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An Investigation of Minerals, Vitamin C, and Antioxidant Capacity of Four *Bougainvillea* spp. Cultivars

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Abstract

The *Bougainvillea* spp. is an ornamental, medicinal, and edible flower whose edibility has been less addressed in previous studies. This research explored the edible application of four bougainvillea cultivars (*B. glabra, B. glabra* 'Snow White', *B. glabra* 'Scarlett O'Hara', and *B. glabra* 'Louis Wathen'). The bougainvillea were purchased at the full blooming stage from a commercial producer in Talesh County, Guilan province, and their petals were used to determine the contents of minerals, vitamin C, anthocyanin, and antioxidant activity. The results showed that *B. glabra* 'Scarlett O'Hara' had the highest P (32.10 mg/100 g FW), Ca (98 mg/100 g FW), Fe (3.79 mg/100 g FW), and anthocyanin content (31.14 mg/100 g FW). The highest K contents (181.81 and 181.52 mg/100 g FW) were obtained from *B. glabra* and *B. glabra* 'Scarlett O'Hara', and the highest Zn content (0.35 mg/100 g FW) was recorded by *B. glabra*. Also, *B. glabra* 'Snow White' was the weakest in Zn, Fe, P, and anthocyanin contents. *B. glabra* 'Snow White' and *B. glabra* 'Louis Wathen' were the best in vitamin C content, and *B. glabra* 'Snow White' and *B. glabra* were the best in antioxidant capacity. In general, it was found that all four cultivars had nutritional value. However, the best were *B. glabra* 'Scarlett O'Hara' due to its mineral content and *B. glabra* 'Snow White' due to its vitamin C content and antioxidant capacity.

Keywords: Anthocyanin, Antioxidants, Edible flower, Nutritional value, Ornamental plants.

INTRODUCTION

Flowers have traditionally been used to beautify and scent the environment and treat many diseases. Flower application in cuisine for improving the organoleptic properties and visual beauty of foods and beverages has increased in recent years. The flowers of medicinal and aromatic plants, fruit trees, and vegetables are the main categories of edible flowers, which are rich in nutrients, vitamins, and antioxidants and have numerous health benefits, such as anti-cancer, anti-diabetics, antibacterial, antiviral, antifungal, and anti-inflammatory activities (Pensamiento-Niño *et al*., 2024; Pires *et al*., 2023). Although edible flowers are abundant as natural sources of nutrients and biologically active compounds, naturally occurring flowers need to be tested for their chemical and biological features to avoid the likely risks, especially their toxicity. So far, 180 species, 100 genera, and 97 families of edible flowers have been detected (Saurabh and Barman, 2020; Pensamiento-Niño *et al*., 2024), consumed fresh as salad. In addition, edible flowers are used as a raw material in preparing foods, such as stews, cakes, beverages, seasonings, and desserts (Zhao *et al*., 2019).

An essential reason for using flowers in cuisine is the appealing color of their petals. In addition to beautification and attraction of pollinators, pigments endow the flowers with antioxidant properties. Therefore, pigments, especially anthocyanins, are a crucial category of chemicals in flowers that can increase their nutritional and medicinal value due to their vigorous antioxidant activity and other beneficial physic-chemical and biological properties (Benvenuti *et al*., 2016).

In addition to pigments, edible flowers contain water, fiber, proteins, fats, carbohydrates, vitamins, minerals, sugars, free amino acids, alkaloids, organic acids, phenols, and antioxidants in abundant amounts (Fenanndes *et al*., 2019; Barani *et al*., 2022). Although the nutritional value and many health benefits of edible flowers have been reported, they are generally consumed at very low levels due to the lack of their detection and the fear of their toxicity. So, applied research is required to increase public awareness and introduce new edible flowers.

A research study on the nutritional and medicinal value of 23 rose cultivars revealed that the flowers, especially red cultivars, were an excellent edible source of phenolic compounds, vitamin C, and anthocyanins (Kalisz *et al*., 2023). Jadhav *et al*. (2023) reported that many edible flowers are a good source of vitamin C. Nicknezhad *et al*. (2022) introduced the marigold, gladiolus, yucca, and chrysanthemums as a new source of minerals, vitamin C, and antioxidants. Bayanifar *et al*. (2024) investigated different chrysanthemum cultivars in terms of their vitamin C, minerals, and antioxidant compounds. They revealed that the studied cultivars could partially meet the body's mineral requirements. Pourzarnegar *et al*. (2023) found that rose cultivars, especially the cultivars in red, were acceptable edible sources of vitamin C and antioxidant compounds.

