

Chemistry

Evaluation of the antioxidant and photoprotective potential of plants from the Atlantic Forest of southern Bahia

Avaliação do potencial antioxidante e fotoprotetor de plantas da Mata Atlântica do sul da Bahia

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ABSTRACT

Study of the many therapeutic and pharmacological actions of plants that result from the benefits of secondary metabolites, is an important part of appreciating and understanding the rich biodiversity of Brazilian biomes. The objective of this study was to investigate the antioxidant and photoprotective properties of extracts optimized through simple centroid planning of a solvent mixture of *Copaifera lucens* Dwyer, *Schnella angulosa* (Vogel) Wunderlin, and *Miconia albicans* (SW) Triana collected from the Atlantic Forest of southern Bahia, Brazil. The results showed that all extracts were dependent on DPPH concentration for antioxidant activity. *S. angulosa* (EC_{50} 21.37 $\mu\text{g mL}^{-1}$ and IAA 1.40) and (EC_{50} 37.21 $\mu\text{g mL}^{-1}$ and IAA 0.80) were considered active. However, photoprotective activity was not significant ($FPS < 6$).

Keywords: Vegetable extract; Photoprotection; Antioxidant, Optimized extraction; Phenolics

RESUMO

As ações terapêuticas e farmacológicas de plantas, decorrente da ação de metabolitos secundários é uma estratégia importante para a valorização e conhecimento da rica biodiversidade do Brasil. Neste contexto, este estudo objetivou a investigação por métodos *in vitro* da ação antioxidante e fotoprotetora de extratos otimizados por planejamentos de mistura de solventes de *Copaifera lucens* Dwyer, *Schnella angulosa* (Vogel) Wunderlin e *Miconia albicans* (SW) Triana coletadas na Mata Atlântica o Sul da Bahia, Brasil. Os resultados mostraram a dependência da concentração de DPPH na atividade antioxidante e extrato de *S. angulosa* (CE_{50} 21,37 $\mu\text{g mL}^{-1}$ e IAA 1,40) e (CE_{50} 37,21 $\mu\text{g mL}^{-1}$ e IAA 0,80) foram considerados ativos. Entretanto, a atividade fotoprotetora não foram significativas ($FPS < 6$).

Palavras-chave: Extrato vegetal; Fotoproteção; Antioxidante, Extração otimizada; Fenólicos

1 INTRODUCTION

From prehistoric times, plants have been utilized by man as a source of food and medicine. Medicinal effects are most often due to the presence of secondary metabolites that act beneficially to prevent or treat pathologies, such as chronic diseases, cardiovascular diseases, and inflammatory diseases, among others (PEREIRA; CARDOSO; GRAÇAS, 2012).

Phenolics are bioactive compounds that act as antioxidants by sequestering free radicals (FRs), and are associated with numerous properties, such as skin photoprotection (KUMAR *et al.*, 2019). Simple phenols, phenolic acids, flavonoids, coumarins, stilbenes, hydrolysable and condensed tannins, lignans, and lignins constitute the majority of bioactive constituents found in plants that provide a variety of health benefits (KUMAR; GOEL, 2019; MARKHAM *et al.*, 1998).

An excess of FRs in the body has been associated with a number of deleterious effects, including DNA damage and pathologies such as melanoma and non-melanoma skin cancer (CÓRDOVA; NAVAS, 2000; D'ORAZIO *et al.*, 2013; NAUSER; GEBICKI, 2019). The use of antioxidants extracted from plants has been gaining prominence due to their health benefits and their ability to regenerate or prevent this oxidative damage (ALVES *et al.*, 2010; FLORES *et al.*, 2021). In addition, plant extracts rich in antioxidants have become an alternative for photoprotection and prevention of diseases associated with exposure to solar radiation (DE SOUZA; CAMPOS; PACKER, 2013).

The diversity of plants found in the Atlantic Forest of southern Bahia is a potential source of bioactive molecules waiting to be discovered. In addition, the region contains communities with immeasurable traditional knowledge about the use of plants, that has been passed down from generation to generation (TABARELLI *et al.*, 2005; BRASIL 2020). In order to contribute to the body of knowledge regarding species in this ecosystem, the current study proposes an investigation of the photoprotective and antioxidant actions of *Copaifera lucens* Dwyer, *Schnella angulosa* (Vogel) Wunderlin, and *Miconia albicans* (SW.) Triana.

