

Original Article

Chemical characterization, antioxidant and photoprotective activity of extracts from *Mimosa setosa* Benth var. *paludosa* (Fabaceae)

Caracterização química, atividade antioxidante e fotoprotetora dos extratos de *Mimosa setosa* Benth var. *paludosa* (Fabaceae)

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Abstract

Mimosa setosa Benth var. *paludosa*, popularly known as *arranha-gato*, *pom-pom*, *jurema-branca*, or *maricá-peludo*, is widely used both for the regeneration of degraded areas and in traditional medicine practices, particularly in the form of leaf infusions to promote sleep. This species, like others of its genus, produces bioactive compounds derived from secondary metabolism that may act in the prevention of diseases associated with oxidative stress and damage caused by ultraviolet radiation. This study investigated the antioxidant and photoprotective potential, as well as performed the phytochemical screening of crude leaf and root extracts of *M. setosa*. The antioxidant activity was evaluated *in vitro* using the DPPH and ABTS assays, while the photoprotective action was assessed through *in vitro* tests. Total phenolic and flavonoid contents were determined using the Folin–Ciocalteu method. Phytochemical screening was carried out by thin-layer chromatography (TLC), employing silica as the stationary phase and suitable solvent systems as the mobile phase. The results revealed the presence of phenolic compounds, flavonoids, terpenes, saponins, and steroids in the leaf extracts. The antioxidant capacity of the crude extracts was expressed as IC_{50} , with values of $6.72 \pm 0.71 \mu\text{g/mL}$ for leaves and $13.17 \pm 3.75 \mu\text{g/mL}$ for roots using the DPPH method, and $60.29 \pm 0.50 \mu\text{g/mL}$ for leaves and $36.24 \pm 0.48 \mu\text{g/mL}$ for roots using the ABTS method. Total phenolic contents were $380.16 \pm 17.78 \text{ mg GAE/g}$ for leaves and $182.33 \pm 3.78 \text{ mg GAE/g}$ for roots, whereas total flavonoid contents were $72.22 \pm 2.62 \text{ mg QE/g}$ for leaves and $182.33 \pm 3.78 \text{ mg QE/g}$ for roots. The extracts did not exhibit significant photoprotective activity, with SPF values ranging from 0.072 to 0.91 mg/mL for leaves and from 0.012 to 0.035 mg/mL for roots, falling below the minimum threshold established by ANVISA for photoprotective efficacy. These findings indicate that the crude extracts of *Mimosa setosa* possess antioxidant activity, partly attributed to their high flavonoid and phenolic contents; however, the secondary metabolites present do not confer significant photoprotective effects.

Keywords: phytochemistry, medicinal and secondary metabolites.

Resumo

Mimosa setosa Benth var. *paludosa*, popularmente conhecida como *arranha-gato*, *pom-pom*, *jurema-branca* ou *maricá-peludo*, é amplamente utilizada tanto para regeneração de áreas degradadas quanto em práticas de medicina tradicional, especialmente na forma de infusão das folhas para induzir o sono. Esta espécie, como outras de seu gênero, produz substâncias bioativas derivadas do metabolismo secundário que podem atuar na prevenção de doenças associadas ao estresse oxidativo e aos danos causados pela radiação ultravioleta. Este estudo investigou o potencial antioxidante e fotoprotetor, bem como realizou a triagem fitoquímica dos extratos brutos de folhas e raízes de *M. setosa*. A atividade antioxidante foi avaliada *in vitro* pelos métodos DPPH e ABTS; a ação fotoprotetora foi mensurada por ensaios *in vitro*. Os teores de fenólicos e flavonoides totais foram determinados pelo método de Folin–Ciocalteu. A triagem fitoquímica foi realizada por cromatografia em camada delgada (CCD), utilizando sílica como fase estacionária e solventes apropriados como fase móvel. Os resultados revelaram a presença de compostos fenólicos, flavonoides, terpenos, saponinas e esteroides nos extratos foliares. A capacidade antioxidante dos extratos brutos foi expressa como EC_{50} , com valores de $6,72 \pm 0,71 \mu\text{g/mL}$ para folhas e $13,17 \pm 3,75 \mu\text{g/mL}$ para raízes pelo método DPPH, e de $60,29 \pm 0,50 \mu\text{g/mL}$ para folhas e $36,24 \pm 0,48 \mu\text{g/mL}$ para raízes pelo método ABTS. Os teores fenólicos foram de $380,16 \pm 17,78 \text{ mg EAG/g}$ para folhas e $182,33 \pm 3,78 \text{ mg EAG/g}$ para raízes, enquanto os teores de flavonoides foram de $72,22 \pm 2,62 \text{ mg EqC/g}$ para folhas e $182,33 \pm 3,78 \text{ mg EqC/g}$ para raízes. Os extratos não apresentaram atividade fotoprotetora significativa, com valores de FPS variando entre 0,072 e 0,91 mg/mL para folhas e entre 0,012 e 0,035 mg/mL para raízes, ficando aquém do mínimo estabelecido pela ANVISA para eficácia

