva et al. 2020], as well as those that are collected (wild) such as maguey pulquero (Agave salmiana Gentry), aloe (Aloe vera (L.) Burm f.), colorin (Ervthrina coralloides DC.), izote (Yucca faxoniana Sarg.), among others [Sotelo et al. 2007, Juárez et al. 2018, López et al. 2018]. It is common for them to be cooked and combined with other ingredients to create local recipes that are highly valued from a gastronomic point of view by haute cuisine restaurants and as cultural heritage for the population where they are generated, because they often possess a very nice aroma, shape, taste and texture [Li et al. 2017].

A particular case is the dahlia, an ornamental plant included in the genus Dahlia (Asteraceae: Coreopsideae) composed of 37 species [Shimizu and Ichimura 2013], most of them are native to Mexico and are considered for flower production cutting, potting and as a structural element in the design and management of gardens [Lu et al. 2015, Ohno et al. 2016]. However, segments of the population that wish to change and improve their eating habits (vegans and vegetarians), have included it in their diet [Lara et al. 2014,



Grzeszczuk et al. 2016], which is not new, since it is known that in some indigenous groups of Oaxaca, Mexico, the petals of the dahlia are used in the preparation of small cakes, salads, desserts and as a garnish in various local dishes [Grzeszczuk et al. 2016, Fernandes et al. 2017].

Among the most important consumer demands, the concentration of compounds that have the ability to prevent, reduce, delay or inhibit the harmful effect caused by the production and excessive accumulation of reactive oxygen species (superoxide radical anion, hydrogen peroxide, hydroxyl ion, among others) [Benvenuti et al. 2016], and with this, contribute to health care, when consumed [Rodríguez et al. 2017]. This is one of the reasons for the increase in the production and commercialization of crops such as blueberry, raspberry, pepper, aromatic species and in general products with yellow, blue and red tones [Chen et al. 2018, Frias-Moreno et al. 2019]. In this sense, dahlia flowers have a great diversity of shapes, sizes, as well as colours, due to their high concentration of phenolic compounds (gallic acid, caffeic acid, hydroxybenzoic acid, chlorogenic acid, synaptic acid

and coumaric acid) and flavonoids (quercetin, naringenin, hesperidin and routine) [Lara et al. 2014], compounds that are associated with their high antioxidant capacity [Ohno et al. 2016, Wu et al. 2018], characteristics that make them attractive to be consumed as a fresh product [López et al. 2018]. That is why the objective of this research was to determine the nutritional value, the bioactive compounds and the antioxidant capacity in ligulate flowers of dahlia with different colour.

MATERIAL AND METHODS

Plant material, crop management and experimental design. The present research was carried out with ligulate flowers of six dahlia clones (*Dahlia* × *hortorum*) with different colours: white (L* 82.05, C* 19.41, h° -73.04), yellow (L* 80.05, C* 62.17, h° -73.47), variegated (L* 28.07, C* 59.15, h° 14.67), pink (L* 48.28, C* 25.73, h° 6.85), purple (L* 44.66, C* 24.00, h° 6.61) and cherry (L* 30.14, C* 15.32, h° 62.15) – Figure 1. The initial concentration of bioactive compounds and antioxidant capacity is also determined (Tab. 1).

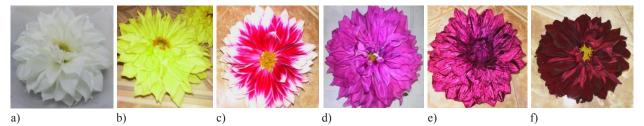


Fig. 1. Colour of the ligulate flowers (*Dahlia* \times *hortorum*) evaluated in this study a) white; b) yellow; c) variegated; d) pink; e) purple and f) cherry

Colour	VC	TP	TC	TFl	ТА	AC
White	0.10	8.94	0.12	7.02	0.006	58.45
Yellow	0.13	5.78	0.50	40.48	0.00073	60.12
Variegated	0.12	11.50	0.27	10.95	0.32	59.65
Pink	0.18	13.58	0.51	57.89	0.11	60.25
Purple	0.15	10.02	0.79	82.98	0.63	57.28
Cherry	0.16	13.98	0.15	58.58	1.68	47.25

Table 1. Initial concentration of bioactive compounds and antioxidant capacity in ligulate flowers of dahlia (D. × hortorum)

VC: vitamin C (mg ascorbic acid 100 g⁻¹); TP: total phenols (mg GAE 100 g⁻¹); TC: total carotenoids ($\mu g \beta$ -carotene g⁻¹); TFI: total flavonoids (mg QE 100 g⁻¹); TA: total anthocyanins (mg cyanidin-3-glucoside 100 g⁻¹); AC: antioxidant capacity (mg AAEVC 100 g⁻¹). Data is expressed on fresh weight basis.