C. lucens and *S. angulosa* are species belonging to the Fabaceae family that are endemic in the Atlantic Forest. *C. lucens* is a 15- to 25-meter-tall tree that is found in Bahia, Espírito Santo, Minas Gerais, Rio de Janeiro, and São Paulo, where it is popularly known as red *copaífera*, *pau-óleo*, *pau-óleo-copaíba*, or *copaiba* (COSTA, 2021). The oil extracted from the trunk is used in cosmetics, phytotherapeutics, and for the healing of wounds, skin diseases, and throat and lung infections (PIERI; MUSSI; MOREIRA, 2009). Studies also report activity against promastigote forms of *Leishmania amazonenses* (SANTOS *et al.*, 2008). However, despite the popular use and presence of bioactive chemical compounds within the plant oils, studies of the leaves were not found in the literature, making this an important object of this study.

S. angulosa, formerly known as *Bauhinia angulosa* Vogel, is a native shrub that occurs in Bahia, Espírito Santo, Minas Gerais, Rio de Janeiro, São Paulo, Paraná, and Santa Catarina, where it is known as *escada de macaco* and traditionally used in the treatment of diarrhea, kidney disorders, and back pain (SOUZA, 2020). Although there is no record of any studies on its pharmacological properties or chemical composition, the presence of flavonoids, lactones, terpenoids, steroids, triterpenes, tannins, and quinones is noted in the genus (SILVA; FILHO, 2002).

Another studied plant, *M. albicans*, is a tree of the Melastomaceae family found in tropical regions. In Brazil it is known as *canela-de-velho*, and its leaves and aerial parts are widely consumed in teas prepared by infusion and in ointments for the treatment of rheumatism, arthrosis, and arthritis (CORRÊA *et al.*, 2021; QUINTANS-JÚNIOR *et al.*, 2020). Some authors attribute the antioxidant, anti-inflammatory, and analgesic properties to the presence of pentacyclic triterpenes, phenolic acids, and flavonoids (CORRÊA *et al.*, 2021; CUNHA *et al.*, 2017; LIMA *et al.*, 2018).

The search for species with pharmacological properties that thrive in the Atlantic Forest is an important part of understanding and valuing the local biodiversity, which has been reduced over the years as a result of advancing destruction of vegetation. The objective of this study was to investigate the antioxidant and photoprotective

action, through in vitro methods, of extracts optimized through mixture design from *Copaifera lucens* Dwyer, *Schnella angulosa* (Vogel) Wunderlin, and *Miconia albicans* (SW) Triana, collected from the Atlantic Forest of Southern Bahia, Brazil.

2 MATERIAL AND METHODS

Specimens of *Copaifera lucens* Dwyer, *Schnella angulosa* (Vogel) Wunderlin, and *Miconia albicans* (S.W) Triana were randomly collected from a fragment of the Atlantic Forest in Southern Bahia, Brazil, located within the Pau Brasil Ecological Station, near BR 367, Porto Seguro, BA (16°23'2.5"S and 39°10'57.7"W) in November 2018 and February 2019. After botanical identification, exsiccates from each species were documented in the Professor Geraldo C.P. Pinto (GCPP) Herbarium at the Federal University of Southern Bahia, Sosígenes Costa Campus (Porto Seguro) under the numbers GCPP 01055, GCPP 0919, and GCPP 1110, respectively.

The plant material was dried in a forced air circulation oven for 72 hours at 30°C and then ground in a knife mill and stored in dark plastic bags until the beginning of the extractive processes.

2.1 Preparation of Extracts through Mixture Design

The extraction process was carried out using solvent mixtures through an experimental design of SCD Simplex mixtures, as described by Scheffe (1963), with the response factor associated with the dependent and independent variables using the Statistica® 6.0 (StatSoft) software. In mixture of 60 mL of methanol (M), ethyl acetate (A), and trichloromethane (T) solvents, 5 g of the stem, leaf, and root of *C. lucens*, *S. angulosa*, and *M. albicans* (Table 1) were macerated two consecutive times for 48 hours in a closed system. Subsequently, they were evaporated under reduced pressure following the solvent specifications at a temperature of 50°C, with the phenolic contents used as a response factor.

