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Chemical composition and antimicrobial and antioxidant activity of essential oil and various plant extracts from *Prunus myrtifolia* (L.) Urb

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In this study focused on research on plants as a source of alternative and natural antimicrobial substances, the chemical composition of the essential oil from *Prunus myrtifolia* (L.) Urb. was assessed through gas chromatography coupled to mass spectrometry (GC/MS) and phytochemical screening of different extracts (aqueous, ethanolic, ethyl acetate, and hexanic) from the same plant, as well as the antimicrobial effect against the following microorganisms: *Pseudomonas aeruginosa, Salmonella* Typhimurium, *Proteus mirabilis, Klebsiella pneumoni, Escherichia coli, Enterococcus faecalis*, *Staphylococcus epidermidis, Staphylococcus aureus, Bacillus subtilis* and *Candida albicans*, through determination of minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) values, using the micro-dilution broth method. Finally, the goal was to assess the antioxidant activity of essential oil and plant extracts using the DPPH free radical method (2.2-diphenyl-1-picrylhydrazyl). The largest class of volatile compounds identified in *P. myrtifolia* oil belongs to aldehydes represented by benzaldehyde compounds. With respect to antimicrobial activity, all extracts and essential oil showed activity against the microorganisms assessed, with exception of hexanic extract. Among the extracts assessed, aqueous and ethanolic extracts were the most effective. Antioxidant activity of aqueous, ethanolic and ethyl acetate extracts was confirmed; however, antioxidant activity of essential oil and hexanic extract was not observed.

Key words: Antimicrobial activity, gas chromatography–mass spectrometry (GC/MS), native plants, chemical composition, antioxidant activity, essential oil, plant extracts.

INTRODUCTION

Brazil has the largest equatorial and humid tropical forest on the planet and, consequently, little explored extensive plant genetic diversity. With respect to the medicinal potential, only approximately 17% of plants have been studied (Pinto et al., 2002). Exploration of these plants is required, because potentially useful compounds can be lost due to the extinction of some species (Patinõ and Cuca, 2011). Due to this diversity, Brazil came to prominence in the search for potential bioactive compounds that can be used for various purposes, such

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as alternative antimicrobial products for controlling pathogens (Pupo et al., 2007) used in the pharmaceutical and food industries (Cehyan et al., 2012).The family Rosaceae comprises around 100 genera and 3000 species. Concentrated in the northern hemisphere, it is one of the leading families from an economic point of view, showing a few native species in Brazil (Souza and Lorenzi, 2005). Some species have great pharmacological and nutrition potential and are used in popular medicine for the treatment of various diseases and for the maintenance of good health. The genus *Prunus* is composed of approximately 130 species that occur in the northern, southern and southeastern regions of Brazil. Various fruits introduced and consumed in Brazil belong to this genus, such as peaches (*P. persica*), nectarines (*P. persica* var. nucipersica), plums (*P. domestica*), almonds (*P. dulcis*), and cherries (*P. avium*, *P. cerasus*) (Souza and Lorenzi, 2005).

Regarding Brazilian native species of the genus *Prunus*, the species *Prunus myrtifolia* (L.) Urban deserves attention. For being a species of wide geographic distribution, it is synonymous with *P. sphaerocarpa* Hook and *P. sellowii* Koehne (Souza and Lorenzi, 2005).

In recent years, the chemical compositions as well as the antioxidant and antimicrobial properties of plants have gained interest in the search for alternative products. Essential oils can contain from 20 to 60 (or more) diverse compounds and in the most varied concentrations (Bakkali et al., 2008). The analysis requires the application of current analytical methods and adapted instrumentation, which allows assessing the quality of essential oils and ensure the identification of their constituents. Plant extracts are targets of great interest due to the presence of secondary metabolites in their composition, which are substances used against pathogenic microorganisms, insects and herbivorous animals. In addition, they have a varied chemical composition with the presence of terpenoids, alkaloids and coumarins, which often feature antimicrobial activity (Reschke et al., 2007).With the progressive development of synthetic antimicrobial resistance, the biological properties of plant products have been studied in search of alternative products with antimicrobial action (Arya et al., 2010). In this context, essential oils and plant extracts stand out as efficient antimicrobials (Bona et al., 2010).