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fotoprotetora. Esses achados indicam que os extratos brutos de *Mimosa setosa* possuem ação antioxidante, atribuída em parte aos teores elevados de flavonoides e fenólicos, porém os metabólitos secundários presentes não conferem fotoproteção significativa.

Palavras-chave: fitoquímica, metabólitos medicinais e secundários.

1. Introduction

Secondary, or specialized, metabolites are products derived from plant metabolism that are not directly involved in developmental processes such as growth, reproduction, or propagation (Twaij and Hasan, 2022). These low molecular-weight compounds play key roles in plant defense mechanisms and environmental adaptation (Arimura and Maffei, 2017; Salam et al., 2023).

They can be classified according to their chemical structures, and more than 100,000 have already been elucidated (Martins et al., 2013; Wink, 2018; Santos et al., 2019). Because of their diverse biological activities, plants have been used by humans for thousands of years as valuable sources of medicines, perfumes, and biopesticides (Nunes et al., 2018; Yang et al., 2018; Wink, 2018; Mandal et al., 2022).

Among the activities displayed by secondary metabolites, antioxidant and photoprotective properties are particularly attractive to the pharmaceutical industry in the search for novel compounds (Orlanda and Vale, 2015; Custódio et al., 2017; Abramović et al., 2018; Orlanda and Santana, 2018). Antioxidant activity is linked to the mitigation of oxidative stress, which results from an imbalance between the excessive generation of free radicals and the insufficient rate of their removal (Barbosa et al., 2010).

Antioxidant molecules can scavenge free radicals generated by cellular metabolism or external factors, thereby preventing their harmful effects (Abrahão et al., 2010). The accumulation of these radicals promotes biomolecular oxidation and can lead to various human disorders, including cardiovascular disease, cancer, inflammation, and Alzheimer's disease (Sayre et al., 2001; Kaur et al., 2014).

Natural antioxidant compounds may also exert photoprotective effects, as they contain chromophore groups capable of absorbing radiation in the UV range (Orlanda and Vale, 2015). These photostable molecules not only protect the skin against UVA and UVB radiation, preventing damage, but also reduce premature aging and the risk of cancer (Santana et al., 2012; Martins et al., 2013). The pursuit of organic photoprotective agents offers many advantages, including a lower probability of adverse skin reactions and reduced production costs (Augustyniak et al., 2010; Tambor, 2016).

In Brazil, owing to its rich flora, approximately 55,000 plant species have been identified. However, many species remain unstudied, leaving a vast array of secondary metabolites yet to be evaluated and characterized (Costa and Marinho, 2016). Consequently, studies addressing the biological activities and chemical composition of these species are of great relevance (Pereira and Cardoso, 2012).

Mimosa setosa Benth var. *paludosa* is a weedy species of the Fabaceae family, popularly known as *arranha-gato*, *jurema-branca*, or *maricá-peludo*. It is widely distributed throughout Brazil and occurs in Paraguay, inhabiting both disturbed and forested environments (Farias et al., 2013;

Dutra and Garcia, 2014; Dutra and Morim, 2015). The species is frequently used for the regeneration of degraded areas, while its leaves are traditionally prepared as infusions to promote sleep (Nau and Sevegnani, 1997; Campos Filho, 2012; Sperandio et al., 2013; Faria et al., 2016).