These materials were obtained from the Experimental Agricultural Field "San Martin" at Universidad Autonoma Chapingo (UACh) [19°29'23"N, 98°53'37"W and at 2,246 m above sea level (masl)] with an average annual temperature and precipitation of 18.9°C and 619.3 mm, respectively.

The cut of the flower head (inflorescences) was made during the spring-summer agricultural cycle (the month of August). This activity was carried out in the first hours of the day (between 6–7 in the morning) for the purpose of reducing the mechanical damage. Subsequently, the flowers (flower head) were transported in dry ice boxes to the plant physiology laboratory of the Department of Plant Science (UACh). The propagation and obtaining of the plant material, was done during in the first week of March 2017 with the sowing in boxes with peat, in greenhouse, of the tuberous roots obtained from the previous cycle, which buds emerged after 15 days. The field transplant was performed in the second week of April 2017, where the distance between plants and rows was 0.5 and 0.7 m, respectively. The supply of water and nutrients was carried out with a frequency of 1-2 times per week with a drip irrigation system and a water expenditure of 0.5-3.0 L plant⁻¹, depending on the ambient temperature, relative humidity and phenological stages of the crop. Fertilization based on a previous analysis of soil fertility and the fertilization formula 120–0–200, which was divided into two applications (at transplantation and prior to the beginning of flowering). Weed control and some crop management practices, such as corking, were handled manually.

The experimental design was completely randomized with five replications (biological replicates), where the experimental unit consisted of a clamshell type packing with dimensions of $181 \times 121 \times 86$ mm with 50 ± 0.5 g of inflorescences formed by ligulate and tubular flowers (flower head) and maintained at 8°C (refrigeration) and relative humidity of 80%, simulating the handling given for cut flowers. The evaluation was carried out during a period of six days, because on the eighth day the flowers showed very visible symptoms of dehydration and senescence (data not shown), therefore, the data collection process was interrupted.

Parameters evaluated. To determine moisture, dry matter, crude protein, raw fat and fibre, the method indicated by AOAC 934.01, 2001.11, 954.02, 962.09

[AOAC 2016] was used. The determination of vitamin C (VC) was carried out by the method proposed by the AOAC 967.21 [AOAC 1990]. The data was expressed as mg ascorbic acid 100 g⁻¹ fresh weight (FW). The content of total phenols (TP) was determined according to the Folin-Ciocalteu method described by Waterman and Mole [1994]. The results were expressed as mg gallic acid equivalents 100 g⁻¹ FW (mg GAE 100 g⁻¹). The total carotenoids were determined according to the technique described by Lichtenthaler [1987]. The data was reported in $\mu g \beta$ -carotene g⁻¹ FW. The content of total flavonoids (TFl) was quantified according to the method proposed by Chang et al. [2002]. The results were expressed as mg quercetin equivalents per 100 g of fresh weight (mg QE 100 g⁻¹ FW). The content of total anthocyanins (TA) was determined with the differential pH method described by Giusti and Wrolstad [2001]. The results are expressed as mg cyanidin-3-glucoside 100 g⁻¹ FW. The modified 2,2-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) method was use to measure antioxidant capacity (AC) [Ozgen et al. 2006]. The results were expressed in AAEVC (antioxidant activity equivalent to vitamin C). The data obtained were analysed by means of variance analysis (ANOVA) and multiple means comparison using the Tukey's test ($P \le 0.05$), in which the statistical analysis package SAS (Statistical Analysis System version 9.3) was used.