The search for new natural antioxidants has increased and led food, cosmetics and pharmaceutical industries to focus their searches on materials of plant origin. Plant antioxidants are very varied, but the phenolic compounds have been considered responsible for greater antioxidant capacity, being represented by flavonoids and isoflavones, tannins, lignans, and xanthones, among others (Razavi et al., 2008).The goal of this study was to determine the chemical composition of the essential oil and various plant extracts from *P. myrtifolia*, as well as their antimicrobial effect against different microorganisms, such as: *Pseudomonas aeruginosa* (ATCC 27853), *Salmonella* typhimurium (ATCC 14028), *Proteus mirabilis* (ATCC 25933), *Klebsiella pneumoniae* (ATCC 13883), *Escherichia coli* (ATCC 25922), *Enterococcus faecalis* (ATCC 19433), *Staphylococcus epidermidis* (ATCC 12228), *Staphylococcus aureus* (ATCC 25923), *Bacillus subtilis* (CCD-04) and *Candida albicans* (ATCC 10231). Finally, we aimed to assess the antioxidant activity of the essential oil and plant extracts.

MATERIALS AND METHODS

Plant material

The leaves of *P. myrtifolia* were collected in the western region of the State of Parana, Brazil (24°57' S - 53°28' W), in January and February 2013. The material was identified and incorporated into the Herbarium of the West of Parana State University (UNOP) under number 25 J. Silva, J. P. B.

The leaves collected were dried in an oven with air circulation at 40°C for 48 h and subsequently ground using a cutting mill with less than 0.42 mm granulometry. The plant material ground was stored protected from the light until its use for the production of extracts.

Obtaining aqueous extract (W)

We added 20 g of the ground plant material to a container with distilled water that was kept in a rotary shaker at 220 × g for 24 h. Subsequently, the material was filtered in filter paper (Whatman Nº 1) and centrifuged at 5000 x g for 15 min. The supernatant material was collected and the final concentration was 200 mg/ml. The extract was stored at 4°C until use.

Obtaining of organic extracts

The organic extracts were obtained according to the methodology described by Ceyhan et al. (2012) with modifications. Ethanol (95%), ethyl acetate and hexane were used as organic solvents. Starting with 10 g, the ground plant material was added to 100 ml organic solvent and placed in a rotary shaker at 220 x g for 24 h. Subsequently, it was filtered in filter paper (Whatman Nº 1) and centrifuged at 5000 × g for 15 min. The supernatant material was collected and submitted to roto-evaporation in order to remove the solvent. The extract obtained was diluted at a concentration of 150 mg/ml for ethanolic extracts (ET) and ethyl acetate (EA) and at a concentration of 6 mg/ml for hexanic extract (H) with 10% dimethyl sulfoxide (DMSO), following the proportion of its weight and volume. The extracts obtained were stored at 4°C until use.

Phytochemical screening

The main secondary metabolites were detected in accordance with the methodology developed by Matos (1997).

Essential oil extraction (EO)

Nearly 70 g of fresh leaves of *P. myrtifolia* in 600 ml distilled water were submitted to standard water steam dragging methodology for three hours using Clevenger-type equipment. The oil was collected directly with no addition of solvent and stored at 4°C.

Chemical composition analysis

The constituents of the essential oil were identified through gas chromatography coupled to mass spectrometry (GC-MS) and the determination of their Kovats retention index (KI).