Table 1 – Solvent mixture design from the extraction process

Treatment	M	A	T
	mL		
3	0	0	60
5	30	0	30
4	30	30	0
6	0	30	30
2	0	60	0
7	20	20	20
7	20	20	20
7	20	20	20
1	60	0	0

Source: Authors (2022)

Methanol (**M**), Ethyl Acetate (**A**), and Trichloromethane (**T**)

The percentage yield of dry mass (R%) for the sample was calculated using equation 1:

$$R\% = (me/mv) \times 100 \quad (1)$$

Where, me = extract mass and mv = plant mass

2.2 Total Phenolics (TP)

Values for total phenolics (TP) were obtained using an adapted version of the *Folin Ciocalteu* method (NEVES *et al.*, (2009) (MATHEW; SUBRAMANIAN, 2014). Twenty μL of the sample, 200 μL of distilled water, 20 μL of *Folin-Ciocalteu* reagent, and 60 μL of sodium carbonate were placed in a 96-well microplate. The microplate was then incubated for one hour at 45°C, after which a reading was taken using a Thermo Scientific Multiskan Go UV-VIS spectrophotometer at 760 nm. The blank sample consisted of 200 μL water, 20 μL *Folin-Ciocalteu*, 20 μL ethanol, and 60 μL sodium carbonate solution, and the control for samples and standards consisted of 280 μL distilled water with 20 μL of the sample

or reference. Concentrations between 5.46 - 350 $\mu\text{g mL}^{-1}$ of gallic acid were used as the reference for phenolics.

The phenolic content was determined by interpolating the absorbance of the samples onto a calibration curve constructed with the standard data and expressed as milligrams of gallic acid equivalent per gram of extract (mg g^{-1}). Validation of the method was performed using the detection limits (DL) and quantification limits (QL). The DL represents the lowest concentration of the substance that can be detected and was obtained from the equation: $\text{DL} = 3.3 \times s/S$, where "s" is the mean standard deviation of the blank sample, and "S" is the angular coefficient of the line. The QL, which represents the lowest concentration of the substance that can be measured, was calculated using the equation: $\text{QL} = 10 \times s/S$ (RIBANI *et al.*, 2004).

2.3 Antioxidant Activity (AA)

The antioxidant capacity was determined through the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical sequestration method. In this evaluation, the change from violet to yellow color results from the reduction of the DPPH radical, monitored at 517 nm in a UV/visible spectrophotometer after incubation in the dark (FURLAN *et al.*, 2015).

To perform this evaluation, 50 μL of methanol and 500 $\mu\text{g mL}^{-1}$ of extracts were placed in the 96-well microplate for the initial concentration and carrying out of the serial dilution. Following this, 250 μL of the methanolic solution of DPPH (40 $\mu\text{g mL}^{-1}$) was added. After incubation for 20 min, a reading was taken using a Thermo Scientific Multiskan Go UV-VIS spectrophotometer at 517 nm. All assays were performed in triplicate and gallic acid was the reference substance.

The blank sample was prepared using 50 μL of the extract solution and 250 μL of methanol. For the verification of 100% activity, 250 μL of DPPH solution and 50 μL of methanol were used. From the absorbance values, the antioxidant activity percentage was calculated using equation 2:

$$AA\% = (Abs2 - Abs1) / Abs2 \times 100 \quad (2)$$

Where AA% is the percentage of antioxidant activity, Abs1 is the absorbance of sample reduced by its respective blank, and Abs2 is the absorbance of the negative control (methanol and DPPH).

The average effective concentration (EC_{50}) necessary to inhibit 50% of the DPPH and the Antioxidant Activity Index (AAI) were obtained from the straight-line equation of the graph of sequestration percentage versus concentration, Equation 3 (SCHERER; GODOY, 2009):

$$IAA = \frac{[DPPH]}{EC_{50}} \quad (3)$$

Where IAA = Antioxidant Activity Index, [DPPH] = DPPH concentration, and CE_{50} = effective concentration.

2.4 Photoprotective activity

The in vitro photoprotective activity was determined using the extracts of higher phenolic content from the mixture planning. The extracts of each plant with methanol concentrations of 1000, 500, 250, 125, and 62.5 $\mu\text{g mL}^{-1}$ (methodology adapted from Mansur *et al.*, 1986) were measured in a Multiskan Go model Thermo Science® spectrophotometer at 290 to 320 nm (UVB) with quartz cuvettes having an optical path of 1 cm (PEREIRA *et al.*, 2020).