Species of the genus *Mimosa* are well known for their richness in bioactive metabolites (Majeed et al., 2021). Approximately 145 active compounds have been identified within the genus, encompassing chalcones, alkaloids, flavonoids, indoles, terpenes, terpenoids, saponins, steroids, amino acids, glycosides, flavonols, phenols, lignoids, polysaccharides, lignins, and fatty acid salts and esters (Rizwan et al., 2022). Nevertheless, only a few species have been investigated in depth, underscoring the need for further studies to uncover their full medicinal potential (Rizwan et al., 2022).

Given this context, the search for new natural compounds that help combat oxidative stress and enhance skin photoprotection represents a sustainable approach to the use of native flora. Thus, this study aimed to identify the main classes of compounds and evaluate the antioxidant and photoprotective activities of leaf and root extracts of *M. setosa*.

2. Materials and Methods

2.1. Collection of plant material

Leaves and roots of *Mimosa setosa* were collected from adult individuals in a degraded forest area (3°10'46.9" S, 52°09'47.0" W), located in the municipality of Altamira, Pará State, Brazil. The collected material was stored in paper bags, washed with running water, and subsequently dried in a forced-air oven at 45 °C until a constant weight was reached (Braga et al., 2017).

2.2. Preparation of extracts

The preparation of extracts followed the methodology proposed by Jatobá et al. (2016). The dried plant material (leaves or roots) was separately ground in a knife mill (1.5 mm sieve) until a fine powder was obtained and subjected to a cold extraction process.

For the preparation of crude extracts from leaves (LCE) and roots (RCE), the powdered material was macerated with methanol at a 5:1 ratio (methanol/plant material; v/w) until exhaustion and left to stand for three days at 25 °C, protected from light. The solution was filtered through gauze and filter paper, and the solvent was removed using a rotary evaporator at 45 °C under 600 mmHg pressure. The concentrated material was then lyophilized to obtain the crude extracts.

The crude leaf extract was solubilized in methanol and subjected to liquid-liquid fractionation with the following solvents and proportions: hexane (8:2, v/v), dichloromethane (7:3), and ethyl acetate (6:4). The resulting fractions, hexane (LHF), dichloromethane (LDF), and ethyl acetate (LEAF), were concentrated using a rotary evaporator.

The extraction yield was calculated based on the dry biomass before extraction and expressed as a percentage (Yield %) (Mustafa et al., 2019) (Equation 1).

$$\text{Yield (\%)} = \left(\frac{\text{Weight of crude extract}}{\text{Total dry biomass}} \times 100 \right) \quad (1)$$

2.3. Antioxidant activity

To evaluate the antioxidant activity, two methods were employed: the 2,2-diphenyl-1-picrylhydrazyl free radical scavenging assay (DPPH) and the 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) radical cation assay (ABTS). All analyses were performed in triplicate.

The DPPH assay followed the procedure described by Brand-Williams et al. (1995), with modifications proposed by Rufino et al. (2007). Stock solutions of the crude leaf and root extracts of *M. setosa* were prepared by dissolving 5 mg of each extract in 25 mL of 20% methanol. The solutions were subsequently diluted in water to obtain the following concentrations: 2.5, 5, 7.5, 10, 12.5, 15, 17.5, and 20 µg/mL. Then, methanol and 2 mL of methanolic DPPH solution were added to each tube. The mixtures were vortexed and kept in the dark for 30 minutes. The blank was prepared under the same conditions, except that no extract was added. Absorbance was measured at 517 nm using a spectrophotometer.

For the ABTS assay, stock methanolic solutions were prepared at 10 mg/10 mL for both crude leaf and root extracts of *M. setosa* (Rufino et al., 2007). The tests were performed in triplicate. The solutions were diluted in methanol to obtain the following concentrations: 200, 300, 400, 500, 600, 700, and 800 µg/mL. Next, 3 mL of ethanolic ABTS solution was added to each sample, and the mixtures were left to stand for six minutes in the dark. Absorbance was then measured at 734 nm using a spectrophotometer. The blank was prepared as described above, using only ethanol.