GC-MS

Analysis of oil from *P. myrtifolia* was carried out using a Thermo-Finnigan GC-MS system, composed of a FOCUS GC gas chromatograph (Thermo Electron), coupled to a DSQ II mass spectrometer (Thermo Electron) and a TriPlus AS automatic injector (Thermo Electron). Chromatographic separation was performed with an HP-5ms fused silica capillary column (30 m long, 0.25 ID and 0.25 µm film; composition of 5% phenyl-95% dimethylpolysiloxane).

The temperature of the injector was 250°C. Samples and patterns of alkanes were injected using the split mode with a split ratio of 1:25. The programming of the temperature used was: 50°C maintained for 2 min; temperature rise to 180°C at a ratio of 2°C min-1; followed by an increase to 290°C at a ratio of 5°C min^{-1}. The interface between the GC and MS was maintained at 270°C and the temperature of the ionization source of the mass spectrometer was 250°C. The identification of the components was performed by comparing their retention times with those obtained in the literature (Adams, 2007) for the same compounds analyzed by means of Kovats retention index.

Microorganisms used

To perform the antimicrobial activity test of the essential oil and plant extracts from *P. myrtifolia*, we used 5 gram-negative bacteria (*Pseudomonas aeruginosa* ATCC 27853; *Salmonella* Typhimurium ATCC 14028; *Proteus mirabilis* ATCC 25933; *Klebsiella pneumoniae* ATCC 13883; and *Escherichia coli* ATCC 25922), 4 Gram-positive bacteria (*Enterococcus faecalis* ATCC 19433; *Staphylococcus epidermidis* ATCC 12228; *Staphylococcus aureus* ATCC 25923; and *Bacillus subtilis* CCCD-B005) and *Candida albicans* ATCC 1023 as yeast.

Microorganisms previously kept at -20°C were recovered in enriched medium (Brain Heart Infusion) and incubated at 36°C for 24 h. After this period, they were re-suspended in 0.9% sterile saline solution to obtain the standard inoculum at a concentration of 1×10^8 UFC/ml on the MacFarland scale. Subsequently, dilutions were performed in 0.9% sterile saline solution in order to obtain a final inoculum at a concentration of 1×10^5 UFC/ml, with the exception of *C. albicans* that was used at the final concentration of 1×10^6 UFC/ml.

Determination of minimum inhibitory concentration (MIC)

Essential oil

The MIC of the essential oil was determined using the broth microdilution method. We used 96-well plates, according to the CLSI document M31-A317 with modifications. We added 200 µl of EO from *P. myrtifolia*, at a concentration of 7000 µg/ml with Mueller-Hinton broth (MH) for bacteria and RPMI for yeast in the first well and, after homogenization, successive dilutions were held, obtaining final concentrations from 7000 to 13.67 µg/ml. Aliquots (10 µl) of microorganisms' dilution were distributed in each well containing the EO in its final dilutions. The plates were incubated at 36°C for 24 h. After turbidity was observed, each well received an aliquot of 10 µl of 0.5% triphenyl tetrazolium chloride (TTC). After three more hours of incubation at 36°C, the MIC was defined as the lowest concentration of oil in µg/mL able to prevent microbial growth (Sartoratto et al., 2004).

Plant extracts

The MIC of extracts was determined using the broth microdilution method proposed by Ayres et al. (2008) with modifications. Aliquots (10 µl) of dilution were distributed in 96-well microtitre plates, containing 150 µl of MH broth (double concentration) for bacteria and RPMI for yeast, with the previous addition of extracts. The extracts were diluted in concentrations between 100 and 0.04 mg/mL (W), between 75 and 0.035 mg/mL (ET and EA), and between 3 and 0.0012 mg/mL (H). The plates were incubated at 36°C for 24 h. After turbidity was observed, we followed the same assessment standards used for the essential oil.