Methanol was used as a blank sample and its absorbance values were used in equation 4 to obtain the Sun Protection Factor (SPF) values. The entire experiment was performed in triplicate and efficacy was considered to be an $SPF \geq 6$ (DAL PRÁ *et al.*, 2017; ORLANDA; VALE, 2015; ROSA *et al.*, 2008).

$$SPF = FC \cdot 290 \sum_{320} EE(\lambda) \cdot I(\lambda) \cdot ABS(\lambda) \quad (3)$$

Where FC = correction factor (equal to 10); \sum = wavelength interval from 290 to 320 nm, EE (λ) = erythematogenous effect of radiation of wavelength λ ; I (λ) = intensity of sunlight at wavelength λ ; and Abs (λ) = absorbance reading at wavelength (λ). The EE x 1 values used were: 0.0150 (290 nm), 0.0817 (295 nm), 0.2874 (300 nm), 0.3278 (305 nm), 0.1864 (310 nm), 0.0839 (315 nm), and 0.0180 (320 nm) (MANSUR *et al.*, 1986).

2.5 Statistical Analysis

The statistical analyses were performed using analysis of variance (ANOVA) with a sample number equal to 3 and a confidence index compared to the student's-t table value of 2.92 at the 95% confidence level. All results were represented as the mean \pm standard deviation (SD) and data treatment and graphing were performed using Excel and Statistic 6.0.

3 RESULTS AND DISCUSSION

3.1 Mixture Planning and Total Phenolics

The first step in studying the chemical constituents of plants is extraction and the selection of the extracting solvent plays a crucial role in the procedure (AZMIR *et al.*, 2013; LIMA *et al.*, 2020). Statistical mixture planning makes it possible to select the solvent most efficient for acquiring the enriched fraction of the metabolites of interest, by decreasing the number of experiments and identifying the synergistic and antagonistic effects of the solvents used. In addition, it decreases the amount of volatile solvents and residues that are normally observed in solid-liquid or liquid-liquid processes (NOVAES *et al.*, 2018).

The efficiency of Centroid Simplex Planning through quantitative responses is described in Table 2. The extraction yields ranged from 0.02% to 35.5% (m/m) dry mass. However, this response is considered imprecise and of low quality, since the high yield does not guarantee

that the extracted constituents are responsible for the activity attributed to the plant (OLIVEIRA, 2014) and losses may also occur through the incorrect handling of the physical procedures adopted.

In this planning scheme, the total phenolics (TP), used as the optimized response parameter, varied from 7.97 to 120.96 mg g⁻¹, 23.30 to 145.30 mg g⁻¹, 11.20 to 260.90 mg g⁻¹, and 11.89 to 100.20 mg g⁻¹ in the treatments of *C. lucens* leaves (CLL), *S. angulosa* root (SAR), *S. angulosa* stem (SAS), and *M. albicans* stem (MAS), respectively. Experiments 7 (CLL), 1 (SAR), 5 (SAS), and 4 (MAS) produced the highest amount of phenolics.

Table 2 – Total Phenolic (TP) content for each plan, with dried samples

Plan	Plant							
	CLL		SAR		SAS		MAS	
	Y (%)	TP ± SD (mg g ⁻¹)	Y (%)	TP ± SD (mg g ⁻¹)	Y (%)	TP ± SD (mg g ⁻¹)	Y (%)	TP ± SD (mg g ⁻¹)
3	5.00	20.87 ± 2.99	2.68	23.30 ± 1.06	0.02	11.20 ± 1.5	0.14	16.52 ± 0.80
5	4.87	58.43 ± 3.82	19.32	25.61 ± 0.14	2.38	260.90 ± 5.51	9.17	93.08 ± 0.68
4	35.56	82.42 ± 6.13	4.63	56.54 ± 1.46	7.49	106.64 ± 3.11	1.06	100.20 ± 1.49
6	5.10	7.97 ± 0.59	1.85	24.93 ± 0.83	0.43	45.87 ± 1.85	4.82	11.89 ± 0.53
2	3.19	23.57 ± 0.53	1.00	26.63 ± 0.43	0.44	104.72 ± 1.08	0.14	17.45 ± 0.59
7	20.02	120.96 ± 1.37	6.25	57.45 ± 1.30	6.41	231.17 ± 1.95	5.53	35.95 ± 0.85
1	7.99	118.25 ± 1.53	8.43	145.30 ± 1.27	6.74	91.62 ± 1.88	9.53	26.34 ± 2.86