The *Bougainvillea* spp. from the family of Nyctaginaceae is native to hot and semi-hot areas of South America, Peru, Argentina, and Brazil. As an ornamental spiral, the bougainvillea has many fans. Furthermore, it has medicinal and edible applications. Over 105 biologically active compounds have been detected in different parts of the bougainvillea, used for disease treatment in traditional medicine (Saleem *et al*., 2021). There is no significant information about the nutritional value of bougainvillea. Given the need to find new edible flowers and detect their nutritional compounds in each region, this research investigated and compared four bougainvillea cultivars, including *B. glabra* with purple flowers (Kumar *et al*., 2017), *B. glabra* 'Snow White' with white bracts (Gupta *et al*., 2009), *B. glabra* 'Scarlett O'Hara' with magenta flowers (El-Sayed *et al*., 2020), and *B. glabra* 'Louis Wathen' with orange bracts (Thompson, 2011) as an edible source.

MATERIALS AND METHODS

An experiment based on a completely randomized design was conducted in the summer of 2023 to study four bougainvillea cultivars (*B. glabra*, *B. glabra* 'Snow White', *B. glabra* 'Scarlett O'Hara', and *B. glabra* 'Louis Wathen') (Fig. 1). The flowers were purchased at the fully blooming stage from a commercial producer in Talesh County, Guilan province, and were transferred to the study site (Islamic Azad University, Rasht), taking care of all handling precautions. Then, the healthy flowers that had no symptoms of diseases or mechanical damage were packed in lidded containers in 10-branch groups and were kept at 4°C during the experiment and trait assessment.

B. glabra | *B. glabra* 'Snow White' | *B. specabilis* 'Scarlett O'Hara' | *B. buttiana* 'Louis Wathen'

Fig. 1. The bougainvilleas flowers used in the research.

Assessment of traits

Minerals (N, P, Ca, K, Fe, and Zn)

To measure N content, 0.5 g of the fresh petal was mixed with acids (100 ml of sulfuric acid, 6 g of salicylic acid, and 18 ml of H_2O_2). After 24 hours, the samples were subjected to digestion using an electric heater. The resulting samples were used to measure the N content using the Kjeldahl method. Finally, the following equation was used to find the N percent:

$$N(\%) = 0.56 \times t \times (a - b) \times \frac{V}{W} \times \frac{100}{DM}$$

In which *t* is the concentration of the acid used for titration in mol/l, *a* is the amount of the acid used in the sample in ml, *b* is the amount of acid used for control in ml, *V* is the volume of the extract derived from the digestion in ml, *W* is the petal sample weight for digestion in g, and *DM* is the dry matter percent of the petal.

To measure the P, K, Ca, Fe, and Zn contents, some petals were converted into ash at 550°C. Then, 1 g of the petal ash was mixed with 1 ml of nitric acid in a 250-mL Erlenmeyer, and it was added with soda 10% and the MOROXAED mixture. Then, the Ca content was determined by titration using ethylenediaminetetraacetic acid (EDTA), the K content was determined by flame-photometry, and the P content was determined by adding the nitro-vanadate-molybdate reagent and using spectrophotometry. The Fe and Zn content of the samples was also evaluated using an atomic absorption device and drawing the standard graph (Rezaee *et al*., 2004).

Vitamin C content

To measure the vitamin C content, 2 g of the fresh petal was extracted using 5 ml of liquid nitrogen. Then, 15 ml of meta-phosphoric acid 3% was added to the extract and mixed with it. The filtered sample was adjusted to 10 ml by adding meta-phosphoric acid 3% and titrated with 2,6-dichlorophenolindophenol until a light pink color emerged. Finally, the vitamin C content of each sample was calculated using the following equation (Mazumdar and Majumder, 2003):

$$Vitamin\ C = \frac{e \times d \times b}{c \times a} \times 100$$

In which *a* is the sample weight, *b* is the amount of meta-phosphoric acid used for extraction, *c* is the solution taken for titration, *e* is the amount of color solution consumed for the sample, and *d* is the color factor. The following equation gives d:

$$d = \frac{0.5}{The\ amount\ of\ color\ solution\ used\ for\ titration\ of\ the\ standard\ sample}$$

Petal anthocyanin

To measure the anthocyanin content, the petal extract was derived using acidic methanol (pure methanol + hydrochloric acid). The resulting extract was filtered through Whatman filter paper and adjusted to 50 ml by adding distilled water. Then, the absorbance was read at 535 nm with a spectrophotometer (APEL, PD-103UV), and the petal's anthocyanin content was estimated by the following equation (Mazumdar and Majumder, 2003):

$$Anthocyanin\ content = \frac{e \times b \times c}{d \times a} \times 100$$

in which *e* is the sample weight, *b* is the sample volume for measurement, *c* is the total solution generated, *d* is the volume of the sample taken, and *a* is the reading by the spectrophotometer.

