Seasonal variation in phytochemicals and nutraceutical potential of *Begonia nelumbiifolia* consumed in Puebla, México

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Abstract *Begonia nelumbiifolia* is a traditional edible plant consumed and commercialized in the northern highlands of Puebla, México. The present study reports the seasonal variation in proximate analysis as well as organic acids, carotenoids and flavonoids content in both leaves and stalks of this plant. The stalks contained low concentrations of protein ($\sim 3\%$), fiber ($\sim 1.5\%$) and nitrogen free extract ($\sim 0.26\%$) during the time of study. Both organs showed contents of oxalic acid ($91\text{--}103\text{ mg } 100\text{ g}^{-1}\text{ FW}$), ascorbic acid ($50\text{--}65\text{ mg } 100\text{ g}^{-1}\text{ FW}$), lutein ($1\text{--}2.5\text{ mg } 100\text{ g}^{-1}\text{ FW}$), β -carotene ($1\text{--}3\text{ mg } 100\text{ g}^{-1}\text{ FW}$), quercetin ($1.3\text{--}2.7\text{ mg } 100\text{ g}^{-1}\text{ DW}$) and rutin ($0.5\text{--}1.7\text{ mg } 100\text{ g}^{-1}\text{ DW}$). Antioxidant activity against DPPH was observed by the stalk extracts from 30% methanol (IC_{50} , $0.21\text{--}0.37\text{ mg L}^{-1}$), pure methanol (IC_{50} , $0.14\text{--}0.21\text{ mg L}^{-1}$) and hexane: acetone (IC_{50} , $0.77\text{--}1.21\text{ mg L}^{-1}$). In vitro anti-HMG-CoA reductase (IC_{50} , $0.07\text{--}0.36\text{ mg L}^{-1}$) and anti-alpha-glucosidase (IC_{50} ,

$0.28\text{--}0.43\text{ mg L}^{-1}$) activities were observed in extracts from the edible stalks from pure methanol and 30% methanol. The leaf extracts from 30% methanol inhibited the growth of *Pseudomonas syringae* pv. tabaci TBR2004 (MIC, $254\text{ }\mu\text{g mL}^{-1}$), *P. syringae* pv. tomato DC3000 (MIC, $423\text{ }\mu\text{g mL}^{-1}$), *P. syringae* pv. glycinea (MIC, $605\text{ }\mu\text{g mL}^{-1}$) and *Clavibacter michiganensis* AB299158 (MIC, $162\text{ }\mu\text{g mL}^{-1}$). *B. nelumbiifolia* contained valuable phytochemicals associated to nutraceutical and biological properties. However, the consumption of the fresh stalks must be carefully considered because of the high oxalate content.

Keywords *Begonia nelumbiifolia* · Nutraceuticals · Antioxidant · Anti-HMG-CoA · Anti-alpha-glucosidase

Introduction

Approximately 30 *Begonia* species have been taxonomically described between the state of Veracruz and the northern highlands of Puebla, Mexico, thus far (Basurto-Peña et al. 2003; McMillan et al. 2006). Some species commonly grow in coffee plantations in those geographical areas, but only a few are considered edible and/or medicinal (Martínez et al. 2007; Alfaro 1984). *Begonia nelumbiifolia* and *Begonia heracleifolia* are two of these plants frequently consumed in those regions; however, their nutraceutical potential and biological properties are still unknown (Laferriere et al. 1991). Native people of the northern highlands of Puebla usually use the boiled stems of *Begonia nelumbiifolia*, locally known as “xocoyoles” or “quelites”, to prepare traditional dishes mixed with beans, scrambled eggs and soups (Basurto-Peña et al. 2003; Mapes and Basurto 2016). Because of the perennial condition of this plant, fresh stalks are commercialized in local

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marketplaces throughout the year. The “xocoyoles” have a hard, acid, fruity taste like lemon so it is used as a condiment. On the other hand, the plant leaves are ground to prepare cataplasms for treating soft tissue injuries such as contusions, abrasions, strains and sprains due to their chilling effect on inflamed parts. Considering the use of this traditional crop by the native people of México, we did a proximate analysis looking for main chemical components and some in vitro biological activities of this plant. The seasonal variation of these components and activities between 2014 and 2015 was also evaluated.