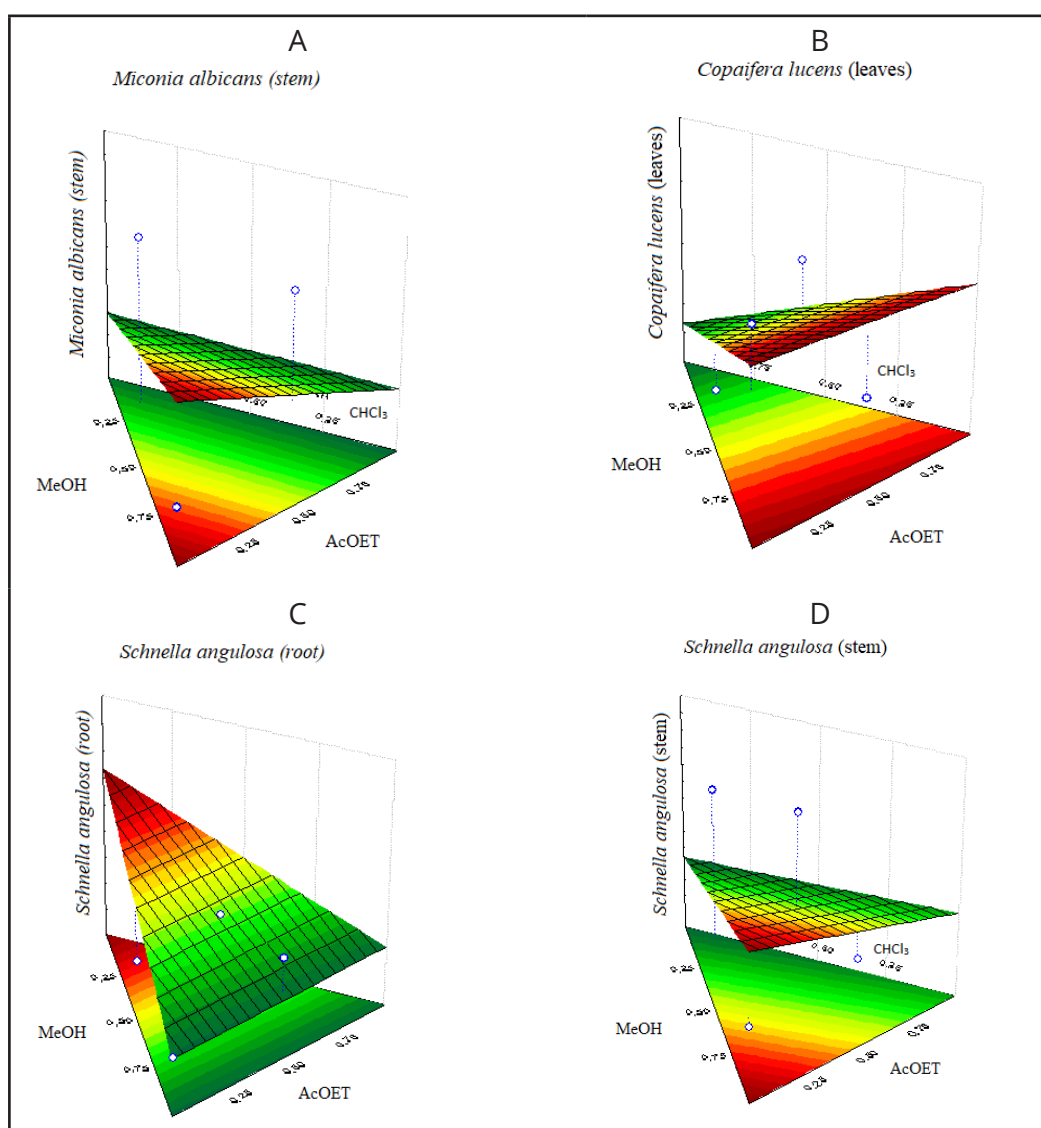
Source: Authors (2022)

Y = Yield; TP = Total Phenolics; DP = Standard Deviation; CLL= *Copaifera lucens* Leaves; SAR = *Schnella angulosa* Root; SAS = *Schnella angulosa* Stem; MAS = *Miconia albicans* Stem

The experimental design of the response factor on an equilateral triangle contour surface as a function of solvent composition is shown in Figure 1. And all phenolic values had variations for both plants and solvents.

