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Original Article

Phytochemical composition and antioxidant capacity of the aqueous extracts of *Malva sylvestris* L. and *Malva pseudolavatera* Webb & Berthel.

[Composición fitoquímica y capacidad antioxidante de los extractos acuosos de *Malva sylvestris* L. y *Malva pseudolavatera* Webb & Berthel.]

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Abstract

Context: *Malva* spp. have been widely used in the world as traditional remedies. In Ecuadorian markers *Malva sylvestris* and *Malva pseudolavatera* are the species most commercialized. However, the scientific information about *M. pseudolavatera* is little.

Aims: To determine the chemical composition and antioxidant capacity of the aqueous extracts of the leaves of both species.

Methods: Aqueous extracts were partitioned with butanol and the fractions obtained were analyzed by GC/MS. Total extracts were evaluated as antioxidant using FRAP, DPPH and ABTS assays.

Results: Differences in the chemical composition were found between the extracts. Both extracts showed the presence of polysaccharides, phenolic acids and fatty acids, but in *M. sylvestris* were identified polyols, which there was not observed in *M. pseudolavatera*. The aqueous extracts showed free radical scavenging and ferric reducing power capacities *in vitro* assays, being *M. sylvestris* the most promissory antioxidant, possibly due to the presence of polyols in the extract. In the present research were reported 39 phytochemical compounds by first time for the specie *M. pseudolavatera*.

Conclusions: Aqueous extracts of the leaves of *M. sylvestris* and *M. pseudolavatera* showed antioxidant capacity associated with the presence of phenolic acids, polysaccharides and flavonoids in the extracts.

Keywords: antioxidant; gas chromatography; *Malva pseudolavatera*; *Malva sylvestris*; phytochemicals.

Resumen

Contexto: Las especies de *Malva* son muy empleadas mundialmente como remedios tradicionales. En los mercados ecuatorianos se comercializa mayoritariamente las especies *Malva sylvestris* y *Malva pseudolavatera*, sin embargo, sobre esta última especie la información científica es escasa.

Objetivos: Determinar la composición química y capacidad antioxidante de los extractos acuosos de las hojas de ambas especies.

Métodos: Los extractos acuosos fueron particionados con butanol para posterior análisis por CG/EM. La capacidad antioxidante de los extractos totales fue evaluada mediante los ensayos de FRAP, DPPH y ABTS.

Resultados: Se encontraron diferencias en la composición química de los extractos. En ambas especies se observó la presencia de polisácaridos, ácidos fenólicos y ácidos grasos; sin embargo, en *M. sylvestris* se identificaron polioles que no fueron observados en *M. pseudolavatera*. Los extractos acuosos mostraron capacidad secuestradora de radicales y poder reductor del hierro en los ensayos *in vitro* realizados. *M. sylvestris* resultó ser la especie más promisoria como antioxidante, probablemente asociado a la presencia de polioles. En el estudio se reportaron, por primera vez, 39 compuestos químicos para la especie *M. pseudolavatera*.

Conclusiones: Los extractos acuosos de las hojas de *M. sylvestris* y *M. pseudolavatera* mostraron capacidad antioxidante asociada con la presencia de ácidos fenólicos, polisacáridos y flavonoides en los extractos.

Palabras Clave: antioxidante; cromatografía de gases; fitoquímica; *Malva pseudolavatera*; *Malva sylvestris*.

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INTRODUCTION

During time medicinal plants have been used as traditional medicines, remedies, potions and oils without any knowledge about the bioactive compounds contained inside, but just considering results of hundreds of centuries of man experimenting (Bernardini et al., 2018). These ethnobotanical references are the first clinical evidence about the efficacy of natural products like therapeutic agents, however, phytochemical, pharmacological and toxicological studies are necessary to corroborate the ethnomedical information (Patra et al., 2020).