Materials and methods

Plant material

Wild *Begonia nelumbiifolia* Schltdl. et Cham., was collected in Yaonáhuac, Puebla, México (19°56'55"N 97°26'26"W; 1997 MASL) during spring (March 2014), summer (July 2014), fall (October 2014) and winter (January 2015). The taxonomical identity of the plant was certified at the Herbarium of Facultad de Ciencias, UNAM, México (FCME-UNAM), where a reference voucher 150,043 was deposited. Five independent seasonal samples ($n = 5$) were collected and analyzed in each season to determine their proximate and chemical composition as well as the biological activities.

Chemicals

All solvents were analytical or HPLC grade from J.T. Baker. Analytical standards of organic acids, carotenoids, flavonoids, DPPH radical and enzymes were from Sigma Aldrich Co. St. Louis Mo.

Proximate analyses

The basic proximate analyses of *B. nelumbiifolia* were estimated in accordance with the AOAC standard protocols (2005). Method 926.08 was used for moisture and dried matter calculation. Method 920.23 was used for the crude protein content calculation. Methods 923.03 and 962.09 were used for ash and crude fiber determinations, respectively. Finally, method 920.39 was used for the crude fat content determination. All assays were performed in quintuplicate and the results were expressed in percentage $\pm$ standard deviation.

Extraction and determination of organic acids

100 g of fresh stalks or leaves were ground in a sterile mortar and extracted with 100 mL of cold 30% methanol in

darkness with vigorous shaking at 4 °C for 12 h. Extracts were filtered with Whatman No. 1 paper and concentrated in a Büchi R-200 rotary evaporator to approximately 60 mL. Chlorophylls and phenols were removed twice with an equivalent volume of an aqueous solution of 4% lead acetate and 1% polyvinylpyrrolidone (MW 40,000). Then the extract was centrifuged at 7300 RCF for 10 min at 4 °C, and the supernatant was completely dried in a centrifuge concentrator (RVC 2–25 CD) for 3 h. The resulting powder was resuspended in 500 μ L of methanol HPLC grade and filtered through Titan[®] filters containing a nylon membrane of 0.45 μ m pore diameter. Filtrates were stored in 1.5 mL amber glass vials at –20 °C until use. For RP-HPLC analysis, 20 μ L of extracts were injected by triplicate in a Hewlett Packard 1050 system coupled to a HP G1306A diode array detector, equipped with a Varian Chromosphere C₁₈ column (5 μ m, 250 $\times$ 4.6 mm). The conditions including the isocratic method, mobile phase, temperature of the column, flow rate and absorbance were as described (Nour et al. 2010). The abundance of oxalic, malic, lactic, ascorbic and citric acids was calculated from respective calibration curves with authentic standards.

Extraction and determination of carotenoids

100 g of fresh stalks or leaves were ground in sterile mortar and extracted with 200 mL of cold hexane: acetone (1:1 v/v) added with 0.5% butylated hydroxytoluene (BHT), with vigorous shaking at 4 °C for 12 h in darkness. Extracts were filtered and reduced to 50 mL for further saponification according to Pintea et al. (2003). Then, 50 mL of distilled water was added along with 50 mL of 10% NaCl. The two layers of the mixture were separated in a sedimentation funnel after vigorously shaken. The upper layer containing carotenoids was collected, reduced to dryness under N₂ stream, suspended in 500 μ L acetone and filtered as previously described for organic acids. Samples were stored at –20 °C in amber glass vials until use. HPLC analysis was carried out with 20 μ L injected by triplicate in a C₁₈ column Nucleosil ODS (5 μ m, 250 $\times$ 4.6) according to Pintea et al. (2003). The abundance of lutein, cryptoxanthin, α -carotene and β -carotene was estimated from respective calibration curves with authentic standards.

Extraction and determination of flavonoids

100 g of lyophilized leaves or stalks were powdered in sterile mortar and immediately extracted twice with 300 mL 100% methanol. The extracts were filtered and reduced to dryness until a green–yellow gum. For analysis, gums were separated using the same conditions for column chromatography and HPLC (20 μ L) described by De Oliveira et al. (2001). For HPLC analyses, 20 μ L were

injected in the same LC apparatus and the column used was the Varian Chromosphere C₁₈ (5 μ m, 250 $\times$ 4.6 mm). The abundance of rutin, quercetin and kaempferol was estimated by respective calibration curves using authentic standards.

In vitro antioxidant activity of edible stalks

The antioxidant activity against DPPH was assayed with all previous extracts in accordance with Villa-Ruano et al. (2015). The extracts were concentrated to dryness in a RVC 2–25 CD centrifuge concentrator and suspended in the same solvents systems to obtain concentrated stock solutions (100 mg mL⁻¹). Assays were done in quintuplicate with the five different seasonal samples and a dose–response test of 0.01–2.0 mg mL⁻¹. The seasonal IC₅₀ was obtained by linear regression. Butylated hydroxytoluene (BHT) was used as a reference standard.