That said, the binary mixture of methanol and ethyl acetate for *M. albicans* was most efficient in extracting TP. Figure 1A displays the maximization of constituents with a contour surface and reddish coloration, for maximum contents of $100.20 \pm 1.49 \text{ mg g}^{-1}$, indicating the contribution of a synergistic effect between solvents.

Figure 1 – Contour surface for the experimental design of the simplex-centroid linear mathematical model as a function of solvent composition for *M. albicans* (A), *C. lucens* (B), and *S. angulosa* (C and D)



Source: Authors (2022)

* MeOH= Methanol; CHCl₃= Trichloromethane; AcOET= Ethyl acetate

In the *C. lucens* extraction (Figure 1B), methanol and the ternary mixture of methanol, trichloromethane, and ethyl acetate ranged from TP = 118 to 120 mg g⁻¹. Methanol is more advantageous as it eliminates the use of organochlorines and reduces the possibility of contamination through the use of three solvents. For *S. angulosa* root (Figure 1C), methanol was most efficient (FT= 145.30 ± 1.27 mg g⁻¹). In contrast, the binary and ternary mixtures are more effective for the stem (Figure 1D), with optimized responses of TP = 260.90 ± 5.51 mg g⁻¹ (ethyl ac. and trichloromethane) and FT= 231.17 ± 1.95 mg g⁻¹ (methanol, ethyl ac., and trichloromethane), demonstrating the differences in chemical composition of the plant. For all experiments, the DL and QL values of the TP content were 0.11 and 0.36 mg g⁻¹, respectively, confirming the analytical method as a suitable quantitative response for analysis of the plants. Statistically, the results were significant at a confidence level of (95%), n = 3, CV (coefficient of variation) ≤ 0.1% and with values ≥ 2.92 (student's-t value).

Despite the scarcity of information in the literature regarding chemical and pharmacological approaches to *S. angulosa* and *C. Lucens*, comparison with previous studies on *M. albicans* shows that the extraction of phenolics using the binary mixture of ethyl acetate and methanol was higher than the values published by Pieroni *et al.*, (2011) and Corrêa *et al.*, (2021) for the methanolic extract from the leaves (70.04 mg g⁻¹) and fruit of *M. Albicans* (43.68 ± 0.50 mg g⁻¹). Obviously disregarding the experimental differences, mixture planning proves to be more efficient.

Binary solvent mixtures are always recommended for extractions of compounds having different solubility and hydrophobicity, where using a single universal solvent for plant extraction is practically unfeasible (ALMEIDA *et al.*, 2018; BRUM; ARRUDA; REGITANO-D'ARCE, 2009; ZORN *et al.*, 2017).

Experimental planning makes it possible to find the best conditions for extraction of bioactive constituents, while minimizing the use of unnecessary, often toxic solvents (HANDA *et al.*, 2016; SCHEFFE, 1963). Many authors used the procedure for more efficient extractions in natural matrices. For example, Dal Prá *et al.* (2017) optimized

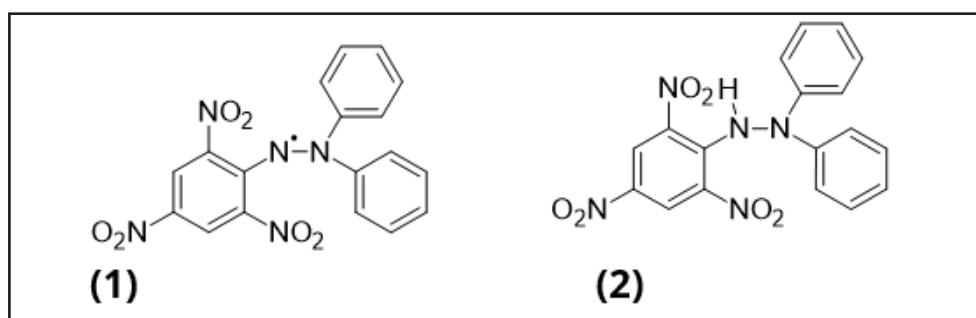
palm extraction conditions in an ultrasound-assisted methodology, and the results demonstrate antioxidant compounds and extracts with a high solar protection factor.

3.2 Antioxidant Evaluation

The use of antioxidants is an efficient approach to reduce damage caused by excess FR, which are often associated with a number of pathologies. In a broad definition, antioxidants are any substance that, regardless of concentration, effectively inhibits or sequesters FR, either in the initiation or propagation stage of the oxidative process.