Malva genus, popularly known as mallow, grows spontaneously in almost all of Europe and the Mediterranean region; however, their culture has been extended to intertropical region due to their application as ornamental, textile, food and medicinal plants (Paloschi de Oliveira et al., 2019). It has 25-40 species, of which, *M. neglecta*, *M. parviflora*, *M. sylvestris* and *M. verticillata* are the most reported in the specialized literature (Azab, 2017). In addition, *Malva* species are traditionally used like diuretic, spasmolytic, antidiarrheic, expectorant, laxative, antitussive and anti-inflammatory (Sharifi-Rad et al., 2020). In America, *M. sylvestris* is commercialized in the traditional markers of Argentina, Mexico, Chile, Colombia, Bolivia, Peru and Ecuador (Acosta et al., 2017; Bussmann et al., 2016; Villanueva-Solis et al., 2020). However, the most popular mallow commercialized in Ecuadorian markers is the leaves of *M. pseudolavatera* for decoctions; on which, the scientific studies that support its traditional use are little (Tinitana et al., 2016).

In this context, the present research is led to determine the phytochemical composition and antioxidant capacity of the aqueous extracts of *Malva pseudolavatera* and *Malva sylvestris* growing in Ecuador according to their traditional usage.

MATERIAL AND METHODS

Plant material

The leaves of *M. sylvestris* and *M. pseudolavatera* were collected in November 2019 in the city of Riobamba, province of Chimborazo, in the Andes at 2750 meters above sea level (1°40'15.5"S 78°38'49.6"W). A sample of each species was deposited in the GUAY herbarium of the Faculty of Natural Sciences of the University of Guayaquil, Ecuador, where a voucher specimen (No. 13118 and 13119, respectively) was deposited. The species were genetically characterized by Sarmiento et al. (2020). The leaves were washed with potable water and dried in a Mettler Toledo oven at 40°C to a constant weight, they were crushed in a Pulvex mill with blades to a particle size of 2 mm.

Obtaining and fractionating of the aqueous extracts

Twenty grams of drug were extracted with 100 mL of distillated water by decoction during 15 min. and partitioned with butanol. The fractions were dried and stored for further analysis (Miranda and Cuéllar, 2000).

Gas Chromatography tandem Mass Spectrometry (GC/MS) analysis

The butanol and aqueous fractions of *M. sylvestris* and *M. psuedolavatera* were analyzed by GC/MS on an Agilent Technology instrument following a previous analysis described by Miranda et al. (2020), 10 µL of dry fractions were derivatized with 100 µL of BSTFA and collocated in water bathroom at 80°C during 2 hours. An DB-5MS column (30 m, 0.25 mm ID × 0.25 µm) was used. The temperature of the oven was increased from 70°C until 300°C at 5°C/min. The inlet and detector temperatures were 250°C and 230°C, respectively. Helium gas was used as the mobile phase at flow of 1 mL/min. The electron gun of mass detector liberated electrons having energy of about 70 eV. The identification of compounds was done using the NIST 2011 data base.

Antioxidant protocols

Ferric Reducing Antioxidant Power (FRAP)

The FRAP assay was done according to Benzie and Strain (1996) with some modifications. The FRAP reagent was prepared in situ by mixing 0.1 mol/L of sodium acetate buffer (pH 3.6), 10 mmol/L of 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ) and 20 mmol/L of ferric chloride (10: 1: 1, v: v: v) and then warmed at 37°C before using. Aqueous extracts were prepared at the concentrations of 25, 50, 100, 150 and 200 µg/mL. Test samples (30 µL) and water (90 µL) were allowed to react with 900 µL of the FRAP solution for 30 min in the dark. Readings of the colored product (ferrous tripyridyltriazine complex) were then done at 593 nm. The blank consisted of 120 µL of water and 900 µL of reagent. The results were expressed as µmol equivalent of ascorbic acid, according to the standard curve of ascorbic acid (100-1000 µmol/L). The results were expressed as µmol equivalents of ascorbic acid (EAA) and µmol equivalents of $FeSO_4$, interpolating the optical density (OD) of the samples in the calibration curves of both reference substances at the concentrations of 100, 200, 400, 500 and 800 µM. The calibration curves for ascoribic acid and ferric sulfate were Y =

0.00046X + 0.09948 (R^2 = 0.9889) and Y = 0.00049X + 0.11502 (R^2 = 0.9887), respectively.

