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Comparison between maceration and ultrasound-assisted extraction of white bracts with spring flowers (*Bougainvillea spectabilis* Willd.) And evaluation of their potential antioxidant activity

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Abstract. Obtaining bioactive compounds depends on the extraction method, with maceration being the most well-known. This method is commonly used with ethanol as an extractor solvent and requires a long period to complete the process. Alternative methods, such as ultrasound-assisted extraction, have been applied to optimize these processes. In this context, to evaluate the effect of extraction time, the ultrasound-assisted and the maceration extraction methods were performed on white bracts and flowers from *Bougainvillea spectabilis* Willd. (Nyctaginaceae). The antioxidant activities and fatty acid content of the different extracts were determined. The amount of phenolic compounds obtained for the maceration technique was greater, however, in the evaluation of the potential antioxidant activity by the DPPH and $ABTS^+$ free radicals scavenging, no significant difference was verified when compared to the extraction by ultrasound for 30 minutes. In the determination of fatty acids by gas chromatography coupled with mass spectrometer it was found that the highest contents were of palmitic, linoleic, and stearic acids for both extraction methods. The 30 minutes of extraction by ultrasound proved to be the time necessary to quickly and efficiently extract the bioactive compounds from *B. spectabilis*. Thus, the use of ultrasound-assisted extraction, when compared to the maceration method, is an alternative for obtaining plant extracts in a short time, reducing the instability that some compounds
Keywords: Extraction methods, phenols, fatty acids, gas chromatography

Introduction

The search for new bioactive compounds, from the secondary metabolism of plants, that can be used by the pharmaceutical, cosmetic, and food industries has been intensified in the last decade (RODRÍGUEZ-PÉREZ et al., 2015). Among the compounds of greatest interest, phenolic compounds stand out, to which antifungal, antibacterial, antiviral, anti-inflammatory, antiallergic, and cardioprotective activities are attributed, among others (AL-SNAFI, 2019; CÉSPEDES et al., 2008; CHOI et al., 2008 ., 2013; ŠAMEC et al., 2010; WALSH et al., 2019; WENG et al., 2019).

Species of the Bougainvillea genus are originally from South America and have attractive bioactive compounds for the development of new products. B*ougainvillea spectabilis* Willd. (Nyctaginaceae) (INTERNATIONAL PLANT NAMES INDEX (IPNI), 1799), known as primavera, três-marias and flor-de-papel, stands out for presenting phenolic compounds with potential antioxidant activity (ALI et al., 2005; CHAIRES-MARTÍNEZ et al ., 2009; SWAMY et al., 2012), potential antibacterial activity, and antidiabetic activity (BHAT et al., 2009; CHAIRES-MARTÍNEZ et al., 2009; JAWLA; KUMAR; KHAN, 2012).

To obtain these compounds, the extraction process is basically the most important step of the analysis, which happens through the action of a solvent in direct contact with the plant cell structure (MEREGALLI et al., 2020; PHAM et al., 2020). Aspects such as type of solvent, temperature, pH of the medium, and time of extraction directly interfere in the process of obtaining bioactive compounds, and as a result can alter their biological activities (NADEEM et al., 2018).

The commonly used techniques, so called conventional, are based on the extraction power of the solvents, the need for agitation, and the application of high temperatures, which often makes the process flawed by interfering in the obtention of the compounds, in addition to requiring a long period

of time to finish the process (SAFDAR et al., 2017; SIMÕES et al., 2010). To overcome the limitations of these methods, new and promising techniques have been used, such as extraction with supercritical fluid, extraction assisted by pulsed electric field, microwave, or ultrasound (SOQUETTA; TERRA; BASTOS, 2018).

In general, the alternative techniques aim to optimize and increase the selectivity and yield of the extracted compounds, to decrease the use of solvents providing less harmful and safer products, and to reduce energy consumption and the time to obtain the extracts (OAK; BERMAMASCO; GOMES, 2018; SAFDAR et al., 2017). In this context, ultrasound-assisted extraction stands out for its ease of use, for presenting good extraction yields, and for having a more accessible cost when compared to other alternative techniques such as microwave and supercritical fluid (FERNÁNDEZ-BARBERO et al., 2019; SOQUETTA; TERRA; BASTOS, 2018; TIAN et al., 2013).