The antioxidant activity was expressed as the half-maximal inhibitory concentration (IC₅₀, mean ± standard deviation), which represents the concentration required to scavenge 50% of the DPPH or ABTS radicals. IC₅₀ values were calculated using the linear regression equation obtained from the radical inhibition curves. Ascorbic acid was used as the reference standard for DPPH, and Trolox for ABTS.

2.4. Photoprotective activity

The *in vitro* photoprotective potential of the crude leaf and root extracts of *M. setosa* was determined using the method proposed by Mansur et al. (1986), with slight modifications. Each extract (10 mg) was diluted in 10 mL of analytical-grade ethanol, and the assays were conducted in triplicate.

Spectrophotometric measurements were performed in the wavelength range of 260 to 400 nm, at 5 nm intervals, to determine the absorbance of the extracts in the UVB (260 to 320 nm) and UVA (320 to 400 nm) regions. The blank (control group) was prepared under the same conditions, using ethanol instead of the samples.

The sun protection factor (SPF) was calculated according to the Equation 2 proposed by Mansur et al. (1986):

$$\text{SPF} = \text{CF} \times \sum_{290}^{320} \text{EE}(\lambda) \times I(\lambda) \times \text{Abs}(\lambda) \quad (2)$$

where: SPF = Sun Protection Factor; CF = Correction Factor; EE(λ) = erythral effect spectrum of radiation at wavelength λ; I(λ) = solar intensity at wavelength λ; and Abs(λ) = absorbance of the sample at wavelength λ.

2.5. Determination of total phenolic and total flavonoid contents

2.5.1. Total phenolic content.

The total phenolic content of the methanolic crude extracts (root and leaf) of *Mimosa setosa* was determined using the Folin-Ciocalteu (FC) method with modifications (Peres et al., 2009). Each crude extract (0.01 g) was dissolved in 10 mL of methanol. From each methanolic solution, 100 µL were transferred to a tube, followed by 1 mL of ultrapure water and 250 µL of Folin-Ciocalteu reagent. After standing for 5 min, 600 µL of 15% sodium carbonate solution (Na₂CO₃) were added, and the volume was brought to 5 mL by adding 3.1 mL of ultrapure water. The mixture was kept in the dark for 90 min, and absorbance was read at 750 nm using a spectrophotometer. Results were expressed as mg of gallic acid equivalents per g of dry weight (mg GAE/g DW) (Ferrera et al., 2016).

2.5.2. Total flavonoid content.

Total flavonoids were quantified according to Woisky (1996) and Banov et al. (2006). Each crude extract (0.01 g) was dissolved in 10 mL of methanol. Aliquots of 1,500 µL of the sample were mixed with 100 µL of 5% aluminum chloride (AlCl₃), followed by 3,400 µL of 5% acetic acid. Samples were left to stand for 30 min. The blank followed the same protocol using methanol. Absorbance was measured at 425 nm. Results were expressed as quercetin equivalents per g of dry weight (mg QE/g DW).

2.6. Thin-layer chromatography (TLC)

Qualitative phytochemical screening of crude extracts (leaf and root) and leaf fractions (hexane, dichloromethane, and ethyl acetate) was performed by thin-layer chromatography (TLC) to assess the nature of the separated compounds (Agatonovic-Kustrin et al., 2015). TLC analyses employed the following mobile phases: hexane/acetone (8:2, v/v) and ethyl acetate/methanol/water (10:1:0.35, v/v/v), following an increasing polarity gradient. The stationary phase consisted of silica gel plates (TLC silica gel 60, F254, MERCK®). Samples were applied using glass capillaries (Agatonovic-Kustrin et al., 2015).

The chromatographic plates were revealed using chemical reagents applied by spraying, including phosphovanillin solution for lignins, Dragendorff's reagent for alkaloids, anisaldehyde reagent for terpenes and steroids, NP/PEG reagent for flavonoids and phenolic acids, and 5% KOH for coumarins. The spots were subsequently visualized under UV light at 254 and 365 nm (Alves et al., 2011). The retention factor (R_f) was calculated as the

ratio between the distance traveled by each band and the distance traveled by the solvent front.