RESULTS AND DISCUSSION

Nutritional value: According to the moisture data obtained in the proximate analysis (Tab. 2), its average content was 91.75%, where purple flowers stand out (92.62%) with respect to white flowers (90.66%), however, it did not exceed what was shown by the rest of the colours analysed (yellow, variegated, pink and cherry), which values fluctuated between 91.13 and 91.92%. Lara et al. [2014], when evaluating ligulate flowers (purple, yellow, pink, white, cherry, orange and red) in several dahlia species (D. australis, D. appiculata, D. brevis, D. coccinea, D. campanulata and D. pinnata), report not having found variation with respect to this variable, however, the range of their data was similar (88 to 92%) to that found in this study and where, coincidentally, for the white flower, indicated the same value. On the other hand, Sotelo

Colour	Percentage						
Coloui	М	DM	СР	RF	F		
White	90.66 b	9.32 a	13.18 d	3.44 a	15.53 abc		
Yellow	91.92 ab	8.08 c	14.84 cd	3.38 a	14.68 bcd		
Variegated	91.76 ab	8.24 c	16.61 bc	2.89 a	13.99 cde		
Pink	91.13 ab	8.87 ab	18.54 ab	3.38 a	16.54 a		
Purple	92.62 a	7.37 d	19.25 a	3.05 a	15.81 ab		
Cherry	91.57 ab	8.44 bc	14.84 cd	2.89 a	12.55 e		

Table 2. Nutritional value on a dry basis of ligulate flowers of dahlia (D. × *hortorum*)

Means with the same letter within columns are equal according to the Tukey's test (P \leq 0.05).

M: moisture; DM: dry matter; CP: crude protein; RF: raw fat; F: fibre

et al. [2007] in other species of Mexican plants with flowers and edible inflorescences such as *A. salmiana* Gentry, *A. vera* Miller, *Arbutus xalapensis* Kunth, *C. pepo* L., *Erythrina Americana* Mill., *Erythrina caribaea* Krukoff & Barneby and *Yucca filifera* Chabaut, report a humidity percentage between 86 and 93.2. Additionally, in petals of other flowers such as *Parkia biglobosa* (Jacq.) R.Br. ex G.Don and *Tagetes erecta* L., lower values are reported with 75.5 \pm 0.87% and 83.39 \pm 0.17%, respectively [Hassan et al. 2011, Navarro et al. 2015], which could suggest that the dahlia may be highly susceptible to the development of fungi and bacteria, as part of the interaction between its high moisture content and environmental temperature as indicated by Li et al. [2017].

The relationship between moisture content and DM was observed along with its behaviour at the time of its evaluation, that is, although the flower head with white ligulate flowers had lower moisture content, it had the highest DM value (9.32%), similar case occurred with pink (8.87%). Results that coincide with those reported in aloe (A. vera (L.) Burm f.) by López et al. [2018], as well as in eight colours of ligulate flowers of different dahlia species [Lara et al. 2014]. With respect to the data obtained from CP, the pink and purple flowers (18.54 and 19.25%, respectively) stood out, which exceeded the data reported in different shades of dahlia colour by Lara et al. [2014] (0.8-4.0%), as well as that reported in other comestible flowers of T. erecta L. (1.32%) and Tropaeolum majus L. (1.99 $\pm 0.06\%$) and Spilanthes oleracea var.

fusca (Lam.) DC ($2.84 \pm 0.11\%$) [Navarro et al. 2015]. According to these results, the gastronomic use of dahlia flowers can be considered an important source of daily protein supply in human nutrition.

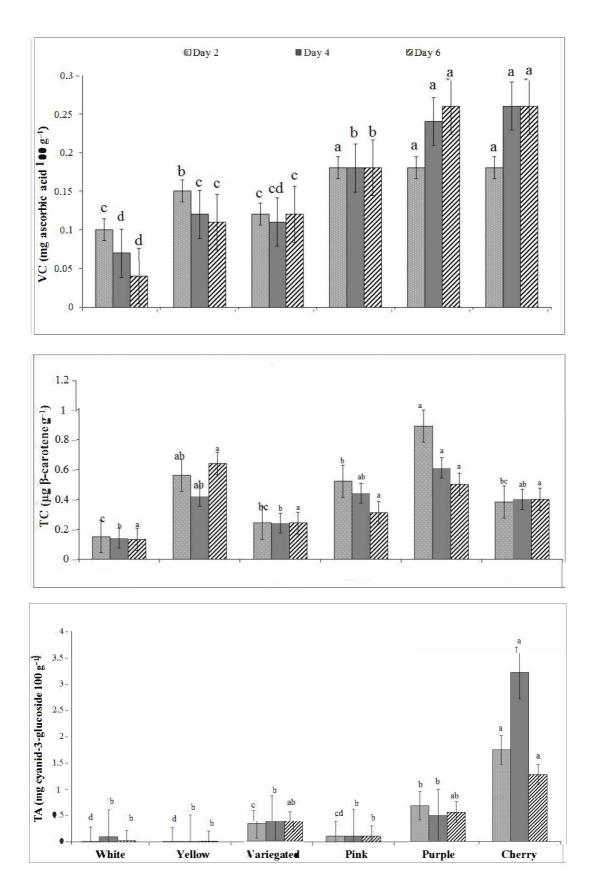
The flowers head did not show significant changes in the RF content with respect to the colour of the analysed material, which data fluctuated between 2.89 and 3.44%. In contrast, higher values are reported by Sotelo et al. [2007] in flowers of other plants such as aloe $(164 \pm 2 \text{ g kg}^{-1} \text{ dry weight})$ and arbutus (A. xalapensis Kunth) (113 ± 2 g kg⁻¹ dry weight). If we consider that consumers are currently looking for foods with low caloric levels, dahlia flowers can be an excellent alternative to maintain or reduce their body weight [Lara et al. 2014, Moldovan et al. 2017], also considering that there are statistical differences between them $(P \le 0.05)$ with respect to fibre content, which varied between 12.55 and 16.54% from the different colours of ligules, where rose, purple and white were highlighted with values of 16.54, 15.81 and 15.53%, respectively. These results surpass what is indicated for other flowers cultivated in Mexico such as roselle (Hibiscus sabdariffa L.) and cassava (Y. filifera Chabaut) [Sotelo et al. 2007].