Ultrasound works by causing lysis in the cell wall structure of plants through cavitation by microbubbles of the liquid phase, facilitating the diffusion of metabolites through the plant membranes to the solvent, causing a considerable decrease in the extraction time (GONZÁLEZ-CENTENO et al., 2014).

Thus, the aim of the study was to use ultrasound-assisted extraction and asses the decrease in the time taken to obtain phenolic compounds, to evaluate the potential antioxidant activity of *B. spectabilis* white bracts and flowers, and to compare these results with the extraction by maceration.

Methods

Plant material obtention

The white bracts and flowers of *Bougainvillea spectabilis* Willd (Nyctaginaceae) were collected in the municipality of Sinop, Mato Grosso (11° 51 '52 "S 55° 30' 29" W) and identified in the Mato Grosso North Center Herbarium (CNMT) of the Federal University of Mato Grosso, *Campus* of Sinop, where the exsiccata of the plant was deposited under the registration number CNMT 10413.

The plant material was sent to the Quality Control laboratory at UFMT, Campus of Sinop, where they were screened, then dried in a forced convection drying oven at a temperature of 40 ± 1 °C, for a period of 48 hours. Subsequently, the material was ground in a mill, identified, and stored at room temperature sheltered from light.

Reagents

All reagents and solvents used in the analyzes were of analytical grade. Ethyl alcohol, methyl alcohol, aluminum chloride ($AlCl_3$), and sodium carbonate (Na_2CO_3) were supplied by Synth (Diadema, SP, Brazil) and the Folin-Ciocateau solution by Dinâmica (Indaiatuba, SP, Brazil). 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2′-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid (ABTS™), 6-Hydroxy-2,5,7,8-tetramethyl chroman-2-carboxylic acid (Trolox™), and gallic acid were purchased from Sigma Aldrich Co. (St. Louis, MO, USA).

Extracts obtention

Maceration

The white bracts and flowers were subjected to extraction by maceration in absolute ethanol (EtOHa) in the proportion of 1:8 (m/v) at room temperature for a period of seven days with manual agitation every 24 hours (SIMÕES et al. , 2010). After that, the mixture was filtered, and the solvent was removed on a rotary evaporator at 50 °C. The extract was weighed and stored sheltered from light and under refrigeration.

Ultrasound-assisted extraction

For the ultrasound-assisted extraction, a mixture of plant material and EtOHa was used in the proportion of 1:8 (m/v). The mixture was put in six bottles, and each one was sonicated for different periods (15, 30, 60, 120, 180, and 240 minutes) in an ultrasonic bath (Cristófoli®) in a frequency of 42 kHz and 170 W of power, at room temperature. Then the material was filtered, and the solvent was eliminated in a rotary evaporator (Fisatom®) at 50 °C. The extracts were weighed and stored sheltered from light under refrigeration.

Determination of fatty acids by Gas Chromatography coupled to Mass Spectrometry (GC-MS)

The fatty acid composition of the extracts was determined in a gas chromatograph (Shimadzu - GCMS-QP2010 Ultra) connected to a mass spectrometer (QP2010 Ultra), using helium as the carrier gas (1.0 $mLmin^{-1}$) under the following conditions: 1 µL of sample injection volume; Split 3:1; column: HP5-MS; MS source: 230 °C; Quad MS: 150 °C; gradient: 140 °C (2 minutes), 4 $°Cmin^{-1}$ to 180 °C, 0.5 $°Cmin^{-1}$ to 200 °C, 5 $°Cmin^{-1}$ to 250 °C (3 minutes); Full time: 70 minutes.

The extracts were esterified according to Jham et al. (1982), and then fatty acids were identified based on the retention time of the standard (Lipid Standards Sigma-Aldrich: FAMEs mixtures C8:0 – C24:0), injected under the same conditions.