2.7. Statistical analysis

Means of treatments for antioxidant activities, total phenolic and flavonoid contents, and photoprotective responses were compared by analysis of variance (ANOVA), followed by Tukey's post hoc test at 5%. Analyses were performed in R using RStudio and the vegan package (R Core Team, 2022).

3. Results

3.1. Extraction yields

Extraction yields are presented in Table 1 for crude root and leaf extracts, as well as for their respective leaf

fractions. Among the fractions, the dichloromethane fraction showed the highest yield (Table 1).

3.2. Thin-layer chromatography

Crude extracts and fractions from *M. setosa* leaves displayed flavonoid-class compounds, evidenced by yellow coloration after reaction with NP/PEG, and suggested the presence of terpenes and saponins by purple/violet tones upon reaction with anisaldehyde (IUPAC: 4-methoxybenzaldehyde) (Reich and Schibli, 2007). No changes were observed with the other revealing reagents (Table 2).

3.3. Antioxidant activities (DPPH and ABTS)

DPPH radical-scavenging results showed that the crude leaf extract exhibited the highest antioxidant capacity ($p < 0.05$) compared with the root and with the reference standards Trolox and ascorbic acid. Root values did not differ statistically from the standards ($p > 0.05$) (Table 3).

Table 1. Yield values (%) relative to the final weight obtained for each sample of extracts and fractions of *Mimosa setosa*.

<i>Mimosa setosa</i>		
EXTRACTS	LEAF (%)	ROOT (%)
Crude extract	12.21	6.7
Hexane fraction	12.5	–
Dichloromethane fraction	30	–
Ethyl acetate fraction	5.0	–

(–) Absent.

Table 2. Classes of chemical compounds elucidated by the TLC method in crude leaf and root extracts of *Mimosa setosa*.

Classes	LCE	LHF	LDF	LEAF	RCE
Alkaloids	–	–	–	–	–
Phenolic	+++	+++	+++	+++	–
Coumarins	–	–	–	–	–
Steroids	–	–	–	–	–
Flavonoids	+++	+++	+++	+++	+
Lignins	–	–	–	–	–
Saponins	++	+	++	++	+
Terpenes	++	++	++	++	–

Note: “+” indicates the presence and “–” indicates the absence of compounds. (–) not detected; (+) low presence; (++) moderate presence; (+++) high presence. Leaf crude extract (LCE), leaf hexane fraction (LHF), leaf dichloromethane fraction (LDF), leaf ethyl acetate fraction LEAF, and root crude extract (RCE).

Table 3. Antioxidant capacity (IC₅₀ in µg/mL) of crude leaf and root extracts of *Mimosa setosa*, determined by the DPPH and ABTS free radical assays.

<i>Mimosa setosa</i>	DPPH IC ₅₀ (Mean ± SD) µg/mL	ABTS IC ₅₀ (Mean ± SD) µg/mL
LCE	6.72 ± 0.71 ^b	60.29 ± 0.50 ^b
RCE	13.17 ± 3.75 ^a	36.24 ± 0.48 ^c
Trolox (standard)	–	181.46 ± 2.33 ^a
Ascorbic acid (standard)	10.11 ± 0.66 ^a	–

The values presented in the table represent the mean and standard deviation of IC₅₀, obtained from triplicate measurements of the leaf and root extracts. Different letters in the same column indicate statistically similar activity according to the ANOVA and Tukey tests at a 5% probability level. (–) Ascorbic acid is not used as a reference value in the ABTS assay. Leaf crude extract (LCE) and root crude extract (RCE).

For the ABTS method, the root extract exhibited higher antioxidant activity than the leaf extract ($p < 0.05$), as evidenced by lower IC_{50} values, indicating greater radical scavenging capacity (Table 3). In addition, the IC_{50} values obtained were lower than those of the standards, supporting the antioxidant efficacy of the extracts (Table 3).

3.4. Flavonoids and phenolics

Total phenolic and flavonoid contents are shown in Table 4. The crude leaf extract had higher levels than the root extract flavonoids: 325.20 mg QE/g; phenolics: 380.16 mg GAE/g. The root extract contained 72.22 mg QE/g in total flavonoids and

182.33 mg GAE/g in total phenolics (Table 4). Both leaf and root displayed higher values for phenolics (Table 4).