Antioxidant capacity

The antioxidant capacity of the petal samples was determined by Brand-Williams *et al.*'s (1995) method for which 1 g of fresh petal was extracted using 10 ml of pure methanol. The extracts were kept at room temperature for 2 hours and then filtered through Whatman filter paper. Then, 50 µl of the extract was mixed with 950 µl of 2,2-diphenyl-1-picrylhydrazyl (DPPH) and was kept in darkness at room temperature for some minutes. Then, the absorbance of the samples was read with a spectrophotometer (APEL PD-303UV) at 515 nm, and the following equation was used to determine the antioxidant capacity:

$$\%DPPHsc = (A_{cont} - A_{samp}) \times 100/A_{cont}$$

in which $DPPH_{sc}\%$ represents the percent inhibition of free radicals, A_{cont} represents the absorbance of the sample and DPPH, and A_{samp} represents the absorbance of DPPH.

Statistical analysis

The collected data were analyzed in the SPSS19 software suite, in which they were subjected to analysis of variance, as well as the comparison of means by the LSD test at the P <0.01 level.

RESULTS

Minerals

Based on the results of ANOVA, the cultivars significantly (P < 0.01) differed in minerals, including N, P, Ca, K, Fe, and Zn (Table 1). The comparison of means showed that *B. glabra* 'Scarlett O'Hara' had the highest levels of N, P, Ca, and Fe. The highest Zn content was recorded by *B. glabra*, whereas the two cultivars *B. glabra* and *B. glabra* 'Scarlett O'Hara' had the lowest one. The lowest N, P, Fe, and Zn contents belonged to *B. glabra* 'Snow White'. On the other hand, *B. glabra* 'Louis Wathen' recorded the lowest Ca and K contents (Table 2).

Table 1. Analysis of variance of the effect of different cultivar on petals minerals in *Bougainvillea* spp.

S.o.V	df	N	P	Ca	K	Fe	Zn
Treatment	3	0.00066**	15.79**	1796.1**	455.6**	0.315**	0.0154**
Error	16	0.0000125	0.202	0.684	2.29	0.0179	0.000947
CV (%)		1.29	1.476	1.024	0.86	3.83	11.17

**: Significant at P < 0.01 based on the LSD test.

Table 2. Mean comparison of the effect of different cultivar on petals minerals in *Bougainvillea* spp.

	N	P	Ca	K	Fe	Zn
Cultivars	**%**	**(mg/100 g F.W.)**				
B. glabra	0.278[b]	30.74[b]	96.39[b]	181.81[a]	3.53[b]	0.35[a]
B. buttiana "Louis Wathen"	0.267[c]	31.06[b]	63.80[d]	161.54[c]	3.46[b]	0.24[bc]
B. specabilis "Scarlett OHara"	0.286[a]	32.10[a]	98.00[a]	181.52[a]	3.79[a]	0.28[b]
B. glabra "Snow White"	0.260[d]	27.94[c]	64.99[c]	172.88[b]	3.18[c]	0.22[c]

*In each column, means with similar letter(s) are not significantly different (P < 0.05) using the LSD test.

Minerals are necessary for the health and natural functioning of the human body, so a proper diet must be rich in minerals (Huang *et al*., 2020). Flowers are new sources of minerals in the human food basket (Benvenuti *et al*., 2016). As was already mentioned, bougainvilleas with red and purple petals are richer in minerals than those with white and orange petals. Some researchers have stated that although the daily consumption of edible flowers is trivial, their continuous consumption can supply a part of the human need for minerals and biologically active compounds (Araújo *et al*., 2019).