Determination of total phenolic compounds

The concentration of phenolics in the extracts was determined using a spectrophotometric method (ROESLER et al., 2007). A MeOH solution of the extract in the concentration of 500 $µgmL^{-1}$ was used in the analysis. The reaction mixture was prepared by mixing 1000 µL of MeOH solution of extract, 1000 µL Folin-Ciocalteu's reagent, 1000 µL of 7.5% sodium carbonate (Na_2CO_3) solution and water. Blank was concomitantly prepared, containing 1000 µL of metanol (MeOH), 1000 µL of Folin-Ciocalteu's reagent dissolved in water and 1000 µL of 7.5% of Na_2CO_3. The samples were sheltered from light for one hour and the absorbance was determined using spectrophotometer at λmax = 765 nm. The samples

were prepared in triplicate for each analysis and the mean value of absorbance was obtained.

The same procedure was repeated for the standard solution of gallic acid and the calibration curve was constructed. Based on the measured absorbance, the concentration of phenolic compounds was read (μgml^{-1}) from the calibration curve; then the content of phenolics in the extracts was expressed in terms of gallic acid equivalent ($mgGAE.g^{-1}$ of extract) (NEVES; ALENCAR; CARPES, 2009; SANTI et al., 2014).

Determination of total flavonoids

The content of flavonoids in the plant extracts was determined using a spectrophotometric method (SILVA et al., 2017). The sample contained 1000 µL of MeOH solution of the extract in the concentration of 500 μgmL^{-1} and 2000 µL of 5% $AlCl_3$ solution dissolved in MeOH. The samples were incubated for 30 minutes at room temperature. The absorbance was determined using spectrophotometer at λmax = 425 nm. The samples were prepared in triplicate for each analysis and the mean value of absorbance was obtained.

The same procedure was repeated for the standard solution of quercetin and the calibration curve was constructed (3.0 a 9.0 μgmL^{-1}). Based on the measured absorbance, the concentration of flavonoids was determined on the calibration curve; then, the content of flavonoids in extracts was expressed in terms of quercetin equivalent ($mgQE.g^{-1}$).

Evaluation of potential antioxidant activity by the DPPH (2,2-diphenyl-1-picrilhydrazyl) radical scavenging method

The antioxidant activity by the DPPH radical scavenging method was performed as described by Rufino et al. (2007a) and Pires et al. (2017). The extracts were prepared at a concentration of 500 μgmL^{-1} and diluted to concentrations of 8.0 to 40 μgmL^{-1}. From the dilutions, 1000 µL of each sample dilution was transferred to screw-cap test tubes and 3 ml of the DPPH radical was added. The samples were homogenized and after a period of 30 minutes in the absence of light, the absorbance was read at a wavelength of 517 nm. The tests were performed in triplicate and a blank test was performed to correct the absorbance measurements. The results were expressed as EC50, which is the extract concentration needed to reduce 50% of DPPH.

Evaluation of the potential antioxidant activity by the ABTS method (2,2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)

The ABTS radical scavenging method was performed according to Rufino et al. (2007b). The extracts were prepared in EtOHa at a concentration of 500 μgmL^{-1} and then diluted to concentrations of 10 to 30 μgmL^{-1}. 60 µL of the prepared dilutions and 6 mL of the ABTS radical solution were transferred to screw-cap test tubes, and then homogenized and kept sheltered from light for six minutes. All analyzes were performed in triplicate and a blank assay was prepared to correct absorbance measurements. The absorbance reading was performed at a wavelength of 734 nm.

A calibration curve was prepared with Trolox (6-Hydroxy-2,5,7,8-tetramethyl chroman-2-carboxylic acid) in concentrations of 100 to 2000 µM. The potential antioxidant activity was calculated in relation to the Trolox activity, and the results are expressed in µM $Trolox.g^{-1}$ of sample.

Statistical analysis

The determination of total phenolic and flavonoid compounds as well as the antioxidant activity analyzes by the ABTS and DPPH methods, were compared by analysis of variance (ANOVA). Significant differences between the mean values were determined by Tukey's multiple comparison test with 95% significance ($p < 0.05$) and the results were presented with the mean ± standard deviation using the OriginPro program, v 8 (OriginLab©).

Results and discussion

The extraction process of bioactive compounds is characterized as one of the most important stages of an analysis. Factors such as the solvent used, temperature, and especially the extraction time can interfere in the obtention of compounds of interest (MEREGALLI et al., 2020; NADEEM et al., 2018). In addition to conventional extraction techniques, alternative methods that cause less harm to the environment and that can present a higher content of compounds extracted in a shorter period of time have been explored (Jacques et al., 2007).