DPPH radical scavenging assay

The used method was a modification to that described by Brand-Williams et al. (1995) and Kedare and Singh (2011). The assay is based on the reduction of the free radical 2,2-diphenyl-1-picryl-hydrazyl (DPPH•). Aqueous extracts were prepared at the concentrations of 25, 50, 100, 150 and 200 µg/mL. For the analysis, 90 µL of absolute ethanol were added to 10 µL of each test sample and mixed with 900 µL of DPPH reagent (0.075 mg/mL). The equation of the straight line and the R^2 of both reference substances calibration curves were vitamin C: Y = 0.207X + 46.28 (R^2 = 0.9760) and Trolox: Y = 0.172X + 47.84 (R^2 = 0.9835). Absolute ethanol (100 µL) was used as blank. The reaction was left in the dark for 30 min and, subsequently, UV absorbance was measured at 517 nm. Each de-termination was performed in triplicate. Trolox was used as reference compound.

The percentage of inhibition was calculated using the following equation [1]:

$$\text{Inhibition (\%)} = [(\text{Abs control} - \text{Abs sample})/\text{Abs control}] \times 100 \quad [1]$$

Where Abs control: Abs of blank + DPPH and Abs sample: Abs of fraction + DPPH

The mean effective concentration (IC_{50}) was determined with the help of the Statgraphics Plus versión 5.1 statistical program.

ABTS radical scavenging assay

ABTS radical assay was done according to the method described by Re et al. (1999) with slightly modifications. The ABTS•+ stock solution was produced by reacting ABTS aqueous solution (7 mM) with 2.45 mM aqueous solution of potassium persulfate in equal quantities and allowed them to react for 16 h at room temperature in the dark. Then, 980 µL of ABTS•+ solution was mixed with 20 µL of the aqueous extract or standard (ascorbic acid) at different concentrations (100-500 µg/mL). The equation of the straight lines and the R^2 of both reference substances calibration curves were vitamin C: Y = 0.120X + 37.36 (R^2 = 0.9590) and Trolox: Y = 0.127X + 27.42 (R^2 = 0.9860). The mixture was then incubated at room temperature for exactly 30 min in the dark. Distillated water was used as control. The absorbance was measured at 734 nm.

The percentage results of scavenging activity were calculated as % inhibition using above equation [2]:

$$\text{Inhibition (\%)} = [A_{734}\,(\text{ABTS}) - A_{734}\,(\text{extract})]/A_{734}\,(\text{ABTS}) \times 100 \quad [2]$$

Where A_{734} was the absorbance of ABTS and extract at 734 nm.

The mean effective concentration (IC_{50}) was determined with the help of the Statgraphics Plus versión 5.1 statistical program.

Statistical analysis

Data were analyzed using the statistical program Graph Pad Prism 5 (GraphPad Software, Inc., La Jolla, CA, USA). Inter-group statistically significant differences were tested using a one-way analysis of variance (ANOVA) followed by Bonferroni's or Dunnett's *posthoc* tests for multiple comparisons. The results are presented as mean ± SEM. $P<0.05$ was considered statistically significant.

RESULTS

Phytochemical composition

For the phytochemical study of *M. pseudolavatera*, aqueous extract was partitioned with butanol and both fractions were analyzed by GC/MS (Figs. 1-2). In aqueous fraction were identified more compounds than butanol fraction. Aqueous fraction was characterized by the presence of fatty acids, polysaccharides and hydrocarbon compounds while in butanol fraction predominated phenolic acids and polysaccharides. The majoritarian compounds identified in the aqueous extract of *M. pseudolavatera* were D(-)-fructose (5.06%), palmitic acid (4.47%), octadecanoic acid (4.22%), D(-)-fructofuranose (3.80%) and succinic acid (2.28%) (Tables 1-2).

On the other hand, the aqueous extract of *Malva sylvestris* (Figs. 3-4) was characterized by the presence of polyols, fatty acids, polysaccharides and phenolic acids. However, the main compound identified in this extract was the steroid, 3β-17β-5α-androstane (4.55%). Other majoritarian compounds were palmitic acid (2.87%), octadecanoic acid (2.75%) and methyl-α-D-glucofuranoside (2.06%) (Tables 3-4).