Rop *et al*. (2012) argue that the mineral contents of edible flowers have a curative effect and are one of the most necessary aspects of edible flower consumption for human nutrition. Benvenuti *et al*. (2016) reported that flowers, as natural sources of minerals, can partially meet the micro-element and macro-element requirements of the human body. We recorded the Ca content of different bougainvillea cultivars at 63.80-98 mg/100 g FW, which was greater than that of white gladiolus (9.11 mg/100 g FW) and purple chrysanthemum (47.25 mg/100 g FW) (Nicknezhad *et al*., 2022). In addition, all four bougainvillea cultivars recorded higher Fe content than marigold, gladiolus, yucca, chrysanthemum, and hollyhock (0.36-2.54 mg/100 g FW) (Nicknezhad *et al*., 2022). Regarding the Zn level, *B. glabra* 'Louis Wathen' (0.24 mg/100 g FW) and *B. glabra* 'Snow White' (0.22 mg/100 g FW) performed weaker than yucca (0.27 mg/100 g FW) and hollyhock (0.25 mg/100 g FW), but *B. glabra* and *B. glabra* 'Scarlett O'Hara' outperformed yucca, chrysanthemum, gladiolus, marigold, and hollyhock as reported by Nicknezhad *et al*. (2022). The comparison of the bougainvilleas with 20 chrysanthemum genotypes studied by Bayanifar *et al*. (2024) in terms of minerals revealed that all four studied paper flower cultivars were superior to the 20 chrysanthemum genotypes in terms of Fe, Zn, and Ca. Also, the studied bougainvilleas had higher P, Ca, and Fe contents but lower Zn and K contents than begonia, roses, daylilies, and pot marigold (Mlcek *et al*., 2021). Based on the results, it can be said that bougainvilleas, especially the cultivars with red and purple flowers, which had the highest mineral contents in this research, can be included in the food regime as a new source of minerals.

Anthocyanins

The four bougainvillea cultivars differed in anthocyanin content significantly ($P < 0.01$; Table 3). The anthocyanin content ranged from 12.64 to 31.14 mg/100 g FW among them. The lowest was for *B. glabra* 'Snow White', which had white petals, and the highest was for *B. glabra* 'Scarlett O'Hara', which had red petals (Fig. 2).

Table 3. Analysis of variance of the effect of cultivar on anthocyanin, vitamin C and antioxidant capacity in *Bougainvillea* spp.

S.o.V	df	Anthocyanin	Vitamin C	Antioxidant capacity
Treatment	3	320**	1.49**	357**
Error	16	0.145	0.257	0.249
CV (%)		1.62	3.89	0.98

**: Significant at $P < 0.01$ based on the LSD test.

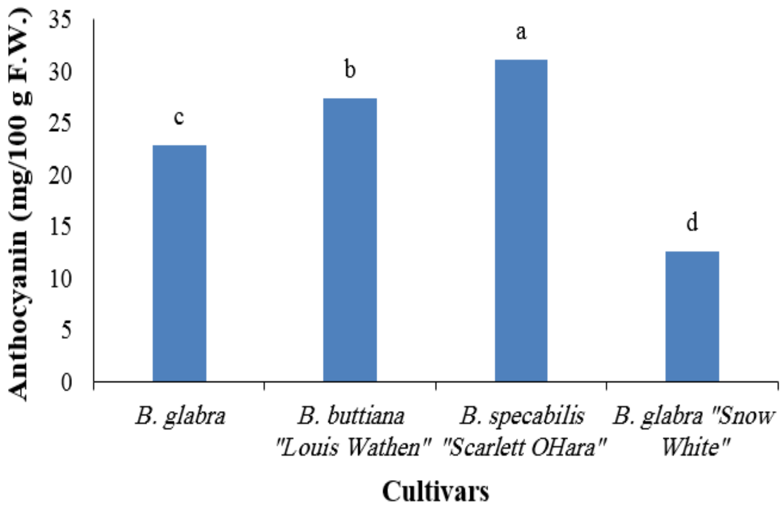

Fig. 2. Effect of cultivar on petals anthocyanin.

Anthocyanins are strong antioxidants that are mainly responsible for a wide range of red, purple, and blue colors in flowers, fruits, and vegetables. These naturally occurring pigments are of high significance in the food industry due to their attractive colors and in the medical industry due to their beneficial curative and antioxidant activities. Humans receive considerable amounts of anthocyanins through the food regime and the consumption of natural resources (Sadighara *et al*., 2012). The mean anthocyanin intake from natural resources has been reported at 180-215 mg/d among US citizens (Lee *et al*., 2011).