3.5. Photoprotective activity

The photoprotective behavior of crude leaf and root extracts was evaluated spectrophotometrically at different concentrations across 260 to 400 nm. As shown in Figure 1 (panels a and b), both extracts exhibited low absorbance in the ultraviolet range (290 to 400 nm), indicating limited or negligible absorption capacity.

Sun protection factor (SPF) values of *M. setosa* (Table 5) did not indicate photoprotective potential at any tested

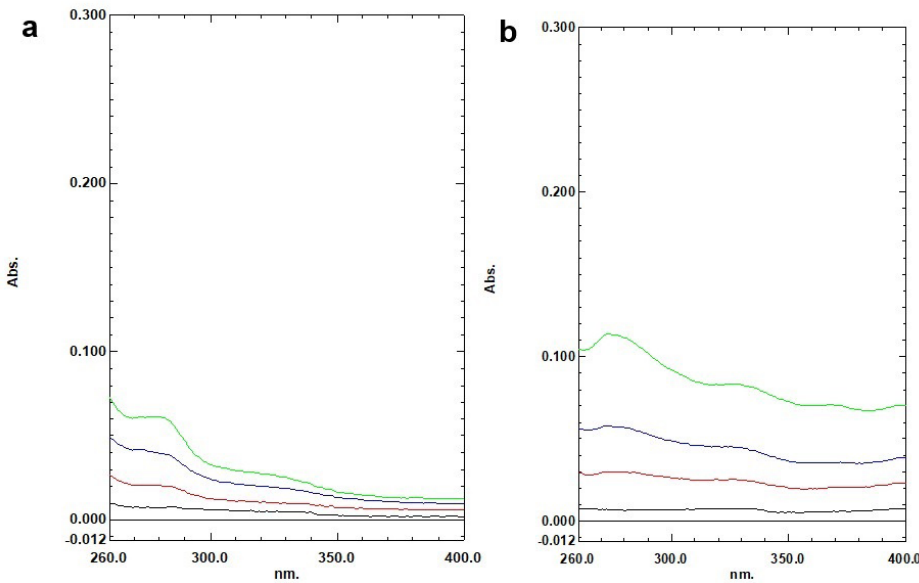


Figure 1. Spectrophotometric analysis of *Mimosa setosa* extracts at different concentrations and their absorbance in the 260 to 400 nm range. The black line represents the concentration of 5 mg/mL, the red line represents 25 mg/mL, the blue line corresponds to 50mg/mL, and the green line to 100mg/mL. (a) Root crude extract; (b) Leaf crude extract.

Table 4. Total phenolic content (mg gallic acid) and total flavonoid content (mg quercetin) of *Mimosa setosa* extracts.

Crude extracts	Flavonoids (Mean $\pm$ SD) mg QE/g	Phenolics (Mean $\pm$ SD) mg GAE/g
LCE	325.20 $\pm$ 5.95 ^a	380.16 $\pm$ 17.78 ^a
RCE	72.22 $\pm$ 2.62 ^b	182.33 $\pm$ 3.78 ^b

The values presented in the table represent the mean and standard deviation of the total flavonoid and phenolic contents, obtained from triplicate measurements of the leaf and root extracts. Identical letters in the same column indicate statistically similar activity according to the ANOVA and Tukey tests at a 5% probability level. Leaf crude extract (LCE) and root crude extract (RCE). GAE = gallic acid equivalent; QE = quercetin equivalent; SD = standard deviation.

Table 5. Sun Protection Factor (SPF) values of crude extracts of *Mimosa setosa* at different concentrations.

Plant tissues	SPF (Mean $\pm$ SD, 5 mg/mL)	SPF (Mean $\pm$ SD, 25 mg/mL)	SPF (Mean $\pm$ SD, 50 mg/mL)	SPF (Mean $\pm$ SD, 100 mg/mL)
LCE	0.072 $\pm$ 0.00 ^a	0.25 $\pm$ 0.01 ^a	0.45 $\pm$ 0.02 ^a	0.91 $\pm$ 0.03 ^a
RCE	0.035 $\pm$ 0.01 ^b	0.12 $\pm$ 0.03 ^b	0.19 $\pm$ 0.02 ^b	0.32 $\pm$ 0.04 ^b

The values presented in the table represent the mean and standard deviation of the Sun Protection Factor (SPF) at different concentrations of the crude leaf and root extracts. Identical letters in the same column indicate statistically similar activity according to the ANOVA and Tukey tests at a 5% probability level. Leaf crude extract (LCE) and root crude extract (RCE).

concentration, since all SPF values were < 6, the minimum threshold established by the Brazilian Health Regulatory Agency (Brasil, 2012) for a product to be considered photoprotective.