The phytochemical composition of the aqueous extract of *M. pseudolavatera* is reported by first time in the present research, contributing to the scientific knowledges of this specie widely commercialized in Ecuadorian markers and compared with *M. sylvestris*, one of the medicinal plants most studied for the scientific community. Qualitative and quantitative differences were observed between both samples. These differences were associated to intrinsic factors, mainly to the genetical variability inter-species, previously reported (Sarmiento et al., 2020a).

For example, palmitic and octadecanoic acids were identified as majoritarian compounds in both samples. However, the abundance relative of these compounds in *M. pseudolavatera* (4.74 and 4.22%, respec-

tively) was higher than *M. sylvestris* (2.87 and 2.75%, respectively). In contrast, the main compound detected in *M. sylvestris* was 3β-17β-5α-androstane (4.55%), which was not observed in *M. pseudolavatera*. In addition, the presence of polyols, such as, arabitol, xylitol and sorbitol only were detected in *M. sylvestris.*

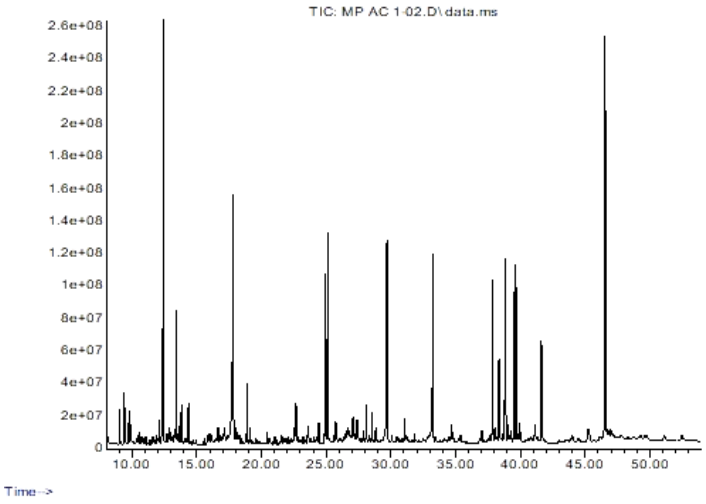

Figure 1. Chromatogram GC/MS of the aqueous fraction of the aqueous extract of leaves of *Malva pseudolavatera.*

Table 1. Compounds identified in the aqueous fraction of the aqueous extract of leaves of *Malva pseudolavatera*.

RT (min)	Compound	RA (%)
9.060	Propanoic acid	0.42/0.02
11.853	4,6-Dimethyldodecane	0.15/0.00
13.406	Succinic acid	2.28/0.04
14.360	Fumaric acid	0.87/0.03
14.703	2-Methyltridecane	0.10/0.01
15.626	Tetradecane	0.11/0.01
18.323	Meso-erythritol	0.15/0.01
19.138	2,3,4-Trihydroxybutiric acid	0.37/0.04
19.596	7-Hexyltridecane	0.12/0.02
20.455	Hexadecane	0.21/0.04
21.523	2,6,10-Trimethylpentadecane	0.15/0.01
23.158	L-(-)-Arabitol	0.12/0.02
24.857	Octadecane	0.14/0.01
25.118	D-(-)-Fructofuranose	3.80/0.15
26.543	Gulconic acid	0.17/0.01
26.651	D-(-)-Tagatose	0.50/0.02
27.911	Nonadecane	0.23/0.04
28.808	7,9-diterbutyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	0.23/0.05
26.692	Palmitic acid	4.47/0.23
30.437	2-Methyltricosane	0.25/0.02
33.242	Octadecanoic acid	4.22/0.16
36.967	D-(-)-Ribose	0.31/0.01

Results of relative abundance (RA) are expressed as medium/standard deviation (n = 3). RT: retention time.