In vitro anti-HMG-CoA reductase and anti-alpha-glucosidase activities of edible stalks

Same extracts prepared for antioxidant activity were used for both enzymatic assays. The putative HMG-CoA reductase inhibition was performed in accordance with Gholamhoseinian et al. (2010), but using Tris–EDTA (0.1 M, pH 7.4) as reaction buffer and Pravastatin (Merck®) as inhibition control. The anti-alpha-glucosidase activity was carried out in accordance with Villa-Ruano et al. (2015), using acarbose (Bayer®) as inhibition control. All assays were done in quintuplicate and a dose–response test of 0.01–1 mg mL⁻¹. The seasonal IC₅₀ was obtained by linear regression.

Antibacterial activity of extracts from the leaves of *B. nelumbiifolia*

The broth microdilution method (Villa-Ruano et al. 2015) was used with the three leaf extracts previously described. The assessment was carried out in quintuplicate using five different seasonal samples on *Pseudomonas syringae* pv. tabaci TBR2004, *P. syringae* pv. tomato DC3000, *P. syringae* pv. phaseolicola NPS3121, *P. syringae* pv. glycinea, *P. stewartii*, *Erwinia carotovora* subsp. *carotovora* and *Clavibacter michiganensis* AB299158. A dose–response curve of 0.01–2.0 mg mL⁻¹ was made for each species to obtain the minimum inhibitory concentration (MIC). Turbidity changes were measured at 600 nm in a Beckman DU 7400 spectrophotometer. Rifampicin and Agrigent Plus® were assessed as antibiotics of reference.

Statistical analyses

Results from the proximal analyses, phytochemicals levels and the antioxidant activities were analyzed and validated by ANOVAs coupled to a Tukey test ($p < 0.01$) using the software GraphPad 6.05.

Results and discussion

Basic nutritional content

The fresh stalks of *B. nelumbiifolia* contained a high percentage of water (Table 1) but a low concentration of protein (~3%) and fiber (~1.5%), comparable to other traditional plant foods such as *Ocimum gratissimum* and the succulent *Opuntia ficus-indica* (Idris et al. 2011; Chiteva and Wairagu 2013). However, in contrast to other plants, the nitrogen free extract had a negligible amount (~0.2%) of assimilable carbohydrates. This property could be of benefit to diabetic people who consume these plants. The seasonal variation of these components was quite stable during the year except for summer time (Table 1) where lipids, protein and fiber levels were higher. Considering that original plants were subjected to different biotic and abiotic factors, the stability of the levels of these components was remarkable. The summer differences may be due to the high rain season which could allow for faster and robust growing plants.

Organic acids content

Oxalic acid was the most abundant organic acid in both leaves (103.84 mg 100 g⁻¹ FW) and stalks (92.12 mg 100 g⁻¹ FW) in all analyzed seasons followed by ascorbic (50.63–61.65 mg 100 g⁻¹ FW), malic (10.87–16.15 mg 100 g⁻¹ FW) and then traces of citric (3.14–4.48–100 g⁻¹ FW) and lactic acids (1.39–2.44 mg 100 g⁻¹ FW) (Fig. 1a–c). These results reveal that *B. nelumbiifolia* biosynthesizes and accumulates more oxalic acid than *Begonia evansiana* (Yang and Loewus 1975). Unexpectedly, the average levels of oxalic acid were slightly higher in leaves (Online Resource Table 1) than in the edible stalks (Online Resource Table 2). A slight accumulation of organic acids was observed in both organs in fall and winter, in comparison to spring and summer which are periods characterized by abundant sunlight (Online Resource Tables 1, 2). This biosynthetic pattern has been observed in other *Begonia* species, where variation in organic acids accumulation was related to light intensity (Yang and Loewus 1975). Oxalic acid has been considered an anti-nutritional factor by the role it plays in the formation of calculi in the kidneys and urinary system (Barot

Table 1 Composition of the stalks from *Begonia nelumbiifolia* collected in different seasons