Determination of the minimum bactericidal concentration (MBC)

The MBC was determined based on the methodology described by Santurio et al. (2007). From the wells in which there was no visible bacterial growth in the MIC test, prior to the addition of TTC, we withdrew an aliquot of 10 µl and inoculated it on the Mueller-Hinton agar surface. The plates were incubated for 24 h at 36°C and, after this procedure; the MBC was defined as the lowest concentration of the extract/oil able to cause the death of the inoculum. The tests of MIC and MBC were carried out in triplicate.

Distilled water, ethanol and ethyl acetate were used as negative control; gentamicin was used as positive control for bacteria; and nystatin was used for *C. albicans* (Table 1). Synthetic antimicrobials were tested at concentrations of 100 to 0.78 mg/ml.

Antioxidant activity

The measurement of the activity of free radicals scavenging (2.2-diphenyl-1-picrylhydrazyl, DPPH) was assessed as described by Scherer and Godoy (2009) and Rufino et al. (2007) with modifications. For the analysis, 0.1 ml of each dilution of samples or patterns were placed in test tubes containing 3.9 ml DPPH radical (0.2 mM) diluted with methanol and homogenized in a test tube agitator. For the negative control, we used 0.1 ml control solution (methyl alcohol, acetone and water) with 3.9 ml DPPH radical, which were homogenized. We used the commercial synthetic antioxidant butylated hydroxytoluene (BHT) following the same procedure used for the negative control. Methyl alcohol was used as whitening agent in order to calibrate the spectrophotometer (UV mini-1240, Shimadzu Co., Japan). The mixtures were incubated in the absence of light at room temperature until measurement. Subsequently, the absorbance at 515 nm was measured using a spectrophotometer and monitored every 30 min until stabilization. The tests were carried out in triplicate.

The DPPH index was calculated using the antioxidant activity equation (%) = [(Abs0 -Abs1) /Abs0] × 100, where Abs0 is the absorbance of the whitening agent and Abs1 the absorbance of the sample.

The concentrations of the samples (extracts and EO) responsible for 50% decrease in the initial activity of DPPH free radical (IC_{50}) were calculated through linear regression of the antioxidant activity.

Statistical analysis

The data obtained by calculating the DPPH index and IC_{50} were analyzed through Tukey test at 5% significance using the Sisvar software (Ferreira, 2007).

Table 1. Minimum inhibitory concentrations (MIC) and minimum bactericidal concentrations (MBC) of distilled water, organic solvents and reference antibiotics on pathogenic microorganisms.

Microorganisms	MIC/MBC (mg/ml)				
	Distilled water	Ethanol	Ethyl acetate	Gentamycin	Nystatin
P. aeruginosa ATCC 27853	Na	Na	Na	6.25/6.25	Nt
S. TyphimuriumATCC 14028	Na	Na	Na	3.125/6.25	Nt
P. mirabilis ATCC 25933	Na	Na	Na	6.25/6.25	Nt
K. pneumoniae ATCC 13883	Na	Na	Na	6.25/6.25	Nt
E. coli ATCC 25922	Na	Na	Na	6.25/6.25	Nt
E. faecalis ATCC 19433	Na	Na	Na	3.125/6.25	Nt
S. epidermidis ATCC 12228	Na	Na	Na	6.25/6.25	Nt
S. aureus ATCC 25923	Na	Na	Na	6.25/6.25	Nt
B. subtillis CCD-04	Na	Na	Na	6.25/6.25	Nt
C. albicans ATCC 10231	Na	Na	Na	Nt	6.25/6.25

Na, No activity (100<); Nt, not tested.

Table 2. Classes of secondary metabolites identified in different extracts from *Prunus myrtifolia*.

Classes of metabolites	Extracts			
	W	ET	EA	H
Tannins	+	+	-	-
Alkaloids	-	-	-	-
Coumarins	-	-	-	-
Saponins	-	+	-	-
Anthocyanins	-	-	-	-
Anthocyanidins	-	-	-	-
Flavonoids	+	+	+	-
Triterpenoids	-	+	-	-
Steroids	-	-	-	-

-, Absent; +, present; W, aqueous extract; ET, ethanolic extract; EA, ethyl acetate extract; H, hexane extract.

