

Evaluation of the Photoprotective Potential and Antioxidant Activity of Plant Extracts from Restinga de Jurubatiba (Brazil)

Gabriel dos Reis^a, Bruna Magalhães Pereira Gomes^a, Lucas Oliveira Rodrigues^b, Leandro Rocha^b, Carlos Augusto de Freitas Peregrino^a, Emeli Moura de Araújo¹⁵*^a

^aDepartamento de Tecnologia Farmacêutica (MTC), Faculdade de Farmácia, Universidade Federal Fluminense, Niterói, Brazil; ^bPrograma de Pós-Graduação em Ciências Aplicadas a Produtos para Saúde, Faculdade de Farmácia, Universidade Federal Fluminense, Niterói, RJ, Brazil;

*Corresponding author: emeliaraujo@id.uff.br

Dermatological damage caused by exposure to solar radiation drives the search for photoprotective formulations that can reach a wider public. Most of the current chemical ultraviolet (UV) filter formulations causes issues such as photoirritation, photosensitization, harmful hormonal activity to pregnant women, and interference in marine ecosystems caused by the buildup of these compounds. Plant metabolites such as polyphenols and flavonoids have structural characteristics similar to the current UV filters. The study focused on the possibility of employing plant extracts collected in the Restinga de Jurubatiba National Park (Rio de Janeiro, Brazil) as UV photoprotectors. *In vitro* sun protection factor (SPF) was determined by spectrophotometric method, and antioxidant activity by 2,2-diphenyl-1-picrylhydrazyl (DPPH) method. Of the analyzed extracts, 7 species reached SPF >1 in 5% solutions, and a mixture containing 6 of them reached the highest SPF of 9.9. They were: *Baccharis reticularia*, *Eugenia pruniformis*, *Myrsine rubra*, *Myrsine parvifolia*, *Eugenia sulcata*, *Myrcia amazonica*, and *Myrcia vittoriana*. In the antioxidant activity test, *Myrsine rubra* had the highest activity, higher than ascorbic acid and Trolox standards. The bibliometric analysis results, there is a recognized knowledge gap regarding studying the photoprotective and antioxidant properties of these native plants from the Brazilian restinga.

Keywords: Sunscreen; sun protection factor; bibliometric analysis; natural UV filter; spectrophotometry.

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Introduction

The sun can emit electromagnetic radiation in wavelengths from infrared, visible, and ultraviolet (UV) ranges. UV radiation can be further subdivided into UVA (320 - 400 nm), UVB (290 - 320 nm) and UVC (220-290 nm) (1). Despite corresponding to 6.8% of the solar radiation that falls on the Earth's surface (2), UV radiation, is the most harmful to human health because it has a short wavelength and, therefore, higher energy (3). UVA and UVB radiation are the main responsible for causing epidermal damage because UVC radiation rarely reaches the Earth surface due to the protective effect of the ozone layer. UVB is about 900 times more reactive on the skin than UVA causing erythema, pigment darkening skin (4,5), epidermal hyperplasia, photoaging, immunosuppression, exacerbation of photodermatoses, deoxyribonucleic acid (DNA) damage, protein degradation and mutations caused by reactive oxygen species (ROS) including free radicals, and photocarcinogenesis (6), which is a major public health problem (7). Among the ways to protect against damage caused by ultraviolet radiation on the skin are clothing, hats or staying in shaded areas, and sunscreens (8). Sunscreens are formulations designed to protect the skin from UVB and UVA radiation, minimizing potential skin damage (9), and usually comprise of one or more UV

filters. UV filters are classified into organic and inorganic ones based on chemical composition and mechanism of action (6).