Bioactive compounds and capacity antioxidant: The data obtained from bioactive compounds and antioxidant capacity evaluated in the ligulate flowers of dahlia are shown in Figure 2. At present, with the changes that have arisen in the habits of feeding between some sectors of the population, the characteristics of external quality (colour, form and size) do not

satisfy the needs of information for the decision making on the part of the consumers and the need arises to indicate nutritional aspects, among them, the content of ascorbic acid. In this research, on the second day of evaluation, a value of 0.18 mg of ascorbic acid 100 g⁻¹ FW was found, with a similar behaviour among the pink, purple and cherry flowers, situation that changed for days 4 and 6, when an increase to 0.24 and 0.26 mg of ascorbic acid 100 g⁻¹ FW (flowers purple and cherry, respectively) was reported, and once comparing these results with those reported in flowers of the same genus but different species, these were higher with the indicated by Lara et al. [2014], in different colours (purple, cherry, pink, orange, yellow, red and white) with maximum values of 0.5 µg of ascorbic acid g⁻¹. However, these same authors indicate no variation between the colours analysed. Li et al. [2017]. when evaluating petals of light, dark colours and complex combinations among these of 46 varieties of Paeonia lacti lora Pall. reported values that fluctuated between 9.77-30.24 mg/100 g FW with a mean value of 14.88 ±4.94 mg/100 g FW, well above our data. Likewise, the process of cellular respiration, which being an oxidative process contributes to the generation of reactive oxygen species [Rodríguez et al. 2017], by which it activates and increases the activity of enzymes such as ascorbate peroxidase and ascorbate reductase responsible for detoxifying the cell, and which can play an important role in using ascorbic acid as a substrate, with the consequent variation in their levels [Lu et al. 2015, Petrova et al. 2016, Chen et al. 2018].

With respect to the concentration of TP, these compounds had an unclear behaviour, however, with the exception of the yellow colour (lower concentration), the rest of the materials presented a similar value between the days of evaluation. In this sense, Yang and Shin [2017] indicate that the variation in the concentration of these compounds is associated with the colour of the flower and its antioxidant activity. Additionally, the extraction methodologies for this type of compound are not homogeneous, which has repercussions on the type of phenolic compounds extracted and the concentration values reported. These results contrast with what was found by Lara et al. [2014] for pink flowers (127.5 mg gallic acid g⁻¹ DW) followed by yellow flowers (102.1 mg gallic acid g⁻¹ DW), however, the data observed in this study were higher than observed by these authors in red flowers (3.6 mg DW g⁻¹ gallic acid). On the other hand, 83.33% (white, purple, cherry, pink and variegated) of ligulate flowers presented contents of TP that exceeded flowers and comestible inflorescences from other species such as camellia (Camellia japonica L) -6.71 mg GAE g⁻¹ DW, globose amaranth (Gomphrena globosa L.) – 6.13 mg GAE g⁻¹ DW, lily (Lilium brownii F.E. Brown ex Miellez) – 6.39 mg GAE g⁻¹ DW - but were similar to those indicated for carnation (Dianthus caryophyllus L.) – 15.40 mg GAE g⁻¹ DW, loquat (Eriobotrya japonica (thunb.) lindl) - 13.19 mg GAE g⁻¹ DW and plumeria (Plumeria rubra L.) – 10.98 mg GAE g⁻¹ DW [Chen et al. 2018] and flowers well known in Mexico as T. erecta L. [Petrova et al. 2016] with values of 5.24 ± 0.03 mg EAG g⁻¹ FW.