RESULTS AND DISCUSSION

The tests conducted for phytochemical screening (Table 2) showed that the aqueous extract had only the classes tannins and flavonoids. The ethanolic extract showed the greatest number of classes of substances: Tannins, saponins, flavonoids and terpenes. The extract with ethyl acetate solvent only showed flavonoids and the hexanic extract did not show positive results for the classes of substances tested.

It is known that the chemical constitution of Rosaceae includes especially tannins (Okuda et al., 1992), flavonoids (Harbone, 1998), triterpenes and steroids (Wallaart, 1980). The data obtained in our research agree with studies of these authors, except for the class of steroids, which was not found in any of the extracts tested.

Three compounds were found in the volatile composition of essential oil from *P. myrtifolia*, and the largest class of compounds identified belonged to aldehydes, represented by benzaldehyde, which constituted approximately 97% of the total area of the chromatogram peaks. It was followed by lower percentages of alcohol classes (3-hexen-1-ol) and esters (benzyl benzoate), with 0.07 and 0.09% total peak area, respectively (Table 3).

These data agree with those found by Ibarra-Alvarado et al. (2009), when they identified the volatile compounds of oil from *P. Serotina*, they also detected benzaldehyde as majoritary compound. It is known that benzaldehyde is one of the main components responsible for the characteristic odor of essential oils (Kerdogan-Orhan and Kartal, 2011) and it is related to various biological activities, such as antimicrobial and antifungal (Fujii et al., 2005).

The results summarized in Table 4 indicate that all

Table 3. Volatile composition of *P. myrtifolia* through GC-MS.

RT	Compound name	KI	Area (%)
5.74	3-Hexen-1-ol	852	0.07
10.22	Benzaldehyde	964	96.96
57.22	Benzyl benzoate	1759	0.09

RT, Retention time; KI, Kováts retention index calculate.

Table 4. Minimum inhibitory concentrations (MIC) and minimum bactericidal concentrations (MBC) of essential oil and different extracts of *P. myrtifolia* on pathogenic microorganisms.

Microorganisms	MIC/MBC			
	EO (µg/ml)	W (mg/ml)	ET (mg/ml)	EA (mg/ml)
P. aeruginosa ATCC 27853	3500/3500	12.5/12.5	9.38/18.75	37.5/75
S. TyphimuriumATCC 14028	1750/3500	12.5/25	18.75/37.5	150/150
P. mirabilis ATCC 25933	3500/7000	12.5/12.5	18.75/18.75	37.5/75
K. pneumoniae ATCC 13883	3500/7000	12.5/12.5	18.75/37.5	37.5/37.5
E. coli ATCC 25922	1750/7000	12.5/25	9.38/37.5	37.5/75
E. faecalis ATCC 19433	1750/7000	12.5/25	9.38/18.75	9.38/18.75
S. epidermidis ATCC 12228	3500/7000	1.56/1.56	1.18/2.35	4.69/9.38
S. aureus ATCC 25923	3500/7000	0.04/0.09	0.07/0.15	2.34/4.68
B. subtillis CCD-04	3500/7000	3.13/6.25	4.69/4.69	4.69/9.38
C. albicans ATCC 10231	3500/7000	6.25/6.25	4.69/9.37	9.38/9.38

EO, Essential oil; W, aqueous extract; ET, ethanolic extract; EA, ethyl acetate extract; Hexane extract, no activity.

extracts and the essential oil tested showed antimicrobial activity against the microorganisms assessed, with exception of the hexanic extract that showed no activity.