Season	Moisture content (%)	Dry matter content (%)	Crude fat content (%)	Crude protein (%)	Crude fibre content (%)	Ash content (%)	N ₂ free extract (%)
Spring	94.46 ± 0.02 ^a	5.54 ± 0.01 ^b	0.51 ± 0.01 ^c	2.93 ± 0.0 ^a	1.26 ± 0.02 ^a	0.55 ± 0.07 ^b	0.29 ± 0.01 ^b
Summer	92.31 ± 0.06 ^a	7.69 ± 0.04 ^a	0.92 ± 0.05 ^a	3.62 ± 0.0 ^a	1.93 ± 0.03 ^a	0.81 ± 0.02 ^a	0.41 ± 0.05 ^a
Fall	93.93 ± 0.09 ^a	6.07 ± 0.02 ^a	0.71 ± 0.08 ^b	3.05 ± 0.0 ^a	1.45 ± 0.03 ^a	0.64 ± 0.01 ^b	0.22 ± 0.04 ^b
Winter	94.23 ± 0.04 ^a	5.31 ± 0.07 ^b	0.53 ± 0.03 ^c	3.22 ± 0.0 ^a	1.32 ± 0.08 ^a	0.59 ± 0.02 ^b	0.11 ± 0.02 ^c
Mean	93.73 ± 0.97	6.15 ± 1.07	0.67 ± 0.19	3.21 ± 0.3	1.49 ± 0.30	0.65 ± 0.11	0.26 ± 0.13

The results are expressed in % plus or minus standard deviation

Different letters indicate means (n = 5) with significant differences by ANOVA-Tukey test ($p < 0.01$)

Fig. 1 HPLC identification of organic acids from *Begonia nelumbiifolia*. *a* Pure standards of oxalic (1), malic (2), lactic (3), ascorbic (4) and citric (5) acids. *b* Organic acids detected in the leaves. *c* Organic acids detected in the stalks

et al. 2015). The traditional preparation of “xocoyoles” or “quelites” in Mexico may avoid these damaging effects, but the fresh consumption of these plants should be avoided (Noonan and Savage 1999). One of the best procedures to reduce the soluble oxalate from edible plants was by cooking these with low fat milk and neutral pH (Simpson et al. 2009). Malic acid was more abundant in stalks (16.15 mg 100 g⁻¹ FW) than in the plant leaves (10.87 mg 100 g⁻¹ FW).

On average, the ascorbic acid was higher in the stalks than in the leaves (Online Resource Tables 1, 2), but endogenous variation ($p < 0.01$) was lower in the stalks being constant throughout the year (Online Resource Tables 1, 2). This fact could be related to the continuous biosynthesis of vitamin C in phloem tissue, which is abundant in the stalks of other edible plants including *Apium graveolens* (Hancock et al. 2003). In accordance with other studies, oxalic and ascorbic acids were biosynthetically related by the degradation of ascorbic acid into

oxalate, threonate and oxalyl threonate in oxalate-accumulating plants including *Begonia evansiana* (Yang and Loewus 1975; Truffault et al. 2016). Considering that other edible stalks such as celery, broccoli and the tuberous roots of sweet potato contain similar or even lower levels of vitamin C (Ishida et al. 2000; Hancock et al. 2003; Koh et al. 2009), the levels of ascorbic acid in the fresh edible stalks of *Begonia nelumbiifolia* suggest its use as a source of this micronutrient.

Carotenoid content

The main carotenoids identified in both organs were β -carotene (1.71–2.30 mg 100 g⁻¹ FW), lutein (1.35–1.97 mg 100 g⁻¹ FW) and β -cryptoxanthin (0.64–0.69 mg 100 g⁻¹ FW) (Fig. 2a–c). However, traces of α -carotene and other unidentified compounds were also detected (<0.1 mg 100 g⁻¹ FW) (Fig. 2b, c). The carotenoids content was higher in leaves (~6 mg 100 g⁻¹)

Fig. 2 HPLC identification of carotenoids from *Begonia nelumbiifolia*. *a* Pure standards of lutein (1), β -cryptoxanthin (2) α -carotene (3) and β -carotene (4). *b* Carotenoids detected in the leaves. *c* Carotenoids detected in the stalks

than in the stalks ($\sim 4 \text{ mg } 100 \text{ g}^{-1}$) during the studied seasons (Online Resource Tables 3, 4). The levels of β -carotene, cryptoxanthin and lutein were more abundant in spring and summer, characterized by more sunlight incidence than other seasons (Online Resource Tables 3, 4). The endogenous levels of carotenoids in the stalks of *B. nelumbiifolia* are like those reported for some Mediterranean edible plants such as purslane (*Portulaca oleraceae*), draba (*Cardaria draba*) and slender sow thistle (*Sonchus tenerrimus*) (Cámara et al. 2016). Remarkably, the levels of lutein and β -carotene were comparable with that of spinach (*Spinacia oleracea*) and beet (*Beta vulgaris*) (Cámara et al. 2016). Per these results, carotenoids from *B. nelumbiifolia* contribute to the intake recommendations of the World Health Organization (Cámara et al. 2016).