In this work, the conventional technique of extraction by maceration for seven days was compared with the ultrasound-assisted technique at different times of extraction (15, 30, 60, 120, 180, and 240 minutes) in order to evaluate the extraction of bioactive compounds from *B. spectabilis'* white bracts and flowers.

The yield of the extracts was obtained from the ratio between the mass of plant material and the mass of the extract, as shown in Table 1.

Among the extractions, the highest yield was presented by the ultrasound for 120 minutes (4.98%), followed by maceration for seven days (4.48%), ultrasound for 240 minutes (3.73%), then 30 and 60 minutes (3.20%).

Ultrasound breaks plant cells through cavitation, increasing the permeability of the compounds towards the extracting solvent, e.g. phenolic compounds. However, with this breaking of the cell, some insoluble compounds present a decrease in the permeability in the solvent and end up reabsorbed by the plant matrix, explaining the decrease in the extracts' yield after 120 minutes of sonication. On the other hand, the absence of this mechanical stimulus in maceration, allows the compounds to remain dissolved in the medium

(ANNEGOWDA et al., 2012; DONG et al., 2010; MELECCHI et al., 2006; ZHAO; KWOK; LIANG, 2007). Thus, the ultrasound method, when compared to other extraction methods, can accelerate the process at low temperatures, causing less damage to the structural and molecular properties of the compounds of the plant matrix.

In order to quantify some fatty acids present in extracts of *B. spectabilis* bracts and flowers, a gas chromatography (GC/MS) analysis was performed for later comparison between the extraction methods and their different time periods.

The presence of fatty acids in species of the genus Bougainville has already been reported by Abarca-Vargas and Petricevich (2018) whose extracts presented a higher concentration of palmitic acid (58.45 to 81.33%), followed by stearic acid (7.10 to 17.95%) and linoleic acid (9.17 to 15.94%) as shown in Table 2.

Table 1. Crude extracts' yield of *B. spectabilis* white bracts and flowers obtained from extraction by maceration and by ultrasound methods

	Extracts in EtOHa		
Extractions	Plant material mass (g)	Extract mass (g)	Yield (%)
M7	34.00	1.5243	4.48
U15	7.03	0.2095	2.98
U30	7.48	0.2936	3.20
U60	50.00	1.6008	3.20
U120	34.00	1.6940	4.98
U180	50.00	1.7428	3.48
U240	50.00	1.8650	3.73

(EtOHa) Absolute ethanol; (M7) Seven days maceration; (U15) Ultrasound for 15 minutes; (U30) Ultrasound for 30 minutes; (U60) Ultrasound for 60 minutes; (U120) Ultrasound for 120 minutes; (U180) Ultrasound for 180 minutes; (U240) Ultrasound for 240 minutes

Table 2. Determination of fatty acids in the *B. spectabilis* white bracts and flowers' ethanolic extracts