Carotenoids are a group of isoprenoid compounds that besides being precursors of vitamin A [Fernandes et al. 2017], are also considered important from the nutritional point of view (antioxidant) [Kishimoto and Ohmiya 2009] and are related to the presence of colourful petals like yellow and red. In this study, the concentration of these organic pigments was very variable, that is, there were tendencies to maintain their concentration (variegated: 0.24 μ g β -carotene g⁻¹), slightly decrease (white: 0.15–0.13 μ g β -carotene g⁻¹, rose: 0.51–0.31 μ g β -carotene g⁻¹ and purple of 0.89– 0.50 μ g β -carotene g⁻¹), however, the yellow flowers showed an increase in the total carotenoid concentration (0.56–0.64 μ g g⁻¹) but its content was similar to what is observed in the colours purple, pink and cherry. Kishimoto and Ohmiya [2009] indicate that the presence of yellow petals in most species that make up the compositae family are the product of the presence of carotenoids, however, in dahlia, it is due to the synthesis and accumulation of buteins, coreopsine and some types of aurones. On the other hand, Lara et al. [2014] reports the presence of this compound in dahlia ligules in the species D. australis, D. appiculata, D. brevis, D. coccinea, D. campanulata and D. pinnata, in which the orange, purple, purple and white ligules stand out, however, as in this study, the values reported are higher than those reported in begonia (Begonia \times tuberhybrida Voss.) – 0.020 µg g⁻¹ FW, common lavender (Lavandula angustifolia Mill.) -0.164 µg g⁻¹ FW) and Salvia splendens Sellow ex



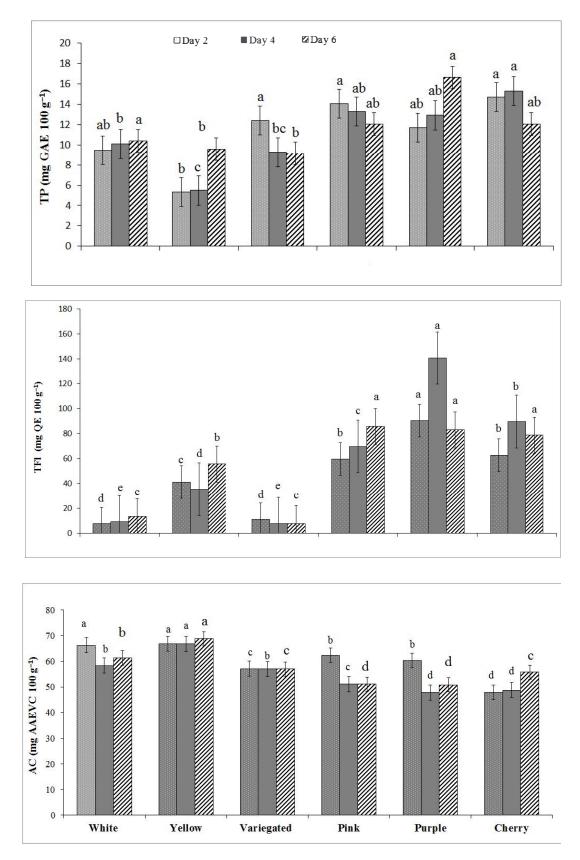


Fig. 2. Bioactive compounds and antioxidant capacity in ligulate flowers of dahlia (D. × *hortorum*). Data shown are mean values ±standard error (n = 5). Bars with the same letter are equal according to Tukey's test ($P \le 0.05$)

VC: vitamin C; TP: total phenols; TC: total carotenoids; TFI: total flavonoids; TA: total anthocyanins; AC: antioxidant capacity. Data is expressed on fresh weight basis.