Organic substances can absorb UV radiation and release it as heat or light in wavelengths (λ) greater than 400 nm. These UV filters are characterized by structures containing disubstituted benzene rings in the ortho or para position, with electron-donating and electron-attracting groups. The main representatives of this class are derivatives of p-aminobenzoic acid (PABA), cinnamates, salicylates, camphor derivatives, octocrylene, phenylbenzimidazole sulfonic acid, triazones, benzophenones, methyl anthranilate, and butyl methoxydibenzoylmethane (10).

Organic UV filters are widely used in various photoprotective formulations due to the variety of substances available. However, studies point out some disadvantages of using organic UV filters, including photodegradation of these substances leading to the formation of free radicals. They can also cause photoirritation and photo-sensitization in some individuals, posing risks to users (11). Another important factor is the absorption of these substances into circulation through the skin, leading to their deposition in adipose tissues, liver, brain, and breast milk, potentially interfering with neuronal synapses and endocrine signaling, making their use not recommended for pregnant and lactating women

(12). Additionally, these filters are bioaccumulated in aquatic environments, which can enter humans through food, raising environmental concerns (13).

Ideally, sunscreen should also have antioxidant activity. Studies show that formulations containing substances capable of scavenging free radicals help delay dermal aging and reduce photodamage caused by UV radiation (14) mediated by ROS generated after UV exposure (6).

The use of natural products is gaining attention from researchers due to their greater safety than synthetics. The presence of flavonoids in plant extracts contributes significantly to photoprotective action due to their UV radiation absorbance characteristics, and their anti-inflammatory and immunomodulatory properties (15). Studies also showed that extracts containing significant concentrations of polyphenols, alkaloids, tannins, and coumarins exhibited photoprotective activity and were important antioxidants (16, 17). So the development of products with large number of components of plant origin has been highlighted in the cosmetic field (6).

Restinga de Jurubatiba is part of the Jurubatiba Restinga National Park, located in the northern part of Rio de Janeiro state (Brazil), covering Macaé, Carapebus, and Quissamã. It is a Quaternary sandy plain, with an area of 146.8 km², containing many habitats and floristic richness. Its soil mainly consists of sand deposits, with shrub vegetation, and the most prevalent family among the listed species is *Myrtaceae* (18). It is a location with abundant natural resources of interest for research.

This work aims to study the photoprotective and antioxidant potential of plant species available in the Jurubatiba Restinga National Park to incorporate them into photoprotective formulations and justify these studies' importance through bibliometric analysis.

Experimental section

Bibliometric analysis

A longitudinal retrospective study was carried out using the Web of Science (WoS) (Clarivate Analytics), Scopus (Elsevier) and *Biblioteca Virtual em Saúde* - or BVS - (which encompasses Pubmed, Lilacs and Medline) databases. The methodological approach evaluated the scientific production published until March 30 2024, and indexed by the previously mentioned databases, referring only to the 13 plant species studied. The search was conducted across all databases within Web of Science (Web of Science Core Collection, Derwent Innovations Index, Grants Index, KCI-Koren Journal Index, Preprint Citation Index, ProQuest Dissertations & Theses Citation Index, SciELO Citation Index). The search query utilized the "topic" field, encompassing title, abstract, keywords, and keywords plus, with the following string: "TS= ("Annona acutiflora" OR "Baccharis reticularia" OR "Eugenia pruniformis" OR "Eugenia sulcata" OR "Erythroxylum ovalifolium" OR "Myrcia amazonica" OR "Myrcia vittoriana" OR "Myrciaria floribunda" OR "Myrsine parvifolia" OR "Myrsine rubra" OR "Ocotea

elegans" OR "Ocotea nonata" OR "Ocotea pulchella") AND TS=(photoprotecti* OR antioxidant)".

In Scopus, the search was conducted across all fields using the following string: "(ALL ("Annona acutiflora") OR ALL ("Baccharis reticularia") OR ALL ("Eugenia pruniformis") OR ALL ("Eugenia sulcata") OR ALL ("Erythroxylum ovalifolium") OR ALL ("Myrcia amazonica") OR ALL ("Myrcia vittoriana") OR ALL ("Myrciaria floribunda") OR ALL ("Myrsine parvifolia") OR ALL ("Myrsine rubra") OR ALL ("Ocotea elegans") OR ALL ("Ocotea nonata") OR ALL ("Ocotea pulchella")) AND (ALL("photoprotecti*") OR ALL ("antioxidant"))".