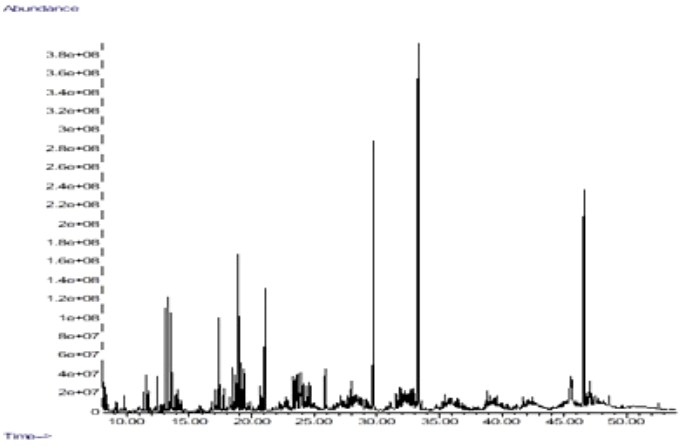

Figure 2. Chromatogram GC/MS of the butanolic fraction of the aqueous extract of leaves of *Malva pseudolavatera.*

Table 2. Compounds identified in the butanolic fraction of the aqueous extract of leaves of *Malva pseudolavatera*.

RT (min)	Compound	RA (%)
9.060	Propanoic acid	0.25/0.02
11.580	Benzoic acid	0.42/0.02
13.406	Succinic acid	0.83/0.18
18.890	Benzoic acid, 3,5-bis(1,1-dimethylethyl)-4 hydroxy-ethyl ester	0.61/0.02
20.283	α-Linoleic acid	0.10/0.04
20.601	2-Methyl-1-hexadecanol	0.16/0.02
21.135	Ribonic acid	0.29/0.07
23.820	1,5-Anhydro-D-Sorbitol	1.14/0.17
25.143	D(-)-Fructose	5.06/0.28
26.670	D-Psicose	1.85/0.14
26.925	β-D-Talopyranose	0.31/0.02
27.007	D-Xylopiranose	0.67/0.02
29.616	Palmitinic acid	0.65/0.03
32.892	β-D-Galactopyranoside, Methyl	0.15/0.01
33.242	Octadecanoic acid	0.15/0.02
35.660	α-D-Galactoside	0.16/0.03
36.474	Uridine	0.20/0.01
43.269	Octadecane, 3-ethyl-5-(2-ethylbutyl)	0.32/0.01

Results of relative abundance (RA) are expressed as medium/standard deviation (n = 3). RT: retention time

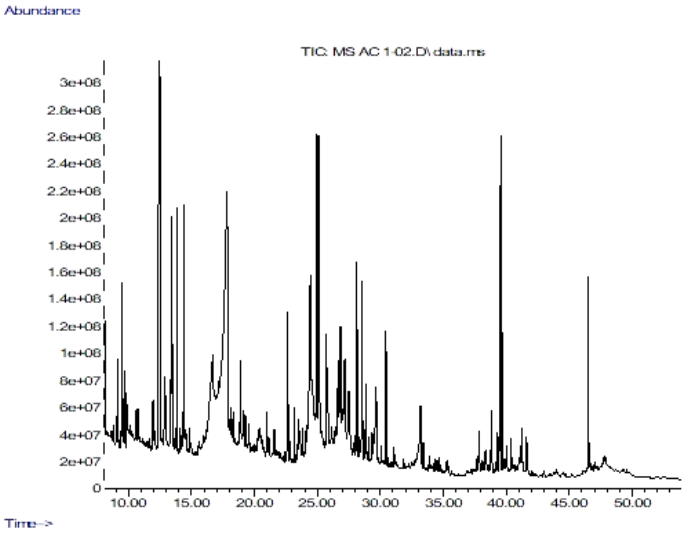

Figure 3. Chromatogram GC/MS of the aqueous fraction of the aqueous extract of leaves of *Malva sylvestris.*

Table 3. Compounds identified in the aqueous fraction of the aqueous extract of leaves of *Malva sylvestris*.