Flavonoid content

Quercetin (1.55–2.44 mg 100 g^{-1} DW), its glycoside rutin (0.72–1.38 mg 100 g^{-1} DW) and kaempferol (0.26–0.68 mg 100 g^{-1} DW) were the most abundant flavonoids detected in both the leaves and stalks of the plant (Fig. 3a–c). However, the levels of those flavonoids were lower in the stalks than in the leaves in all seasons (Online Resource Tables 5, 6). Despite the fact that the levels of flavonoids were considered lower in comparison to broccoli and cauliflower (Miean and Mohamed 2001), the abundance of quercetin was similar to the white radish (*Raphanus sativus*) and kailan (*Brassica alboglabra*) (Miean and Mohamed 2001). Flavonoids in edible green stems are limited; however, asparagus (*Asparagus officinalis*) and *B. nelumbiifolia* could be considered as good alternatives due to their rutin, quercetin and kaempferol content (Shahidi et al. 2011).

Antioxidant activity

Antioxidant assay revealed that all the crude preparations from edible stalks of *B. nelumbiifolia* stabilized the DPPH radical. Methanol extracts were the most effective inhibiting this radical, followed by 30% MeOH and hexane: acetone extracts (Table 2). Considering the MeOH dielectric constant, extracts containing this solvent probably had a significant number of flavonoids, ascorbic acid and carotenoids (Shalmashi and Eliassi 2008). In contrast, high levels of ascorbic acid in 30% MeOH extracts could reasonably clarify the antioxidant activity on DPPH. On the other hand, the low levels of carotenoids in hexane: acetone extracts could be associated to the relatively high IC_{50} values to DPPH. It has been reported that methanol and acetone extracts of asparagus and broccoli had significantly greater antioxidant activity than their water extracts (Sun et al. 2007). Similarly, this work demonstrates the

Fig. 3 HPLC identification of flavonoids from *Begonia nelumbiifolia*. a Pure standards of rutin (1), quercetin (2) and kaempferol (3). b Flavonoids detected in the leaves. c Flavonoids detected in the stalks

antioxidant effectiveness of pure methanol and methanol-aqueous preparations of *B. nelumbiifolia* but contrarily, a less effect of hexane: acetone extracts.

Inhibition of HMG-CoA reductase and alpha-glucosidase enzyme activities

The HMG-CoA reductase was inhibited by both 30% MeOH and pure methanol extracts (Table 3). Aqueous-methanol extracts containing mainly organic acids were slightly more effective than those with just methanol. Oxalic acid was a main component of the 30% MeOH extracts and its inhibitory effect on HMG-CoA from rat hepatocytes was proposed in 1983 (Stacpoole et al. 1983). However, no more information on this subject has been published since then. The IC_{50} value was more effective in fall and winter when oxalic acid was also more abundant than in other seasons (Online Resource Table 2). Pure MeOH extracts showed similar behavior in comparison to 30% MeOH extracts, but the most effective IC_{50} values were observed in summer and fall (Table 3). Pure methanol extracts contained the flavonoids quercetin, rutin and kaempferol; and rutin has recently been reported as the strongest inhibitor of HMG-CoA reductase among other plant-derived compounds (Suganya et al. 2017). These flavonoids among other phenolics have been used in the treatment of cardiovascular diseases due to their inhibition properties on HMG-CoA reductase (Ademosun et al. 2015). Like HMG-CoA reductase, the alpha-glucosidase enzyme activity was also inhibited by the two alcoholic extracts (Table 3). IC_{50} values of 30% methanol extracts from fall and winter were more effective. The malic acid concentration was slightly more abundant in these extracts than those from other seasons (Table 3). According to the

Table 2 Antioxidant activity against DPPH radical of different plants extracts from the edible stalks of *Begonia nelumbiifolia* in the four seasons of the year