In BVS, the same terms were searched within the title, abstract, and subject using the string: "(("Annona acutiflora" OR "Baccharis reticularia" OR "Eugenia pruniformis" OR "Eugenia sulcata" OR "Erythroxylum ovalifolium" OR "Myrcia amazonica" OR "Myrcia vittoriana" OR "Myrciaria floribunda" OR "Myrsine parvifolia" OR "Myrsine rubra" OR "Ocotea elegans" OR "Ocotea nonata" OR "Ocotea pulchella") AND ("photoprotecti*" OR "antioxidant"))".

Reagents and plant extracts

Ethanol 96% (v/v) was provided by J.T.Baker® (Brazil); 2,2-diphenyl-1-picrylhydrazyl (DPPH), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), n-Hexane and ascorbic acid were provided by SIGMA-ALDRICH (Brazil); Ethyl acetate was provided by TEDIA (Brazil); Methanol P.A. and n-butanol were provided by VETEC (Brazil).

The plant extracts of the species *Annona acutiflora*, *Baccharis reticularia*, *Eugenia pruniformis*, *Eugenia sulcata*, *Erythroxylum ovalifolium*, *Myrcia amazonica*, *Myrcia vittoriana*, *Myrciaria floribunda*, *Myrsine parvifolia*, *Myrsine rubra*, *Ocotea elegans*, *Ocotea nonata*, and *Ocotea pulchella* were provided by Laboratório de Tecnologia de Produtos Naturais/Universidade Federal Fluminense (LTPN/UFF) previously collected in the Jurubatiba Restinga National Park using authorization 13659-9 ICMBio/SISBIO and SisGen AFCF243.

The extracts were prepared from the leaves after drying and crushing. The crude extracts were obtained by exhaustive maceration using ethanol and subsequent solvent evaporation using a rotary evaporator. The crude extract was then fractionated by liquid-liquid extraction using organic solvents with increasing polarities, following the sequence: hexane, dichloromethane, ethyl acetate and n-butanol (19). Each fraction obtained was then dried by freeze-drying.

In vitro SPF determination

The *in vitro* sun protection factor (SPF) of the plant extracts from the 13 species was determined according to the method described by Mansur (20). Absorbance values

for each plant extract at 5 or 10% (w/v) in ethanol 96% (v/v), were determined in triplicate at a final concentration of 0.2 µL/mL in UV/Vis spectrophotometer (Shimadzu, UV 1601) (21).

Also, the SPF of mixtures of the extracts at 5% (w/v) in ethanol 96% (v/v) was analyzed. The dilutions were made to mimic the action of those extracts in the same formulation, with their concentrations being added together.

The absorbance was determined by scanning in the range of 220 to 400 nm, speed of 240 nm/min, low slit opening (1.0) and sampling interval of 1 nm (21).

The SPF determination, equation 1 and the correlation between the erythemogenic effect (EE) and the radiation intensity (I) at each wavelength ($EE \times I$) were adjusted according to Sayre (22).

$$SPF_{\text{spectrophotometric}} = CF \times \sum_{290}^{320} EE(\lambda) \times I(\lambda) \times ABS(\lambda) \quad (1)$$

This equation calculates the photometric SPF by summing the variables, which are the absorbances (ABS) found from 290 to 320 nm at intervals of 5 nm, multiplied by the erythral effect and the intensity of solar radiation at each wavelength ($EE \times I$), which are constants (Table 1), and the correction factor (CF) concerning the *in vivo* test, which is always 10 (23).

Table 1. Values of $EE \times I$ described by SAYRE (1979) (22).