RT (min)	Compound	RA (%)
11.879	Serine	0.42/0.03
20.995	Arabinonic acid	0.24/0.01
21.523	Methyl 2-(acetylamino)-2-deoxy-α-D-Glucopyranoside	0.12/0.06
22.859	D-(+)-Arabitol	0.13/0.00
23.171	Xylitol	0.30/0.00
24.444	3β-17β-5α-Androstane	4.55/1.19
26.136	α-D-Glucopyranosiduronic acid, 3-(5-ethylhexahydroxo-2,4,6-trioxo-5-pyriminidil)-1,1-dimethylpropyl	0.45/0.10
26.562	D-(+)-Gluconic acid δ-Lactone	0.48/0.02
26.645	D-Galactonic acid ϒ-Lactone	0.89/0.02
27.580	D-Sorbitol	0.42/0.03
27.962	Methyl β-D-(+)-Mannopyranose	0.34/0.01
29.667	Palmitic acid	1.41/0.05
30.443	Inositol	0.90/0.55
31.066	Docosane	0.14/0.00
33.198	Stearic acid	1.29/0.07

Results of relative abundance (RA) are expressed as medium/standard deviation (n = 3). RT: retention time.

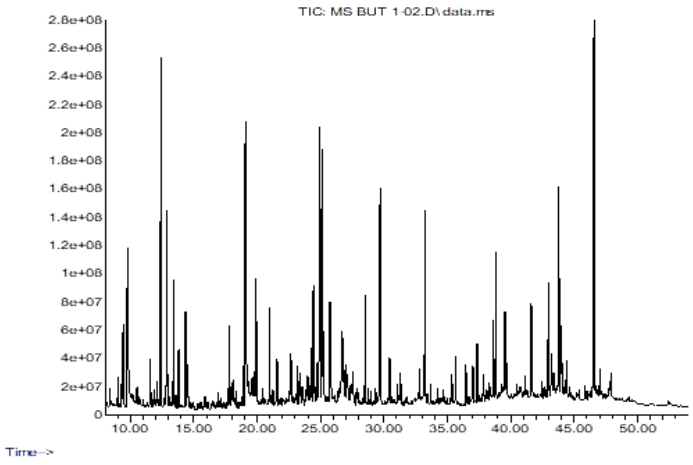

Figure 4. Chromatogram GC/MS of the butanolic fraction of the aqueous extract of leaves of *Malva sylvestris*.

Table 4. Compounds identified in the butanolic fraction of the aqueous extract of leaves of *Malva sylvestris*.

RT (min)	Compound	RA (%)
11.58	Benzoic acid	0.40/0.09
13.399	Succinic acid	1.26/0.04
14.353	2-Butanedioic acid	0.79/0.04
19.316	DL-Phenylalanine	0.64/0.19
19.685	4-Hydroxyphenylethanol	0.33/0.09
23.985	Vanillic acid	0.45/0.02
24.515	Cinnamic acid	0.41/0.03
24.755	Azelaic acid	0.58/0.02
25.748	Methyl-α-D-Glucofuranoside	2.06/0.04
26.543	D-Gluconic acid	0.43/0.08
26.664	D-Psicose	0.77/0.22
26.721	α-D-Mannopyranose	1.02/0.02
26.823	α-D-Galactoside	0.57/0.02
26.925	β-D-Talapyranose	0.23/0.00
27.001	D-Xylanopyranose	0.42/0.02
29.667	Palmitic acid	2.87/0.05
30.456	Isoferulic acid	0.63/0.03
33.249	Octadecanoic acid	2.75/0.06
36.468	Uridine	0.41/0.02
37.028	4-Methylthio-N-phenyl-1,2-carbazoledicarboximide	0.50/0.07
38.815	Monopalmitin	1.80/0.06
39.553	Sucrose	1.60/0.03
42.963	Acubin	1.44/0.06

Results of relative abundance (RA) are expressed as medium/standard deviation (n = 3). RT: retention time

In vitro antioxidant capacity

The radical scavenging capacity of aqueous extracts of *M. pseudolavatera* and *M. sylvestris* was measured using DPPH and ABTS assays. Vitamin C and Trolox were used as positive control. In both extracts, a proportional increase was observed between the antioxidant activity and the concentration of the extracts. The values of IC_{50} (concentration required to scavenge 50% of free radicals) for *M. sylvestris* were lower than *M. pseudolavatera* for both assays (Tables 5-6).