4. Discussion

Extraction efficiency depends on solvent polarity, since different phytochemical classes exhibit distinct solubility profiles. In this context, methanol provided satisfactory yields, suggesting adequate affinity for polar compounds such as phenolics and flavonoids (Oliveira et al., 2016). These results offer a useful basis for future fractionation and isolation of secondary metabolites from the species.

Regarding antioxidant activity, *M. setosa* crude extracts displayed expressive IC_{50} values by DPPH. These results indicate notable reducing potential particularly for leaves, which outperformed ascorbic acid in the DPPH assay. Method-specific differences likely reflect reaction particularities: DPPH primarily involves hydrogen-atom transfer, whereas ABTS responds strongly to single-electron transfer donors (Abramović et al., 2018). Thus, the phenolic- and flavonoid-rich profile of *M. setosa* supports efficient neutralization of both radicals.

Comparable findings were reported by Magalhães et al. (2018) for *Mimosa tenuiflora*, where the ethanolic bark extract showed a lower IC_{50} than other organs. Zhu et al. (2011) emphasized that antioxidant capacities vary by plant organ and depend on compound concentrations, which may explain the divergences observed here for *M. setosa*. For instance, crude extracts of *Vismia guianensis* exhibited IC_{50} values of 6.61 µg/mL (DPPH) and 8.07 µg/mL (ABTS), suggesting stronger scavenging for DPPH but confirmed antioxidant activity in both assays similar to *M. setosa* (Lins et al., 2016).

The DPPH assay measures the scavenging of the stable 2,2-diphenyl-1-picrylhydrazyl radical (Borges et al., 2017). Due to its stability and relatively slow spontaneous decay, only highly reducing substances often phenolics and aromatic acids efficiently react with this radical (Baliyan et al., 2022). In the ABTS assay, the radical cation 2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) is reduced via electrochemical or enzymatic mechanisms by antioxidant compounds (Xavier, 2016). As noted by Floegel et al. (2011), exposure of the ABTS radical to oxygen-donating compounds such as phenolics promotes its reduction. Antioxidant capacity is commonly expressed as IC_{50} the concentration that inhibits 50% of the initial DPPH or ABTS radicals; the stronger the reducing capacity, the lower the IC_{50} (Oliveira, 2015).

The high antioxidant activity observed is associated with elevated phenolic (380.16 mg GAE/g extract) and flavonoid (325.20 mg QE/g extract) contents, supporting the hypothesis that these classes contribute to the observed activity. Prior studies on *Mimosa tenuiflora* and *Mimosa pudica* report similar trends, with phenolics and flavonoids predominating in extracts with strong reducing power (Magalhães et al., 2018; Rajendran and Krishnakumar, 2010). Structurally, hydroxyl substituents on aromatic rings, especially in ortho and para positions, stabilize

the phenoxy radical, explaining the high free-radical scavenging capacity (Soobrattee et al., 2005).

Organ specific variation in metabolite content is expected and reflects environmental conditions and ecological functions of secondary metabolites. Zhu et al. (2011) highlighted that differences in phenolic and flavonoid levels may relate to protective roles in tissues exposed to oxidative stress. Leaves of *M. setosa*, being more exposed to solar radiation and climatic fluctuations, tend to accumulate antioxidants as a defense mechanism. This adaptive role of flavonoids and terpenes in response to biotic and abiotic stress has also been described in other plant species (Vicente and Boscaiu, 2018; Tetali, 2019; Al-Farsi et al., 2018; Cheng et al., 2019; Zhang et al., 2019).