Wavelength (λ) (nm)	EE(λ) x I(λ)
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0839
320	0.0180
Total	1.0000

Evaluation of Antioxidant Activity

The method used for evaluating antioxidant activity was the one described by Mensor and colleagues (24), which uses the stable free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH). The assay was performed only in 6 species that showed the most promising SPF value when mixed. In addition to the plant species, the same DPPH test was conducted on 2 other standards: Ascorbic acid (or Vitamin C), known as a potent natural and non-toxic antioxidant, and Trolox, widely used as a standard in conventional antioxidant activity tests (25).

In this test, the samples were diluted in methanol P.A. in triplicate at 11 concentrations ranging from 1.12 µg/mL to 53.57 µg/mL and mixed with a 3mM DPPH solution in the same solvent. A control solution containing methanol and DPPH at the same concentration as the others and a blank solution containing the extract and methanol were

prepared. The mixtures were incubated in the dark for 30 minutes and read on a UV/Vis spectrophotometer (PG Instruments, T80) at 515 nm. The percentage of antioxidant activity at each concentration was calculated based on mathematical equation 2, which relates the absorbances found in the sample, blank, and control.

$$AA\% = 100 - \left\{ \frac{[(ABS_{\text{sample}} - ABS_{\text{blank}}) \times 100]}{ABS_{\text{control}}} \right\} \quad (2)$$

ABS sample is the absorbance obtained from the solution with the sample and DPPH; ABS blank is the absorbance obtained from the solution with the sample without DPPH; ABS control is the absorbance obtained from the solution without the sample but with DPPH; and AA% is the antioxidant activity found in percentage. By linear regression of the antioxidant activity percentages, the EC_{50} of each plant extract was calculated, representing the concentration (in µg/mL) required for each extract to achieve 50% of antioxidant activity.

Statistical Analysis

All data were analyzed by ANOVA or Tukey test using the GraphPad Prism 8.0 and $p < 0.05$ was considered to be statistically significant.

Results and Discussion

Bibliometric analysis

In the Web of Science platform, 17 articles were found, 300 articles were found in the Scopus platform, and 11 were found in the BVS platform. The titles and abstracts of these works were manually evaluated. Of these, only 4 articles, indexed in all three platforms, assess the antioxidant potential of *Myrciaria floribunda* (26, 27, 28, 29), whereas two, indexed in the Scopus platform, evaluate the antioxidant potential of *Eugenia pruniformis* (30, 31). In other words, a few partially similar studies were identified.

In vitro SPF Determination

The plant species fractioned extracts were available in different fractions such as dichloromethane, ethyl acetate, n-butanol, hexane, and crude extract.

The chemical ability to absorb UV radiation is affected by the nature of the solvent (21), therefore it is important to use the same solvent to compare the results of the *in vitro* SPF assay. The hexane fractions were not tested due to their insolubility in 96% ethanol. According to Pinto (2005) (32), the insolubility of this fraction in this solvent can be explained by its composition mainly of fatty acids, steroids, and hydrocarbons, substances with an apolar character. The total solubility of the extract in 96% ethanol was a crucial criterion adopted because the presence of solids can cause the spreading of

electromagnetic radiation, leading to interference in the obtained result (33).

Different fractions of three species were initially tested (Table 2)

Table 2. *In vitro* SPF results of different fractions of the plant species at 5% (w/v) in ethanol.

Plants species	Fraction	SPF ¹ X ² ± S ³
<i>Annona acutiflora</i>	Ethyl Acetate	0.82 ± 0.04
	n-Butanol	0.66 ± 0.01
<i>Eugenia pruniformis</i>	Ethyl Acetate	1.43 ± 0.12
	Crude	0.75 ± 0.01
<i>Myrcia amazonica</i>	Ethyl Acetate	1.19 ± 0.10
	Dichlorometane	0.27 ± 0.06

¹Sun protection factor, ²Average (n=3), ³Standard deviation.