The comparison between the extracts and control groups showed differences with respect to the assay. For example, in DPPH, both extracts showed higher values of IC_{50} than Trolox and vitamin C. However, *M. sylvestris* showed lower values of IC_{50} than Trolox and higher than vitamin C. The ABTS assay is applicable to hydrophilic and lipophilic antioxidant systems; whereas DPPH assay is generally used for hydrophobic systems (Floegel et al., 2011), which can be explain the differences found in the results.

These results demonstrated that *M. sylvestris* has a higher radical scavenging capacity than *M. pseudolavatera* and Trolox, but lower than vitamin C.

On the other hand, the reducing capacity of the extract was determined by FRAP assay, using vitamin C and ferric sulphate like standards. The extracts showed a positive concentration-response effect, being the aqueous extract of *M. sylvestris* at 200 µg/mL the most active (Table 7).

Table 5. Effect of the aqueous extracts of *Malva sylvestris* and *Malva pseudolavatera* on DPPH radical scavenging capacity.

Concentration (µg/mL)	DPPH radical scavenging (%)			
	Malva sylvestris	***Malva pseudolavatera***	**Vitamin C**	**Trolox**
25	$37.38/0.68^{a}$	$35.63/0.52^{b}$	$51.55/0.82^{c}$	$53.20/0.75^{d}$
50	$43.44/0.91^{e}$	$39.43/0.52^{f}$	$56.45/0.56^{g}$	$57.50/0.75^{g}$
100	$58.50/0.52^{h}$	$55.05/0.45^{i}$	$64.51/0.85^{j}$	$61.06/1.14^{k}$
150	$64.41/0.54^{l}$	$59.90/1.13^{m}$	$82.88/0.60^{n}$	$75.02/0.76^{o}$
200	$74.52/0.52^{p}$	$69.91/0.38^{q}$	$85.03/0.67^{r}$	$82.98/0.22^{s}$
IC_{50}	78.14	95.04	17.97	12.55

The results are expressed as medium/standard deviation. Different letters indicate significative differences ($p<0.05$) by One Way ANOVA followed by Turkey tests.

Table 6. Effect of the aqueous extracts of *Malva sylvestris* and *Malva pseudolavatera* on ABTS radical scavenging capacity.

Concentration (µg/mL)	ABTS radical scavenging (%)			
	Malva sylvestris	***Malva pseudolavatera***	**Vitamin C**	**Trolox**
100	$44.86/0.58^{a}$	$41.83/0.44^{b}$	$46.52/0.73^{c}$	$39.87/1.94^{d}$
200	$53.85/0.60^{e}$	$52.09/0.89^{f}$	$58.94/0.59^{g}$	$53.90/0.89^{e}$
300	$61.04/0.66^{h}$	$58.11/0.59^{i}$	$81.86/0.69^{j}$	$62.06/0.56^{h}$
400	$68.91/0.53^{k}$	$66.46/0.85^{l}$	$88.99/1.02^{m}$	$83.52/1.56^{n}$
500	$83.27/0.67^{o}$	$78.88/0.81^{p}$	$91.98/0.80^{q}$	$88.70/0.27^{r}$
IC_{50}	166.79	193.97	105.33	188.16

The results are expressed as medium/standard deviation. Different letters indicate significative differences ($p<0.05$) by One Way ANOVA followed by Turkey tests.

Table 7. Effect of the aqueous extracts of *Malva sylvestris* and *Malva pseudolavatera* on ferric reduction power antioxidant activity.

Concentration (µg/mL)	µM equivalents of ascorbic acid		µM equivalents of $FeSO_4$	
	Malva sylvestris	***Malva pseudolavatera***	***Malva sylvestris***	***Malva pseudolavatera***
25	$149.67/3.32^{a}$	$142.43/2.17^{b}$	$108.80/3.11^{a}$	$102.00/2.04^{b}$
50	$188.08/8.69^{c}$	$169.28/5.46^{d}$	$144.85/8.16^{c}$	$123.76/8.2^{d}$
100	$251.12/12.10^{e}$	$231.56/14.25^{f}$	$204.03/11.36^{e}$	$185.66/13.38^{f}$
150	$406.20/13.10^{g}$	$376.49/15.26^{h}$	$349.61/12.30^{g}$	$321.72/14.33^{h}$
200	$502.57/10.27^{i}$	$468.51/9.95^{j}$	$440.09/9.64^{i}$	$408.11/9.35^{j}$

The results are expressed as medium/standard deviation. Different letters indicate significative differences ($p<0.05$) by t-students test.