We found that the anthocyanin content was higher in the cultivars with red and orange petals than in those with purple and white petals. A study showed that edible violet flowers with dark (red and purple) colors contained more anthocyanin than those with light-colored petals (Ikeura *et al*., 2023). Islam (2016) reported that the anthocyanin content was higher in red gladioluses than in purple, pink, yellow, and white ones. In Park *et al.*'s (2015) study, the chrysanthemum cultivars with red and purple petals had a higher anthocyanin content than those with orange, green, and white flowers. Similar results were recorded by Bayanifar *et al.* (2024). They revealed that the anthocyanin content was lower in chrysanthemum genotypes with white petals than in those with dark petals, which agrees with our finding about the lower anthocyanin content of the bougainvilleas with white flowers. Since anthocyanins are responsible for generating red, purple, and blue color in plant parts, so there may be a correlation between flower color and anthocyanin content. However, environmental factors, genotype, and growth stage influence anthocyanin content, too (Espejel *et al*., 2019).

Vitamin C

The vitamin C content of the studied cultivars differed significantly at the $P < 0.01$ level (Table 3). It was in the range of 12.24-13.45 mg/100 g FW. *B. glabra* 'Louis Wathen' had the highest vitamin C content, but it did not differ from *B. glabra* 'Snow White' significantly. The lowest was recorded by *B. glabra* 'Scarlett O'Hara' (Fig. 3).

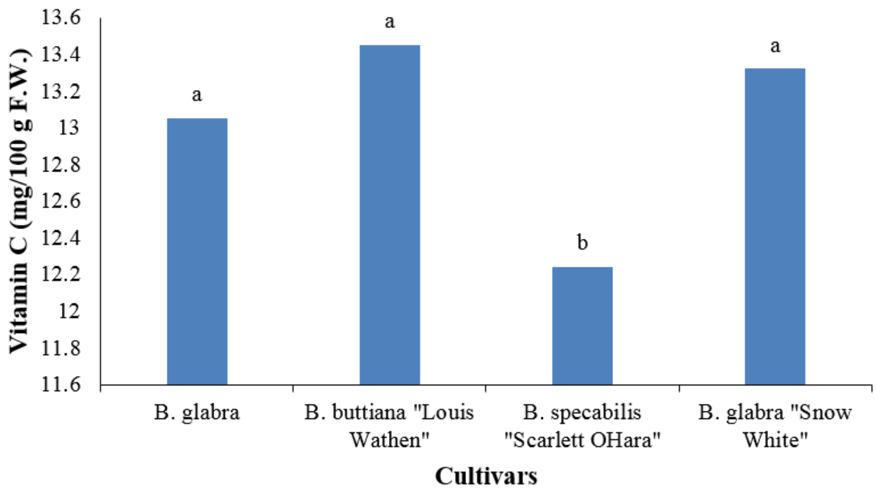

Fig. 3. Effect of cultivar on vitamin C.

Vitamins, including vitamin C, are necessary for body growth and health. The human body does not synthesize vitamin C, so our vitamin C requirement, which amounts to 95-100 mg/day (Bayanifer *et al*., 2024), must be supplied by the food regime. Oranges and kiwifruits, the most famous sources of vitamin C, contain 35 and 93 mg of vitamin C/100 g FW (Demasi *et al*., 2021; Cruz-Rus *et al*., 2012). Although, the bougainvillea cultivars had lower vitamin C content than oranges and kiwifruits, they are a moderate source of vitamin C compared to many edible flowers, as their vitamin C content has been recorded from 2.6 to 44.9 mg/kg FW (Demasi *et al*., 2021). In a study by Bayanifar *et al*. (2024), the vitamin C content of 20 chrysanthemum cultivars was recorded from 11.71 to 13.58 mg/kg FW. In contrast, Nicknezhad *et al*. (2022) found that the vitamin C content varied from 8.16 mg/100 g for chrysanthemum to 30.6 mg/100 g FW for orange marigolds. The comparison of their results with ours shows that the studied bougainvillea cultivars had higher vitamin C content than chrysanthemum flowers and higher vitamin C content than marigold flowers. In the present work, the orange bougainvilleas had the highest, and the white ones had the second-highest vitamin C content, which agrees with the results reported by Nicknezhad *et al*. (2022), according to which orange marigold flowers had the highest and white yucca flowers had the second-highest vitamin C content.

Antioxidant capacity

As is evident in Table 3, the bougainvillea cultivars differed in antioxidant capacity significantly ($P<0.01$). The antioxidant capacity of the studied cultivars varied from 43.7 to 61.06%. The highest was recorded by *B. glabra* 'Snow White' and *B. glabra*, whereas *B. glabra* 'Louis Wathen' and *B. glabra* 'Scarlett O'Hara' were similarly weaker than the other two (Fig. 4).