Antioxidant substances, such as phenolic compounds, help to slow the oxidation rate by inhibition of chemical species or by complexation of metals (PEREIRA; CARDOSO; GRAÇAS, 2012; VIEIRA *et al.*, 2015). Various methods are described in the literature to evaluate the antioxidant capacity of organic substrates (ALVES *et al.*, 2010). However, the stable free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) method is most commonly applied. The test is based on measuring the ability of the DPPH radical (purple) to be reduced by antioxidant substances to hydrazine (pale yellow) (ALVES *et al.*, 2007) (Figure 2).

Figure 2 – Chemical structure of **(1)** Unreduced and **(2)** Reduced DPPH



Source: Authors (2022)

The antioxidant activity determined by the DPPH method for the highest TP experiments are shown in Table 3. The efficiency was measured by the effective concentration (EC_{50}) and the antioxidant activity index (AAI) (SCHERER; GODOY, 2009).

Table 3 – Antioxidant activity of highest TP experiments from mixture planning

Plants	Part	Plan	EC ₅₀ (µg mL ⁻¹)	AAI	TP* (mg g ⁻¹)
<i>C. lucens</i>	Leaves	7	37.21	0.80	121.0
	Root	1	164.0	0.18	145.3
<i>S. angulosa</i>	Stem	5	21.37	1.40	260.9
<i>M. albicans</i>	Stem	4	159.8	0.25	100.2

Source: Authors (2022)

EC₅₀ = Average Effective Concentration; AAI = Antioxidant Activity Index; TP = Total Phenolics

The efficiency of the antioxidant activity is associated with the direct relationship between EC₅₀ and AAI, where a lower value for EC₅₀ and a higher value for AAI indicate effective radical sequestration activity (EVERTON *et al.*, 2021).

Although all of the extracts exhibited the concentration-dependent sequestration capacity, only plan 5 from *S. angulosa* stem (EC₅₀ 21.37 and AAI 1.40) and plan 7 from *C. lucens* leaves (EC₅₀ 37.21 and AAI 0.80) were intensely active, as per the classification described by Reynertson *et al.*, (2008) with an EC₅₀ < 50 µg mL. However, according to the index proposed by Scherer and Godoy (2009), they are classified as strong (1.0 < AAI > 2.0) and moderate (0.5 < AAI > 1.0), respectively. The remaining experiments were considered to have low activity, according to the indices.

High antioxidant potential may be associated with TP, except for *S. angulosa* root (Plan 1), which, despite having a high level of phenolics, curiously showed low antioxidant activity. According to Rockenbach (2008), factors such as solvent polarity and pH can influence this activity, along with the structural characteristics of phenolics.

Phenolic substances are considered to be antioxidant agents (LACHMAN *et al.*, 2010; ANUNCIACÃO *et al.*, 2020; GUEDES *et al.*, 2017), and are often times reported as important for health maintenance because they are associated with numerous

functional health benefits, such as reducing the risk of heart disease, diabetes, and cancer (KUMAR; GOEL, 2019; PALMA-DURAN *et al.*, 2017; SOLEAS *et al.*, 2002). Similarly, there is experimental evidence describing their important role in reducing the onset of degenerative diseases such as Alzheimer's disease (MORAIS *et al.*, 2013) and as a skin photoprotection agent (VIEIRA *et al.*, 2015).

3.3 Evaluation of Photoprotective Activity

Interest in the use of natural products to protect the skin from the deleterious effects of sun exposure is on the rise (CARVALHO *et al.*, 2020). Antioxidants present in plants that can absorb solar radiation have become a target of investigation as a potential agent in anti-aging and photoprotection of the skin (ORLANDA; VALE, 2015).