The essential oil had MIC values ranging from 3500 to 1750 µg/ml over the microorganisms tested. For the majority of microorganisms, the MBC was 7000 and 3500 µg/ml only for *P. aeruginosa* and *S. Typhimurium*. The activity found in the oil can be due to the presence of benzaldehyde in its composition. This compound is environmentally safe when used as an antimicrobial, considering its wide spectrum of inhibitory effect. It is also used as a bactericide and fungicide. Benzaldehyde activity has similarities to the antimicrobial activity of phenols, because it interacts with the surface of the cell and leads to cell death by disintegration of the cell membrane and release of intracellular components (Alamri et al., 2012).

Aqueous, ethanolic and ethyl acetate extracts had MIC values ranging from 0.04 to 150 mg/ml, comparable with standard antimicrobials, which ranged from 3.125 to 6.25 mg/ml. Thus, the extracts were as potent antimicrobials inhibiting the growth of microorganisms' strains as synthetic antimicrobials. With respect to gram-positive microorganisms, the same extracts had smaller MIC (0.04 to 4.69 mg/ml) compared with gentamicin (6.25 mg/ml). Regarding ethanolic extracts, *C. albicans* also had lower MIC value (4.69 mg/ml) compared to nystatin (6.25 mg/ml). When the different plant extracts (aqueous, ethanolic and ethyl acetate), were assessed regarding the gram-negative microorganisms, they had MIC ranging from 9.38 to 150 mg/ml, which were higher concentrations when compared to gentamicin concentrations (3.125 to 6.25 mg/ml). The same ratio found in the MIC was observed with respect to MBC, with values ranging from 0.09 to 150 mg/ml.

A growing number of mechanisms with inhibitory action-such as the secondary metabolites-have been assigned to active compounds present in plant extracts. Thus, the antimicrobial activity observed in aqueous, ethanolic and ethyl acetate extracts can be related to the presence of flavonoids (W, ET, and EA), tannins (ET and W), triterpenoids (ET), and saponins (ET) (Table 2), which have already proved active in different studies described in the literature (Recio et al., 1989).

It is known that the presence of flavonoids is related to most antimicrobial activities of extracts, including antibacterial (Gibbons, 2008) and antifungal potential (Cao et al., 2008). In this study, we observed greater activity against Gram-positive bacteria. This fact can result from the presence of flavonoids, agreeing with the results found by Taleb-Contini et al. (2003). The compounds commonly related to antimicrobial activity,

Table 5. DPPH average and standard deviation (% sequestration) and IC_{50} values of essential oil and different extracts from *Prunus myrtifolia* in the different concentrations tested.

Extracts/oil	Antioxidant activity (%)	IC_{50} (mg/ml)
BHT	95.85±0.07[a]	11.52±0.96[a]
W	91.27±0.67[a]	20.12±0.05[a]
ET	94.12±0.64[a]	15.43±0.0 [a]
EA	78.49±0.98[a]	14.58±0.28[a]
H	2.81±0.039[b]	186.26±0.01[b]
EO	8.69±0.97[b]	175.17±0.99 [b]

Standard error followed by the same letter in the column do not differ through Tukey test ($p<0.05$); EO, Essential oil; W, aqueous extract; ET, ethanolic extract; EA, ethyl acetate extract; H, hexane extract.

such as flavonoids, tannins, saponins, and triterpenes, generally act in the microorganism's membrane or cell wall. Flavonoids act in the bacterial cell through complexes between proteins and the cell wall causing its breakage (Taguri et al., 2004). Tannins act in micro-organisms by preventing their growth through the inhibition of nutrients transport to the cell caused by the formation of complexes between the organism and the cell wall (McSweeney et al., 2001). The action mechanism of triterpenes in microorganisms is related to the breakage of lipophilic compounds of microbial membranes (Bagamboula et al., 2004). Lastly, with respect to the saponins, they act actively in the membrane sterols (Sparg et al., 2004).

The difference between the activity found in the extracts can be attributed to the fact that the components extracted from aromatic plants with antimicrobial activity have greater solubility in solvents like ethanol, compared to hexane, for example (Cowan, 1999). Similarly, the results obtained agree with those found by Rojas et al. (2006) in which the ethanolic extract has antimicrobial activity in comparison with hexane extract, confirming the fact that the latter did not have activity at the concentration tested.