Compound		M7		U15		U30		U60	
		Area	%	Area	%	Area	%	Area	%
Capric acid	C10:0	22195	0.30	21142	0.22	23319	0.21	79303	0.93
Lauric acid	C12:0	72374	0.97	47430	0.49	73300	0.67	90764	1.06
Myristic acid	C14:0	147061	1.98	140714	1.44	227746	2.08	231665	2.71
Palmitic acid	C16:0	5260174	70.75	7490004	76.90	7261629	66.45	4995274	58.45
Palmitoleic acid	C16:1	745	0.01	16778	0.17	8492	0.08	1177	0.01
Stearic acid	C18:0	527761	7.10	1029688	10.57	1433623	13.12	1533846	17.95
Oleic acid	C18:1	1482	0.02	119	N.D	15075	0.14	4713	0.06
Linoleic acid	C18:2	1185497	15.94	893571	9.17	1528290	13.99	1311546	15.35
Arachidic acid	C20:0	4905	0.07	4828	0.05	145664	1.33	51337	0.60
Beenic acid	C22:0	31161	0.42	29507	0.30	2837	0.03	1116	0.01
Erucic acid	C22:1	223	N.D	989	0.01	7242	0.07	612	0.01
Lignoceric acid	C24:0	181521	2.44	65748	0.67	200259	1.83	244868	2.87
Total		7435099	100	9740518	100	10927476	100	8546221	100
Compound		U120		U180		U240			
		Area	%	Area	%	Area	%		
Capric acid	C10:0	3454	0.44	17325	0.26	26756	0.28		
Lauric acid	C12:0	6266	0.80	72395	1.10	110426	1.16		
Myristic acid	C14:0	15525	1.99	207506	3.16	339104	3.57		
Palmitic acid	C16:0	725570	93.15	4057593	61.87	5597698	58.94		
Palmitoleic acid	C16:1	1335	0.17	21622	0.33	1542	0.02		
Stearic acid	C18:0	40	0.01	995628	15.18	1612757	16.98		
Oleic acid	C18:1	28	N.D	2777	0.04	3171	0.03		
Linoleic acid	C18:2	17	N.D	949337	14.48	1325465	13.96		
Arachidic acid	C20:0	977	0.13	2822	0.04	75243	0.79		
Beenic acid	C22:0	238	0.03	14350	0.22	52727	0.56		
Erucic acid	C22:1	255	N.D	892	0.01	10418	0.11		
Lignoceric acid	C24:0	25196	3.23	215806	3.29	342770	3.61		
Total		778901	100	6558053	100	9498077	100		

(N.D.) NOT detected; (M7) Seven days maceration; (U15) Ultrasound for 15 minutes; (U30) Ultrasound for 30 minutes; (U60) Ultrasound for 60 minutes; (U120) Ultrasound for 120 minutes; (U180) Ultrasound for 180 minutes; (U240) Ultrasound for 240 minutes

The extractions by ultrasound in the time periods of 15, 30, 60, 180, and 240 showed no statistical difference between them ($p < 0.05$), and when compared to the method of extraction by maceration, there was an increase in the extraction of stearic acid in these times under sonication. In 120 minutes of sonication, an increase in the content of palmitic acid was observed. The difference presented in the fatty acid profile between both extraction methods can be explained by the possible transformation of linoleic acid into palmitic, by the *β*-cleavage reaction, which transforms unsaturated fatty acids by breaking the molecules into pairs (CARVALHO et al., 2003; GUPTA, 2017; MARTÍNEZ-YUSTA; GUILLÉN, 2014).

Some barriers such as the low obtention of fatty acids in conventional extraction processes can be overcome with the use of alternative techniques, such as ultrasound-assisted extraction (KHOEI; CHEKIN, 2016). Tan et al. (2018) showed in their study that the ultrasound-assisted extraction process was efficient in obtaining fatty acids with increasing sonication time. However, prolonged periods of extraction can lead to the degradation of these compounds.

The extraction of fatty acids using ultrasound proved to be advantageous when compared to the conventional maceration technique, mainly due to the possibility of reducing the extraction time, as well as obtaining greater amounts of unsaturated fatty acids. Still, the obtention of unsaturated fatty acids arouses great interest, knowing that they play an important role in the regulation of biological systems, acting directly in inflammatory processes, antioxidants, sleep induction, hormonal regulation, blood clotting, among other fundamental processes for the maintenance of body homeostasis (FARAG et al., 1989; MURRAY et al., 2003, p. 190; RICE et al., 2011; RICHARD et al., 2008; WANG et al., 2007).

Obtaining bioactive compounds from plants of the Bougainvillea genus are of great interest since they have biological activities described in the literature as antioxidant, analgesic, anti-inflammatory, antiviral, antimicrobial, and neuroprotective (ABARCA-VARGAS; PETRICEVICH, 2018; CHAIRES -MARTÍNEZ et al., 2009; FAWAD et al., 2012).

In the determination of total phenolic compounds, a higher concentration was obtained for the extraction by the maceration technique (52.56 ± 0.52 mgEAG.g^{-1}). For the ultrasound-assisted technique, there was no statistical difference ($p < 0.05$) between the extraction times after 30 minutes, with values ranging from 36.29 ± 0.80 to 40.41 ± 1.25 mgEAG.g^{-1}.As for the determination of flavonoids, the highest content was also obtained for the maceration (5.19 ± 0.06 mgEQ.g^{-1}), while the ultrasound-assisted extraction did not show statistical difference between the extraction times after 30 minutes with values between 4.68 ± 0.01 and 4.80 ± 0.03 mgEQ.g^{-1} (Table 3).