Observing the SPF results obtained in the analysis of *Eugenia pruniformis* (SPF of crude extract = 0.75; SPF of ethyl acetate fractionated extract = 1.43), *Myrcia amazonica* (SPF of dichloromethane fractionated extract = 0.27; SPF of ethyl acetate fractionated extract = 1.19), and *Annona acutiflora* (SPF of n-butanol fractionated extract = 0.66; SPF of ethyl acetate fractionated extract = 0.82), it is visible that the ethyl acetate fractions showed higher values than all the other fractions studied. This is due to substances with greater photoprotective action, such as flavonoids and others with phenolic structures, concentrating in this fraction (32). This first observation established that all the other 10 species be analyzed using only the ethyl acetate fraction (Table 3).

Table 3. *In vitro* SPF results of the ethyl acetate fraction of the plant species at 5% (w/v) in ethanol.

Plants species	Fraction	SPF ¹ X ² ± S ³
<i>Baccharis reticularia</i>	Ethyl Acetate	1.48 ± 0.10
<i>Eugenia pruniformis</i>	Ethyl Acetate	1.43 ± 0.12
<i>Myrsine rubra</i>	Ethyl Acetate	1.40 ± 0.15
<i>Myrsine parvifolia</i>	Ethyl Acetate	1.30 ± 0.02
<i>Eugenia sulcata</i>	Ethyl Acetate	1.20 ± 0.06
<i>Myrcia amazonica</i>	Ethyl Acetate	1.19 ± 0.10
<i>Myrcia vittoriana</i>	Ethyl Acetate	1.05 ± 0.04
<i>Erythroxylum ovalifolium</i>	Ethyl Acetate	0.98 ± 0.06
<i>Annona acutiflora</i>	Ethyl Acetate	0.82 ± 0.04
<i>Myrciaria floribunda</i>	Ethyl Acetate	0.80 ± 0.02
<i>Ocotea pulchella</i>	Ethyl Acetate	0.76 ± 0.03
<i>Ocotea nonata</i>	Ethyl Acetate	0.58 ± 0.01
<i>Ocotea elegans</i>	Ethyl Acetate	0.42 ± 0.04

¹Sun protection factor, ²Average (n=3), ³Standard deviation.

As the bibliometric analysis showed, there are no published articles on the photoprotective potential of the 13 species under study, but there are other studies with other plant species.

Zini (2012) (34) analyzed the *in vitro* SPF *Mikania glomerata* extract produced at Universidade Federal de Juiz de Fora-Brazil (UFJF) and 5 commercial extracts from the species *Mikania glomerata*, *Matricaria chamomilla*, *Ginkgo biloba*, *Aquilella millefolium* and *Aesculus hippocastanum*. The highest SPF found was of the *Mikania glomerata* UFJF extract (SPF = 2.72) which was a 100% (1g dry substance/mL) solution. For the commercial extracts, the highest SPF was from the *Matricaria chamomilla* extract (SPF = 1.51), while the other 4 extracts did not reach SPF > 1. In another study performed by Wróblewska (2019) (35), it was found that a gel made with bamboo extract at 10% obtained SPF < 6.0.

Baccharis reticularia extract at 5% showed the highest SPF value (Table 3), however the extract demonstrated to be insoluble at 10% ethanol. Therefore, only the extracts of *Eugenia pruniformis* and *Myrsine rubra* had their SPF determined in this concentration (Table 4). Both extracts fractionated in ethyl acetate presented the second and third highest SPF at 5% respectively (Table 3).

Table 4. *In vitro* SPF results of plant species`ethyl acetate fraction at 10% (w/v) in ethanol.

Plant species	SPF ¹ X ² ± S ³
<i>Eugenia pruniformis</i>	2.60 ± 0.04
<i>Myrsine rubra</i>	2.84 ± 0.03

¹Sun protection factor, ²Average (n=3), ³Standard deviation.