Many researchers have stated that there is a relationship between antioxidant capacity and flower color. Flowers in dark colors (red and blue) have higher antioxidant capacity than flowers in light colors (Sadighara *et al*., 2012; Benvenuti *et al*., 2016; Ikeura *et al*., 2023). But, *B. glabra* 'Snow White', which had white petals, recorded the highest antioxidant capacity. Similarly, Chen and Wei (2017) argue that the antioxidant capacity of flowers may be influenced by their chemical compounds. Carotenoids and flavonoids may sometimes be more influential on antioxidant capacity than anthocyanins.

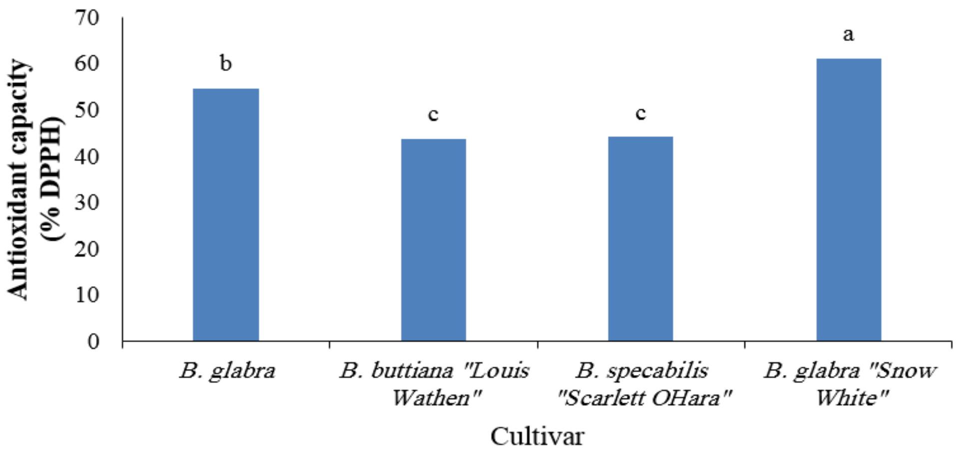

Fig. 4. Effect of cultivar on antioxidant capacity.

Nicknezhad *et al*. (2022) recorded the antioxidant capacity of yucca, marigold, and gladiolus at 47.76, 87.89, and 61.91% HPPH inhibition. So, *B. glabra* 'Snow White' had as much antioxidant capacity as white gladiolus but a higher antioxidant capacity than yucca. However, all four bougainvillea cultivars had lower antioxidant capacities than the marigold.

Anthocyanins and vitamin C are essential antioxidants. According to the correlation test

(Table 4), the anthocyanin content had a negative and significant relationship with the vitamin C content and antioxidant capacity. In the present work, *B. glabra* 'Snow White' had the best, and *B. glabra* had the second-best antioxidant capacity. These two cultivars performed weakly in terms of anthocyanin, but they had acceptable vitamin C content. The correlation between vitamin C and antioxidant capacity was insignificant (Table 4).

Table 4. Correlation between traits.

	An	Vit C	AC	N	P	K	Fe	Zn	Ca
An	1								
Vit C	-0.411*	1							
AC	-0.944**	0.272	1						
N	0.736**	-0.644**	-0.511**	1					
P	0.948**	-0.578**	-0.849**	0.774**	1				
K	0.065	-0.483*	0.229	0.639**	0.173	1			
Fe	0.793**	-0.503**	-0.653**	0.805**	0.822**	0.378	1		
Zn	0.305	-0.012	-0.051	0.602**	0.344	0.567**	0.492**	1	
Ca	0.498**	-0.579**	-0.199	0.886**	0.593**	0.877**	0.673**	0.731**	1

*and**: Significant at $P < 0.05$ and $P < 0.01$ based on the LSD test, respectively. An: Anthocyanin; Vit C: Vitamin C; AC: Antioxidant capacity.

CONCLUSIONS

Although, different bougainvillea cultivars differ in edibility features significantly, they were all good sources of minerals, vitamin C, and antioxidant capacity. The conclusion is that *B. glabra* 'Scarlett O'Hara' can be recommended as a source of minerals, and *B. glabra* can be recommended as a source of vitamin C and antioxidant capacity.

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