Similar outcomes were reported by Gurnani et al. (2016), who observed higher phenolic than flavonoid levels in seeds of *Capsicum frutescens* L. In contrast, Buhian et al. (2016) found in *Muntingia calabura* L. that crude stem extracts had higher phenolic content than leaves, while leaf extracts had higher flavonoids both differing from *M. setosa*, where leaves surpassed roots in both metrics.

Phytochemical screening confirmed multiple classes in leaves flavonoids, terpenes, phenolics, and saponins with intense responses to revealing reagents. Roots showed milder presence of flavonoids, phenolics, and terpenes. In both organs, phenolics and flavonoids predominated, consistent with total-content determinations. Similarly, Rajendran and Krishnakumar (2010) detected glycosides, alkaloids, flavonoids, and phenolics in *Mimosa pudica* leaves by TLC.

Phenolics are among the most abundant plant metabolites and are directly involved in responses to environmental stressors, pathogen attack, nutrient imbalance, temperature extremes, and water availability (Dias et al., 2015; Tsimogiannis and Oreopoulou, 2019). They are strongly linked to the biological activities of species, with flavonoids being a particularly relevant phenolic class (Paula et al., 2010). Leaves often show more intense bands/classes and stronger antioxidant responses because they are the most exposed organs, facing herbivory and pathogens, which stimulate secondary metabolite production (Twaïj and Hasan, 2022). Flavonoids and terpenes are also abundant and play key ecological roles under biotic/abiotic stresses, with insecticidal, allelopathic, antibacterial, antioxidant, and even photoprotective activities (Perez-Vizcaino and Fraga, 2018; Vicente and Boscaiu, 2018; Tetali, 2019).

Photoprotection assays indicated that constituents present in crude leaf and root extracts do not confer effective photoprotection, as SPF values were below the ANVISA cutoff of 6 (Brasil, 2012). Low SPF values have likewise been reported elsewhere, e.g., Gomes et al. (2022) found no photoprotective activity for *Psidium guajava* L. dry extracts at low concentrations, with effects only above 200 µg/mL. Violante et al. (2009) reported no effective photoprotection among various Cerrado species, whereas Orlanda and Santana (2018) observed potential only at the highest extract concentrations for *Ocimum gratissimum* L. According to Rajasekar et al. (2024), photoprotection depends on radiant-energy absorption capacity determined by chromophore

groups, as well as concentration, λ_{max} position, and absorption bandwidth.

Despite the absence of photoprotective effect, the high phenolic levels and strong antioxidant activity suggest potential use of *M. setosa* extracts as adjuvants in cosmetic and pharmaceutical formulations e.g., as stabilizers of organic UV filters and as agents mitigating radiation-induced oxidative stress (Lorigo and Cairrão, 2019). Furthermore, mapping the classes of secondary metabolites and associated bioactivities informs their sustainable utilization and economic potential, supporting biotechnological applications (Pamphile et al., 2017).

5. Conclusion

This study demonstrates that *Mimosa setosa* Benth. var. *paludosa* exhibits high biochemical potential, characterized by the presence of phenolics, flavonoids, and terpenes in leaves and roots. These secondary metabolites are closely associated with the marked antioxidant activity observed, particularly in leaf extracts. Overall, the species can be considered an effective natural source of reducing compounds, acting through complementary hydrogen-donation and electron-transfer mechanisms.

Although no significant photoprotective activity was detected (SPF < 6), the robust antioxidant performance highlights *M. setosa* as a promising plant resource for cosmetic and pharmaceutical formulations aimed at preventing oxidative stress and skin aging. The lack of photoprotection shifts its functional value toward synergistic antioxidant systems and as a natural stabilizing additive for oxidation-sensitive products.

Scientifically, this work expands knowledge on the chemistry and bioactivity of underexplored *Mimosa* species. We recommend continued investigation employing advanced chromatographic and spectroscopic approaches (HPLC-DAD, LC-MS, NMR) to identify major constituents and elucidate molecular mechanisms of antioxidant action. Such advances may consolidate *M. setosa* var. *paludosa* as a valuable source of natural biomolecules with biotechnological, pharmacological, and environmental applications.

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Data Availability Statement

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request. All data supporting the findings of this study are included in the article.

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