Table 3. Phenolic compounds and total flavonoids concentrations in the ethanolic extracts of *B. spectabilis* white bracts and flowers

Codes	Phenols (mgEAG.g^{-1})	Flavonoids (mgEQ.g^{-1})
M7	52.56 ± 0.52^{a}	5.19 ± 0.06^{a}
U15	32.28 ± 0.46^{b}	4.44 ± 0.06^{b}
U30	$38.30 \pm 1.36^{c,d}$	4.80 ± 0.03^{c}
U60	40.41 ± 1.25^{c}	4.68 ± 0.01^{c}
U120	38.90 ± 0.76^{c}	4.72 ± 0.07^{c}
U180	$38.70 \pm 0.80^{c,d}$	4.68 ± 0.03^{c}
U240	36.29 ± 0.80^{d}	4.71 ± 0.03^{c}

(mgEAG.g^{-1}) milligram of gallic acid equivalent per gram of extract; (mgEQ.g^{-1}) milligram of quercetin equivalent per gram of extract; (M7) Seven days maceration; (U15) Ultrasound for 15 minutes; (U30) Ultrasound for 30 minutes; (U60) Ultrasound for 60 minutes; (U120) Ultrasound for 120 minutes; (U180) Ultrasound for 180 minutes; (U240) Ultrasound for 240 minutes. Values with different letters demonstrate statistical diffrence ($p < 0.05$).

The variations in the phenolic compound values of *B. spectabilis* bracts and flowers' ethanolic extracts were superior to other studies and are related to the type of solvent, parts of the plants, color, and species used. Chairez-Martinéz et al. (2009) used extracts from leaves of *B. spectabilis* and obtained a higher content of phenolic compounds in hydroalcoholic extracts (11.10 ± 0.08 mgEAG.g^{-1}), and 2.6 ± 0.06 mgEAG.g^{-1} for methanolic extract. Saleem et al. (2019), when evaluating the methanolic extract from flowers of the species *Bougainvillea glabra* obtained values of 26.04 ± 1.04 mgEAG.g^{-1} for phenols and 20.86 ± 1.09 mgEQ.g^{-1} for flavonoids.

For the ultrasound technique, it was observed that an extraction period of 30 minutes is enough to extract the bioactive compounds since the increase in the extraction time did not affect the contents of the analyzed compounds, presenting no statistical difference ($p < 0.05$).

The extraction time using ultrasound can vary according to the characteristics of the plant matrix, but in most cases, the time window for efficient extraction is around 30 to 60 minutes (GOLTZ et al., 2018; ŞAHIN ; ŞAMLI, 2013; WANG et al., 2008).

The potential antioxidant activity of the extracts obtained was evaluated using the DPPH and $ABTS^{+}$ radical scavenging method (Table 4).

For the DPPH radical scavenging method, the best potential antioxidant activity (EC_{50}) was observed on the ultrasound-assisted technique at 30, 60, 180, and 240 minutes (2.27 ± 0.06 to 3.08 ± 0.10 $g.g^{-1}$). For the $ABTS^{+}$ radical capture method, the extracts obtained by maceration (69.56 ± 1.20 $mM.g^{-1}$) and ultrasound for 30 minutes (67.56 ± 3.66 $mM.g^{-1}$) showed greater antioxidant capacity and presented no statistical difference by the Tukey test ($p < 0.05$) (Figure 1).

Tabela 4- Antioxidants activity of *B. spectabilis* white bracts and flowers ethanolic extracts through DPPH and ABTS radical scavenging methods

Codes	DPPH EC_{50} ($g.g^{-1}$)	ABTS (mMg^{-1})
M7	2.53 ± 0.09^{a}	69.56 ± 1.20^{a}
U15	3.08 ± 0.10^{c}	55.76 ± 2.92^{b}
U30	2.52 ± 0.14^{a,b}	67.11 ± 3.66^{a}
U60	2.30 ± 0.03^{a,b}	56.88 ± 2.52^{b}
U120	2.47 ± 0.09^{a,b}	58.04 ± 1.95^{b}
U180	2.27 ± 0.06^{b}	60.59 ± 0.53^{b}
U240	2.30 ± 0.07^{a}	53.94 ± 2.83^{b}