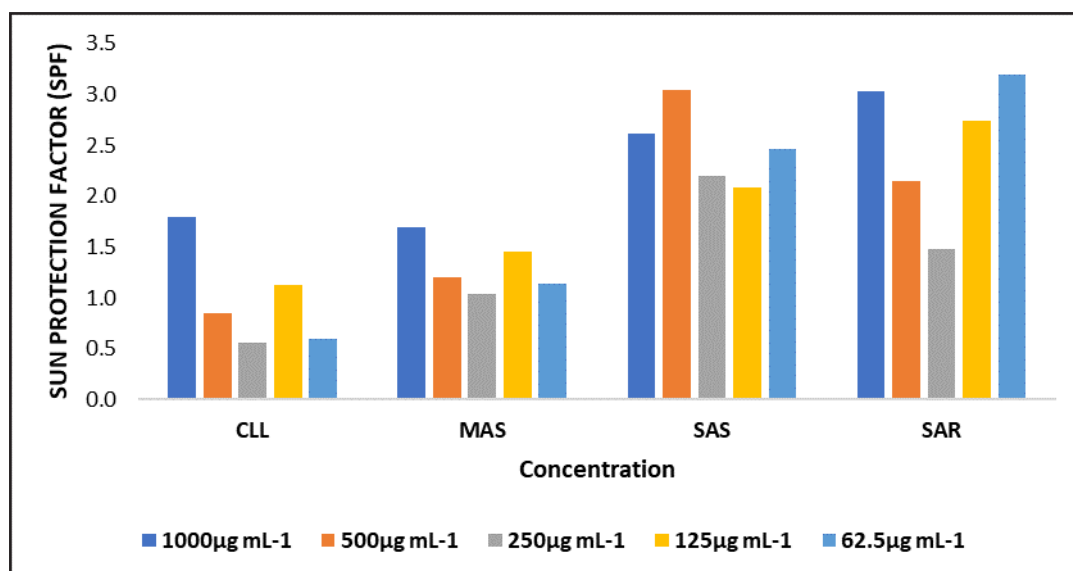
The extracts with the highest concentrations of phenolics were evaluated through the *in vitro* Sun Protection Factor (SPF) test. This method relates the absorbance of the analyzed substance to the erythemal effects and intensity of light at wavelengths in the UVB region between 290 and 320 nm (DAL PRÁ *et al.*, 2017; ORLANDA; VALE, 2015; ROSA *et al.*, 2008).

The calculated SPF values are presented in Figure 3. SAS (Plan 5) and SAR (Plan 1) had the highest SPF values, which may be a result of the concentration of phenolics and antioxidants in the extracts (BARNI; CECHINEL FILHO; COUTO, 2009). However, the Brazilian National Health Surveillance Agency (ANVISA) (RDC No. 30 from July 1, 2012) regulates that cosmetic products or formulations to be used as sunscreens must present an SPF ≥ 6 (six) (BRASIL, 2012). All of extracts displayed weak activity, making them unsuitable for use as sunscreens. However, they should be investigated for use in formulations where they may act synergistically with synthetic photoprotectants.

Numerous studies have reported a photoprotective activity from Brazilian plant extracts, either in isolation or as a supplement to sunscreens. For example, the phytochemical screening of *Pisidium guajava* L. (guava) showed that, despite a low SPF, the association of the extract with a filter based on 2-ethylhexyl- methoxycinnamate

(EHMC), resulted in a significant increase in SPF values (MOTA *et al.*, 2019). In another study conducted by (MOTA *et al.*, 2020), a formulation containing *Nephelium lappaceum* L extract and 7.5% EHMC improved the photoprotective capacity of the sunscreen by 134%. It is also worth noting that the formulations also reduced the toxicity of the synthetic agent as well as the production cost of the sunscreen (MOTA *et al.*, 2019).

Figure 3 – Sun Protection Factor plot of *C. lucens*, *M. albicans*, and *S. angulosa* extracts (CLL = *C. lucens* leaves; MAS = *M. albicans* stem; SAS = *S. angulosa* stem; SAR= *S. angulosa* root)



Source: Authors (2022)

The composition and concentration of phytochemicals are factors that influence the absorption of ultraviolet radiation and the photoprotective action of natural products. Even at low concentration, weakly active constituents can contribute to SPF values, and it is necessary to evaluate purified active components (PEREIRA *et al.*, 2020). In general, compounds such as flavonoids, benzophenones, and phenolic acids have chromophore groups that absorb UV radiation at different wavelengths and are associated with high SPF values (RODRIGUES *et al.*, 2021).

4 CONCLUSION

The results of this study contribute to the knowledge on the pharmacological potential of plants from a region of enormous biodiversity, surrounded by communities that traditionally use them in rituals and medicine to cure and prevent diseases. Although all of the extracts showed a dependence on the DPPH radical concentration, with *S. angulosa* (EC_{50} 21.37 $\mu\text{g mL}^{-1}$ and AAI 1.40) and (EC_{50} 37.21 $\mu\text{g mL}^{-1}$ and AAI 0.80) considered active antioxidants, the photoprotective activity was insignificant ($\text{SPF} < 6$).

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