It is described in the literature that the increase in concentration does not always increase proportionally the SPF result (36), but comparing the results from tables 3 and 4, it was possible to observe that the SPF increased proportionally.

Chiari-Andreo et al. (2020) (37) presented a review regarding the photoprotection properties of phytochemical and secondary metabolites from plants, such as *Hamamelis virginiana* L., *Matricaria recutita* L., *Aesculus hippocastanum* L., *Rhamnus purshiana* DC., and *Cinnamomum zeylanicum* at 3% and 10%, all the extracts showed low SPF values. The results revealed photoprotective activity superiority with the combination of natural products compared with single constituents (37).

In Araújo et al. study (2020) (21), two compounds derived from cashew nut oil (*Anacardium occidentale* L.), identified as V34 and V35, showed low SPF values in individual solutions at 5% (2.9 and 4.2, respectively). However, when tested in a mixture at a concentration of 10%, they showed an SPF of 8.5, which is higher than the sum of the two individual values, demonstrating an increase in action due to the compatibility of the molecules.

Based on this information and the fact that sunscreens usually comprised of one or more UV filters (6), also considering that the minimum SPF value for a formulation to be considered photoprotective, according to Brazil's National Health Surveillance Agency (ANVISA) is SPF 6 (9), extracts that showed SPF values

> 1 at 5% (w/v) *Baccharis reticularia* (SPF = 1.48), *Eugenia pruniformis* (SPF = 1.43), *Myrsine rubra* (SPF = 1.40), *Myrsine parvifolia* (SPF = 1.30), *Eugenia sulcata* (SPF = 1.20), *Myrcia amazonica* (SPF = 1.19), and *Myrcia vittoriana* (SPF = 1.05) were selected to compose mixtures to test the possibility of a synergistic action between them.

The sum of the concentrations (5% or 10%) of the species should correspond to a maximum of 30% (v/w) to make it viable to include the quantity in a sunscreen formulation. Results are presented in Table 5.

Table 5. *In vitro* SPF results of mixtures' plant extracts in ethyl acetate.

M ¹	Plant species	Concentration	SPF ² X ³ ± S ⁴
1	<i>Eugenia pruniformis</i>	5%	3.23 ± 0.07
	<i>Myrcia amazonica</i>	5%	
	<i>Myrsine parvifolia</i>	5%	
2	<i>Eugenia pruniformis</i>	5%	4.31 ± 0.08
	<i>Myrsine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
3	<i>Eugenia pruniformis</i>	5%	5.32 ± 0.06
	<i>Myrsine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrsine parvifolia</i>	5%	
4	<i>Eugenia pruniformis</i>	5%	6.14 ± 0.21
	<i>Myrsine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrcia amazonica</i>	5%	
5	<i>Eugenia pruniformis</i>	10%	4.12 ± 0.05
	<i>Myrsine rubra</i>	10%	
6	<i>Eugenia pruniformis</i>	5 %	6.95 ± 0.16
	<i>Myrsine rubra</i>	10%	
	<i>Baccharis reticularia</i>	5 %	
	<i>Myrcia amazonica</i>	5 %	
7	<i>Eugenia pruniformis</i>	5%	8.50 ± 0.24
	<i>Myrcine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrcia amazonica</i>	5%	
	<i>Myrcine parviflora</i>	5%	
	<i>Eugenia sulcata</i>	5%	
8	<i>Eugenia pruniformis</i>	5%	6.17 ± 0.21
	<i>Myrcine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrcia amazonica</i>	5%	
	<i>Myrcine parviflora</i>	5%	
9	<i>Eugenia pruniformis</i>	10%	9.79 ± 0.09
	<i>Myrsine rubra</i>	10%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrcia amazonica</i>	5%	
10	<i>Eugenia pruniformis</i>	5%	9.99 ± 0.21
	<i>Myrsine rubra</i>	5%	
	<i>Baccharis reticularia</i>	5%	
	<i>Myrcia amazonica</i>	5%	
	<i>Myrsine parvifolia</i>	5%	
	<i>Myrcia vittoriana</i>	5%	

¹Mixture, ²Sun protection factor, ³Average (n=3), ⁴Standard deviation.