In general, aqueous and ethanolic extracts demonstrated inhibitory activity regarding all strains tested in smaller concentrations when compared to ethyl acetate extract, agreeing with Yiğit et al. (2009), who reported antimicrobial activity for ethanolic and aqueous extracts from *P. armeniaca* against Gram-negative and gram-positive bacteria and yeast as *C. albicans*.

With respect to antioxidant activity, it should be noted that the IC_{50} values are inversely related to the percentage of DPPH sequestration, since the higher the rate of sequestration, the lower IC_{50}, establishing a relationship between the values (Table 5).

The results of the antioxidant activity, expressed as IC_{50}, showed no significant difference between the synthetic antioxidant (BHT) and aqueous, ethanolic and ethyl acetate extracts; thus, they can be considered excellent antioxidants. On the other hand, there was significant difference ($p<0.5$) when compared to BHT, essential oil and hexanic extract, and no antioxidant activity was detected in these compounds. The same correlation can be observed in relation to the DPPH sequestration percentage. It is worth mentioning that the IC_{50} determines the minimum sample amount needed to reduce the DPPH free radical absorbance by 50%. However, the analysis of antioxidant activity expressed in percentages can underestimate the real potential of the samples.

According to Gao et al. (1999) phenolic compounds such as flavonoids, triterpenes and tannins are excellent antioxidants. These compounds were found in the phytochemical screening of the extracts tested (Table 2). Ethno-pharmacological data have been reported in studies conducted on the genus Prunus regarding the relationship of antioxidant activity and the presence of flavonoids (Nakatani et al., 2000). The values obtained for the DPPH sequestration index-which is similar to those obtained for BHT, aqueous and ethanolic extracts-agree with the data found by Yiğit et al. (2009).

The non-detection of antioxidant activity with respect to the essential oil may be due to the presence of its majoritary compound, that is, benzaldehyde, which features moderate to low antioxidant activity (Thanh and Hoai, 2012).

The genus *Prunus* has economic importance for the food and phytopharmaceutical industries. The literature reports more than 100 patents involving different *Prunus* species in their formulation for multiple purposes: Skin whitening (Pieroni et al., 2004); sunscreens and anti-aging skin care (Sachdeva and Katyal, 2011); essential oils used in the chemical industry (Bachheti et al., 2012); livestock food (Khanal and Subba, 2001); antimalarial treatment (Muñoz et al., 2000); asthma treatment (Karani et al., 2013); and cardiovascular disease prevention (Negishi et al., 2007).

The increased growth of antimicrobial-resistant micro-organisms commonly used is one of the most serious threats to the successful treatment of microbial diseases. Thus, the search for products that replace synthetic

antimicrobials, such as essential oils and plant extracts, is increasing primarily because they are associated with the treatment of infectious diseases (Bharathi et al., 2010). Therefore, testing new natural compounds with antimicrobial action is of great value.

Within this context, it is worth mentioning the importance of phytochemical studies, since they confirm the biological activities found. It is also worth noting the importance of preliminary studies to determine the activity of these compounds so that they can serve as the basis for subsequent studies in order to isolate different compounds with antimicrobial activity. The antioxidant activity has to be determined, since the compound has to be both antimicrobial and antioxidant.

In conclusion, the presence of flavonoids and terpenoids, among other metabolites, was detected in aqueous, ethanolic and ethyl acetate extracts. With respect to the essential oil, benzaldehyde was found as the majoritary compound. Regarding antimicrobial activity, microorganisms proved susceptible to aqueous, ethanolic and ethyl acetate extracts, and essential oil, demonstrating the antimicrobial potential of *P. myrtifolia*. With respect to antioxidant activity, the ethanolic, aqueous and ethyl acetate extracts had significant values comparable to those of synthetic antioxidant.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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