Extract	Spring	Summer	Fall	Winter	Mean	BHT
30% MeOH	0.32 ± 0.06 ^b	0.37 ± 0.03 ^b	0.25 ± 0.18 ^b	0.21 ± 0.05 ^b	0.29 ± 0.07	0.043 ± 0.001
Hexane:acetone (1:1 v/v)	0.83 ± 0.04 ^a	0.77 ± 0.08 ^a	1.21 ± 0.03 ^a	0.92 ± 0.06 ^a	0.91 ± 0.20	
Methanol	0.21 ± 0.01 ^b	0.23 ± 0.11 ^b	0.14 ± 0.06 ^b	0.25 ± 0.03 ^b	0.21 ± 0.04	

IC₅₀ values are expressed in mg mL⁻¹ plus or minus standard deviation

Different letters indicate means (n = 5) with significant differences by ANOVA-Tukey test (*p* < 0.01)

Table 3 Effects of three different extracts from the stalks of *Begonia nelumbiifolia* on HMG-CoA and alpha-glucosidase enzymes

Extract	Spring	Summer	Fall	Winter	Mean	Standards
30% MeOH	0.22 ± 0.05 [£] / 0.33 ± 0.02 [€]	0.15 ± 0.01 [£] / 0.39 ± 0.04 [€]	0.09 ± 0.08 [£] / 0.32 ± 0.05 [€]	0.07 ± 0.01 [£] / 0.28 ± 0.05 [€]	0.13 ± 0.06 [£] / 0.33 ± 0.045 [€]	0.007 ± 0.0001 ^p / 0.017 ± 0.001 ^c
Hexane:acetone (1:1 v/v)	–	–	–	–	–	
Methanol	0.36 ± 0.04 [£] / 0.43 ± 0.06 [€]	0.24 ± 0.03 [£] / 0.32 ± 0.01 [€]	0.18 ± 0.02 [£] / 0.38 ± 0.07 [€]	0.29 ± 0.03 [£] / 0.40 ± 0.06 [€]	0.27 ± 0.07 [£] / 0.38 ± 0.04 [€]	

IC₅₀ values are expressed in mg mL⁻¹ plus or minus standard deviation (n = 5)

£ Values for HMG-CoA reductase; € values for alpha glucosidase. Standards: pravastatin (p), acarbose (c). A dash (–) indicates absence of activity on assayed concentrations

kinetic and in silico studies, malic acid is a potent inhibitor of alpha-glucosidase (Gou et al. 2015). On the other hand, pure methanol extracts from summer and fall exhibited more effectiveness inhibiting alpha glucosidase. Quercetin and rutin included in the pure methanol extracts have shown relevant inhibitory properties on this enzyme (Li et al. 2009). No effects were found at assayed concentrations with the hexane: acetone extracts on the assayed enzymes (Table 3).

Antimicrobial activity of leaf extracts on phytopathogenic bacteria

The 30% methanol extracts have shown antibacterial activity for some *Pseudomonas* species and *Clavibacter michiganensis* AB299158 (Online Resource Table 7). No inhibitory effects were observed with the other extracts at the assayed concentrations (0.01–2.0 mg mL⁻¹). Despite the fact that the MIC values of the standard antibiotics were more effective than those of the aqueous-methanol extracts, the preparations showed inhibitory effects at less than 1 mg mL⁻¹ in *Pseudomonas syringae* pv. tabaci TBR2004, *P. syringae* pv. tomato DC3000, *P. syringae* pv. glycinea and *Clavibacter michiganensis* AB299158. As a result, this could be a less expensive alternative for the biological control of these phytopathogens (Online Resource Table 7), primarily because biological control of *Pseudomonas*

species and *C. michiganensis* with hydroalcoholic extracts has been poorly explored. A previous work reported the growth inhibition of *Clavibacter michiganensis* and *P. syringae* pv. tomato by the olive mill wastewaters containing citric and other organic acids (Özdemir 2009). In accordance with our results, MIC values from fall and winter extracts were more effective. Coincidentally, these extracts had the highest levels of oxalic and citric acids in leaves (Online Resource Tables 1–7). The selective cytotoxic activity of oxalic acid has been demonstrated on avian pathogenic *Pseudomonas* sp. (Anang et al. 2006). According to these results, the hydroalcoholic extracts of *B. nelumbiifolia* could be useful to design industrial products for the biological control of the assayed bacterial species.

Conclusion

This work demonstrates the nutritional and some nutraceutical properties of the edible stalks of *B. nelumbiifolia* due to the organic acids, carotenes and flavonoids content. The hexane: acetone and alcoholic extracts showed a relevant antioxidant activity on DPPH as well as inhibitory activity on HMG-CoA and alpha-glucosidase enzymes. Additionally, the 30% MeOH extracts showed inhibitory activity on the assayed phytopathogenic bacteria.

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