Mixture 2, that comprised *Eugenia pruniformis*, *Myrsine rubra* and *Baccharis reticularia* at 5% did not show a significant difference (p < 0.05) between mixture 5 that comprised only *Eugenia pruniformis*, *Myrsine rubra*, both

at 10%, demonstrating that doubling the concentration did not increase the SPF, and the addition of the species had a beneficial effect on the SPF of the mixture 2.

The difference between Mixture 2 and 4 was the addition of *Myrcia amazonica* at 5%, and the SPF value was considerably higher and significant different (p < 0.05) at mixture 4 (SPF 6.14), but there was no significant difference comparing mixture 4 with mixture 8, that had the addition of 5% of *Myrcine parviflora*, demonstrating that the addition of this extract was no benefit to SPF value. Formulations 4 (20%), 6 (25%) and 8 (25%) reached the minimum SPF determined by ANVISA (SPF > 6) formulation to be considered photoprotective (9).

In order to further increase the SPF value, the mixture 9 was prepared with *Eugenia pruniformis* (10%), *Myrsine rubra* (10%), *Baccharis reticularia* (5%) and *Myrcia amazonica* (5%), and the SPF value (9.79) was significantly higher (p < 0.05) than mixture 4, but there was no significant difference comparing to mixture 10, that had 6 species at 5%, that had the addition of *Myrsine parvifolia* and *Myrcia vittoriana*. These formulations comprised 30% of the extracts.

When the SPF values of mixtures 7, 9, and 10 are compared with the simple sum of the individual SPF values of each extract (Tables 3 and 4), it is found that Mixture 7 (Sum of individual SPF values = 8; SPF of mixture = 8.5) had a synergistic effect increasing by 6.25% the SPF value, while mixture 10 (Sum of individual SPF values = 8.11) and Mixture 11 (Sum of individual SPF values = 7.85) showed an increase in the SPF value of 20.72% and increase of 27.26%, respectively, representing a hyperchromic effect, showing a synergy among the extracts.

Evaluation of antioxidant activity

The plants chosen to perform this assay were those that were part of mixtures 9 and 10 that presented higher SPF values (Table 5), namely *Eugenia pruniformis*, *Myrsine rubra*, *Myrcia amazonica*, *Myrsine parvifolia*, *Baccharis reticularia*, and *Myrcia vittoriana*. The results obtained were as EC₅₀ (concentration required to obtain a 50% antioxidant effect) in Table 6.

Table 6. Results of the antioxidant activity evaluation.

Sample	□□ ₅₀ (µg/mL)
<i>Myrsine rubra</i>	2.36
<i>Myrcia vittoriana</i>	3.38
<i>Myrcia amazonica</i>	3.91
Ascorbic Acid	4.09
<i>Myrsine parvifolia</i>	4.27
<i>Eugenia pruniformis</i>	4.47
Trolox	5.30
<i>Baccharis reticularia</i>	75.44

When analyzing the results based on EC₅₀ values, it is understood that the lower the value, the higher the antioxidant capacity of the sample, as it requires lower concentrations to reduce the DPPH reagent by 50%.

According to the values found in Table 6, the species *Myrsine rubra* ($EC_{50} = 2.36$), *Myrcia vittoriana* ($EC_{50} = 3.38$), and *Myrcia amazonica* ($EC_{50} = 3.91$) showed statistically significant difference ($p < 0.05$) and better antioxidant activities than the standard Ascorbic acid ($EC_{50} = 4.09$) and Trolox ($EC_{50} = 5.30$), with *Myrsine rubra* having the most significant result, with an EC_{50} 42% lower than ascorbic acid.