Roem. et Shult. ($0.036 \ \mu g \ g^{-1} \ FW$) [Grzeszczuk et al. 2016] but remain at a 10 : 1 ratio with some rose species grown in Brazil (1.25 mg 100 g⁻¹) [Prata et al. 2017]. However, our values are comparable with that reported for *Tagetes tenuifolia* Cav. ($0.992 \ \mu g \ g^{-1} \ FW$) [Grzeszczuk et al. 2016], a flower well known in Mexico for its ritual use and in some cases food [Navarro et al. 2015, Petrova et al. 2016].

Perhaps one of the most studied compounds are the flavonoids due to their wide distribution in fruits and vegetables [Ohno et al. 2016] and for their proven efficiency as iron chelators and as oxygen scavengers [Lu et al. 2015, Wu et al. 2018]. In this sense, the content of total flavonoids showed a very variable behaviour between the days of evaluation and the colour of the flower analysed, this is how the white and variegated flowers showed the lowest values (7.85-13.36 and 7.9–11.03 mg QE 100 g⁻¹, respectively), it should also be mentioned that purple flowers did not vary during the three days of evaluation, with maximum values on day 4 (140.57 mg QE 100 g^{-1}), however, by day 6 this content decreased to 82.98 mg QE 100 g⁻¹, which was statistically similar to 85.72 and 78.69 mg QE 100 g^{-1} , observed in the colours pink and cherry.

Dahlia flowers are recognized by the presence of a wide range of bright colours, which expression is highly associated with the synthesis and accumulation of anthocyanins ($r^2 = 0.967$) similar with other pigments such as carotenoids ($r^2 = 0.996$) [Yamaguchi et al. 1999]. That makes imperative the need to assess their concentration at the time of its cutting and its changes during storage, in this work the ligule of cherry colour remained without significant variation throughout the evaluation period, but showed the highest values (1.26–3.20 mg of cyanidin-3-glucoside 100 g⁻¹ FW), however, at day 6 it was statistically similar to that observed in the variegated and purple flowers with 0.38 and 0.55 mg of cyanidin-3-glucoside 100 g⁻¹ FW. Anthocyanins are the most important functional group of phenolic and water soluble pigments present in plants and are responsible for the presence of horticultural products and their structures (petals) with red, purple and blue epidermis [Shimizu and Ichimura 2009, Benvenuti et al. 2016], where cyanidin 3,5-diglucoside (Ci 3G5G) and pelargonidin 3,5-diglucoside (Pg 3G5G) predominate, however, in white dahlias it is indicated by the presence of a mixture of 3G Pg [pelargonidin 3- (60-malonyl glucoside)] and Ci 3G [cyanidin 3-(60-malonyl glucoside)] and for the case of the yellow ones their presence of these same compounds but a concentration in a trace level content [Yamaguchi et al. 1999]. Likewise, Lara et al. [2014] highlight a behaviour similar to that observed in this study for the pink, purple and cherry tones, because they mention that the highest data were found in the darkest tones (purple and red), in the same way an also are comparable with the data reported for this pigment by Frias-Moreno et al. [2019] in raspberry (*Rubus idaeus* L.).

The highest values of AC found in this study correspond to those shown by the yellow flowers (66.78–68.82 mg 100 g^{-1} AAEVC) which exceeded the rest of the materials, except for the second day, where they were similar with those of white colour (66.31 mg 100 g^{-1} VEAC). Results that do not coincide with Yang and Shin [2017], who when evaluating petals of nine varieties of rose (Rosa hybrida spp.) cultivated in South Korea, report values between 1416.33-2370.77 mg VCE / 100 g⁻¹ FW, where the highest values were presented in the red flowers. On the other hand, Lara et al. [2014] report not having found variation of this parameter with respect to the colour tonality, however, these same authors found the lowest values in the white flowers and in our work was of the highest. Inter- and intraspecific variation, according to Rodríguez et al. [2017] may be associated with the method used to analyse and compare the efficiency of antioxidants present in flowers, because ABTS (2,2-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid) method requires a long time to complete the reaction, depends on the pH and susceptible to interference due to the generation of turbidity with other compounds (carotenoids and proteins), which makes it difficult for the light beam to pass through the spectrophotometer.

CONCLUSIONS

The proximate analyses revealed that petals of pink and purple colour have more protein and raw fibre, likewise similar raw fat values between the analysed colours. Purple flowers showed the highest values of TC, TFI and VC, however, in the latter it was similar to cherry flowers. Chery flowers presented a higher concentration of TP and TA. This work demonstrated

that the colour of ligulate dahlia flowers show variability in their nutritional value, bioactive compounds and antioxidant capacity.

CONCLUSIONS

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