EC_{50} ($g.g^{-1}$) expressed in g of extract /g of DPPH; (mMg^{-1}) Values expressed in millimolar of trolox per g of extract; (M7) Seven days maceration; (U15) Ultrasound for 15 minutes; (U30) Ultrasound for 30 minutes; (U60) Ultrasound for 60 minutes; (U120) Ultrasound for 120 minutes; (U180) Ultrasound for 180 minutes; (U240) Ultrasound for 240 minutes. Values with different letters demonstrate statistical diffrence ($p < 0.05$).

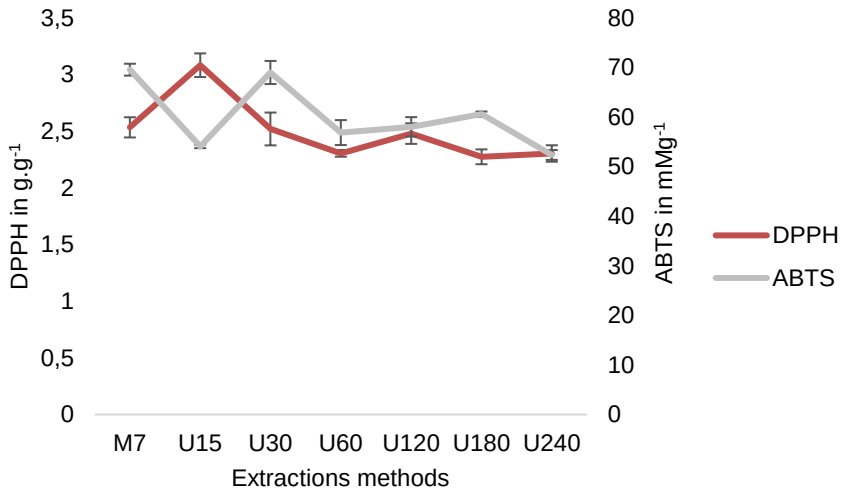

Figure 1. Antioxidants activity of *B. spectabilis* white bracts and flowers' ethanolic extracts through DPPH and ABTS radical scavenging methods. ABTS expressed in millimolar of trolox per g of extract (mMg^{-1}); DPPH expressed in g of extract /g of DPPH ($\mu g.g^{-1}$); (M7) Seven days maceration; (U15) Ultrasound for 15 minutes; (U30) Ultrasound for 30 minutes; (U60) Ultrasound for 60 minutes; (U120) Ultrasound for 120 minutes; (U180) Ultrasound for 180 minutes; (U240) Ultrasound for 240 minutes

Knowing that the DPPH radical reacts strongly with polyphenols and $ABTS^{+}$ with a greater amount of phenolic compounds, when analyzing the results obtained, the potential antioxidant activities were better for the ultrasound extraction methods, even though the maceration method shows a higher content of phenolic compounds extracted.

Thus, the study to optimize the extraction process of bioactive compounds of white bracts and flowers of *B. spectabilis* showed that the use of the ultrasound-assisted technique significantly reduces the extraction time, in addition to presenting less energy and solvent expenses in its process, as well as obtaining potential antioxidant activities similar or better than the extraction process by maceration.

Conclusion

The optimization of the extraction process by the ultrasound-assisted technique proved to be fast and effective in the extraction of bioactive compounds from the plant matrix. Among the time variables used in the ultrasound method, extraction in 30 minutes proved to be as effective as the extraction method by maceration.

Quantification by GC / MS revealed the presence of three main fatty acids in the samples (palmitic, linoleic, and stearic acid), and in the 30-minute ultrasound extraction process, there was no statistical difference when compared to the maceration method.

Although extraction by maceration presented higher levels of phenolic compounds, when analyzing

both techniques concerning their potential antioxidant activity, it was found that the extract obtained by ultrasound in 30 minutes did not show a statistical difference compared to maceration.

In general, the obtention of bioactive compounds by the ultrasound-assisted method should be explored for its ecologically effective characteristic as well as the reduction of extraction time of the bioactive compounds.

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