The species *Myrsine parvifolia* ($EC_{50} = 4.27$) and *Eugenia pruniformis* ($EC_{50} = 4.47$) showed statistically significant difference ($p < 0.05$) and lower antioxidant activity than ascorbic acid ($EC_{50} = 4.09$), but higher than Trolox ($EC_{50} = 5.30$). The results of these species show excellent antioxidant properties. Albuquerque et al. (2016) (30) found a high content of flavonoids and antioxidant activity in *Eugenia pruniformis*, besides potential wound healing effects. Meanwhile, *Myrsine parvifolia* showed promising results in neutralizing venom from *Bathrops* sp. species, with several flavonoids like myricetin, myricetin-3-O- β -arabinopyranoside, quercetin, and kaempferol identified in its ethyl acetate fractionated extract (15).

The use of antioxidants topically has several effects, such as anti-aging, anti-inflammatory, photoprotective, and ultraviolet radiation (UVR) immunosuppression preventative. In theory, the addition of antioxidants to sunscreen formulations could protect against the UVR-induced oxidative stress that could not be prevented by UVR absorption or reflectance, therefore may prevent cancer-causing mutations mediated by ROS (38). Examples of natural antioxidants include ubiquinone or idebenone (CoQ10), alpha-lipoic acid, L-ascorbic acid (vitamin C), and alpha-tocopherol (vitamin E) (38).

The study by Rebêlo et al. (2014) (39), which evaluated the antioxidant activity of *Atemoia* extracts (*Annona cherimola* Mill. and *A. squamosa* L.) using the same DPPH assay and expressed in EC_{50} ($\mu\text{g/mL}$), the highest antioxidant activity was found in the ethanolic extract (EtE) of stems (EC_{50} EtE = 10.44), the second highest in the methanolic extract (ME) of leaves (EC_{50} ME = 29.87), and the lowest in the hexane extract (HE) of leaves (EC_{50} HE = 186.00). According to data from this research, if antioxidant activity is related to the total phenolic content measured in mgEqAG/g (EtE = 207.80; ME = 75.80; and HE = 74.47), and total flavonoids measured in mgEqC/g (EtE = 151.96; ME = 46.86; and HE = 14.90), it is possible to note that the total flavonoid content may have a greater influence on DPPH antioxidant activity than the total phenolic content, since ME and HE have very similar phenolic content, but since ME has triple the total flavonoids of HE, its antioxidant activity EC_{50} was 6 times lower. These data may provide a parameter to identify what may have happened regarding the species *Baccharis reticularia*, which showed the highest SPF value (Table 3) and the lowest antioxidant activity ($EC_{50} = 75.44$) when compared to the other species (Table 6).

Conclusions

Based on the bibliometric analysis results, there is a recognized knowledge gap regarding studying the photoprotective and antioxidant properties of these native plants from the Brazilian restinga.

The hunt for new, more natural, and less harmful substances is still in the spotlight due to the toxic issues that UV filters generate in aquatic systems and living things. Because natural products tend to be more versatile and less hazardous to health, there is a growing economic demand for their utilization.

Usually, antioxidants included in sunscreen formulations are additives to stabilize and attenuate the oxidative effect of UV radiation and help to reduce the risk of non-melanoma skin cancer. Normally the antioxidants used in these formulations do not influence the SPF value (40). However, our research revealed that the species under study exhibited photoprotective activity and may have the ability to add antioxidant properties as well as to contribute to the SPF of the final sunscreen product. As a result, the use of these species in sunscreen formulations showed great promise for the development of multifunctional cosmetic formulations that aim to improve sunscreen effectiveness and shield human skin from UV-induced damage. Hence, our findings provide hitherto unheard-of information about the species under study to the scientific literature.

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Conflict of interest

The authors declare no conflicts of interest.

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