DISCUSSION

Medicinal plants are composed of a wide variety of chemical compounds related to external factors such as environmental conditions and age of the plant (Li et al., 2020; Taylor et al., 2001). Therefore, current drug discovery strategies and modern medicine discard the use of whole plant extracts and are driven by single compound-based medicine. However, the use of extracts like active pharmaceutical ingredients produces a better therapeutic result than individual compounds due to the synergistic effect between the metabolites (Thomford et al., 2018). In this context, phytochemical and biological studies led to identify chemical markers in the extracts are necessary in order to guarantee their quality, efficacy and safety (Rivera-Mondragón et al., 2017).

In the present research, the phytochemical composition and antioxidant activity of the aqueous extracts of *M. sylvestris* and *M. psuedolavatera* were compared. Chemically, both extracts showed presence of fatty acids, polysaccharides and phenolic acids. However, in *M. sylvestris* was observed a wide variety of polyols like xylitol, sorbitol and arabitol, which were not present in *M. pseudolavatera.*

The presence of polysaccharides and phenolic acids in plants has been associated with anticancer, immunomodulatory, antiviral, antibacterial, antioxidant, hypoglycemic and anti-hypercholesterolemic activities (Bandara et al., 2019; Rashmi and Negi, 2020). These compounds can be responsible of the antioxidant activity demonstrated for both extracts by FRAP, DPPH and ABTS assays (Floegel et al., 2011).

In the experimental conditions, *M. sylvestris* showed higher antioxidant effect than *M. pseudolavatera*. In a previous study, the addition of polyols in the extraction solvent increased the antioxidant activity of *Camellia oleifera* (Tsai and Lin, 2019). On the other hand, the presence of flavonoids and phenol compounds have been reported previously in both species. In fact, the content of phenolic compounds and flavonoids in the hydroalcoholic extract of *M. sylvestris* was higher than *M. pseudolavatera* (Sarmiento et al., 2020b). Therefore, the presence of polyols in *M. sylvestris* and its higher content of flavonoids and phenolic compounds can be related to the differences found in the antioxidant activity between the species.

In total, 41 compounds were identified in *M. pseudolavatera*, of which, only palmitic and octadecanoic acid were reported previously (Sarmiento et al., 2020b), the other compounds are reported by first time for this specie. The antioxidant activity of this plant was reported previously in DPPH, ABTS and FRAP assays for the hydroalcoholic extract using the same concentrations (Sarmiento et al., 2020c), but the present research is the first report for the aqueous extract obtained in similar conditions to ethnomedical usage. The aqueous extract showed lower antioxidant activity than hydroalcoholic extracts for the three assays evaluated.

The present research increased the scientific knowledge of *M. pseudolavatera*, a mallow species more commercialized in the Ecuadorian markers. However, *M. sylvestris* showed higher IC_{50} values than *M. pseudolavatera* in the antioxidant assays evaluated.

CONCLUSION

Aqueous extracts of the leaves of *Malva sylvestris* and *Malva pseudolavatera* showed free radical scavenging and ferric reducing power capacities *in vitro* assays, associated with the presence of phenolic acids, polysaccharides and flavonoids in the extracts. *Malva sylvestris* showed a highest antioxidant activity possibly related with the presence of polyols in this specie.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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AUTHOR CONTRIBUTION:

Contribution	Sarmiento GM	Gutiérrez YI	Delgado R	Burbano ZC	Soledispa PA	Jaramillo ND	Vargas LA
Concepts or ideas	x	x	x	x	x		
Design	x		x				
Definition of intellectual content	x	x		x	x		
Literature search	x	x	x	x	x	x	x
Experimental studies	x	x		x	x		
Data acquisition	x	x		x	x		
Data analysis	x						
Statistical analysis	x					x	x
Manuscript preparation	x	x	x	x	x	x	x
Manuscript editing	x		x			x	x
Manuscript review	x	x	x	x	x	x	x

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