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Highlights: (1) Amazonian plants exhibit high antioxidant capacity across various botanical families. (2) Phenolic compounds, such as flavonoids and tannins, are the main antioxidants. (3) Amazonian plants have great medicinal potential for prevention and treatment.

PRE-PROOF

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ABSTRACT

Objective: review the scientific literature that addresses the antioxidant capacity of Amazon plants. Methods: This is an integrative review in English language papers carried out on PubMed/MEDLINE, in the period from 2002 to 2022. Results: The search resulted in 21 eligible papers in which 24 plants with antioxidant capacity were identified, distributed in 15 families. It was found that the studies do not have a standardized methodology, which makes it difficult to compare between plants. Despite this, it was possible to define the Myrtaceae and Lauraceae families as those that have plant species with the highest antioxidant potential, with *Psidium guajava* (32000 µmol TE/g) and *Aniba canelilla* (1.8ppm) being their representatives with the highest potential, respectively. Conclusion: A diversity of Amazonian plants with antioxidant potential was observed in different botanical families. The analysis of studies points to phenolic compounds, such as flavonoids, tannins, chromones, FAE and quercetin, as principal determinants for these species to have this antioxidant capacity. Future developments in this line of research include more studies with the plants with the highest antioxidant capacity with a focus on the analysis of the chemical constituents that make up the samples of the Amazonian plants.

Keywords: Plants. Antioxidants. Amazon ecosystem.

POTENCIAL ANTIOXIDANTE DE PLANTAS DA AMAZÔNIA: UMA REVISÃO INTEGRATIVA

RESUMO

Objetivo: revisar a literatura científica que aborda a capacidade antioxidante de plantas da Amazônia. **Métodos:** Trata-se de uma revisão integrativa em artigos de língua inglesa realizada no <u>PubMed/MEDLINE</u>, no período de 2002 a 2022. **Resultados:** A busca resultou em 21 artigos elegíveis nos quais foram identificadas 24 plantas com capacidade antioxidante distribuídas em 15 famílias. Foi verificado que os estudos não possuem metodologia padronizada, o que dificulta a comparação entre as plantas. Apesar disso, foi possível definir as famílias *Myrtaceae* e *Lauraceae* como as que possuem as espécies vegetais com maior

potencial antioxidante, sendo a Psidium guajava (32000 µmol TE/g) e Aniba canelilla (1,8ppm) suas representantes com maior potencial, respectivamente. Conclusão: Uma diversidade de plantas amazônicas com potencial antioxidante foi observada em diferentes famílias botânicas. A análise dos estudos aponta compostos fenólicos, como flavonóides, taninos, cromonas, FAE e quercetina, como principais determinantes para que essas espécies tenham essa capacidade antioxidante. Desenvolvimentos futuros nesta linha de pesquisa incluem mais estudos com as plantas com maior capacidade antioxidante com foco na análise dos constituintes químicos que compõem as amostras das plantas amazônicas.

Palavras-chave: Plantas. Antioxidantes. Ecossistema amazônico.

INTRODUCTION

Reactive oxygen species (ROS) are molecules or fragments of molecules with high reactivity potential due to unpaired electrons in their molecular orbit (1). In the human body, they can be beneficial or harmful, depending on the concentration that is normally controlled by the metabolic action of antioxidants (2). These molecules can originate from the increase of ROS that can be associated with molecular dysfunctions originating from the endothelium. In addition, part of these ROS are originated from the family of non-phagocytic nicotinamide adenine dinucleotide oxidases. Some mitochondrial electron transport enzymes, xanthine oxidase, cyclooxygenase, lipoxygenase and uncoupled nitric oxide synthase (NOS) can also originate free radicals in cells (3).

In the body there are reactive oxygen species with the potential to cause degenerative diseases, aging and cell death. The formation of ROS in vivo occurs via the catalytic action of enzymes, during the processes of electron transfer that occur in cellular metabolism and by exposure to exogenous factors (4). Antioxidants, in turn, have the ability to neutralize reactive oxygen species (ROS). The removal of excess of these substances is important for the body's oxidative balance (5).

Antioxidants are effective even at low concentrations; for this, they have compounds that donate electrons or a hydrogen radical that causes a reduction in the substrate. These antioxidants can be synthetic or natural, in addition to also being classified as primary or

secondary (6). Among the antioxidants classified as natural, there are tocopherols, vitamin C, carotenoids and phenolic compounds. The chemical structure of these antioxidants is composed of an aromatic ring with a hydroxyl group attached to it, including mainly phenolic acids and flavonoids, which act against the attack of free radicals. In nature, there are natural antioxidants found in plants, in the regions of the stem, leaf, fruit, pulp, regularly having different conformations. They are found in the form of vitamins, essential oils, organic acids, among others (7).

Phytotherapics are substances of plant origin obtained with exclusive use of active plant raw materials in which safety and efficacy are based on clinical evidence and characterized by the constancy of their quality (8). Some of these substances contain antioxidant properties and can act against oxidative stress (9). In the traditional medicine of some regions of the Amazon, the pharmacological potential of the biodiversity is used for the treatment of various diseases through the use of medicinal herbs from local natural resources (10). In recent years, the search for new alternatives of natural origin drugs has grown in areas of biodiversity, such as the Amazon, have been the target of pharmacological studies with the aim of discovering alternative drugs of natural origin (11). The aim of this study was to review the scientific literature that addresses the antioxidant capacity of Amazon plants.

MATERIAL AND METHODS

This is an integrative review in which the selection of papers occurred from the following steps: identification, analysis, eligibility and inclusion. In the first stage, keywords and health descriptors from the Medical Subject Headings MeSH were used to identify works in the National Library of Medicine and National Institutes of Health (PubMed/MEDLINE) database. The crossings for the research were: ("Plants" OR "Plant extracts" OR "Phytotherapy") AND ("Antioxidants") AND ("Amazonian Ecosystem" OR "Amazon"). The filter was also made by month and year, selecting the works published between May 2002 and October 2022.

In the second stage, the titles, abstracts and keywords were read. Studies that pointed out plants from the Amazon ecosystem that have antioxidant potential, proven from quantitative experiments of efficiency levels determination, were prioritized. Duplicate papers

and those that did not correspond to this criterion were excluded from the review. The third stage was similar to the second, with the difference that the full text was read, separating the final sample for inclusion in the review.

After the selection of the papers, the results in IC50 of the antioxidant capacity of the Amazon plants were converted to Parts per Million (ppm) and the results expressed in trolox equivalent were standardized in order to facilitate the comparison between the results obtained. IC50 is a parameter of measure for evaluating the inhibitory activity of a substance in relation to another, thus, the lower the IC50, the more active a compound is (12).

RESULTS

Figure 01 presents a flowchart with the results of the paper selection. Initially, 331 papers were identified, in which, after the evaluations based on the selection criteria, the final sample was closed with 21 papers.

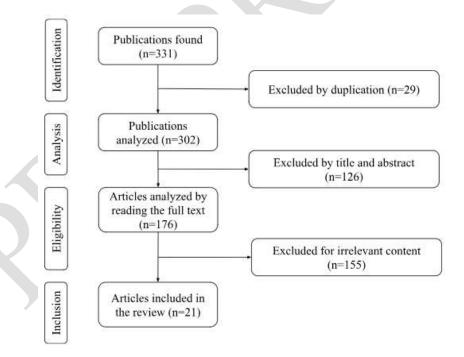


Figure 01. Flowchart of papers selection.

Table 01 presents the title of the 21 references included in this integrative review.

Table 01. Papers selected for this review.

Table 01. Papers selected for this review. Title				
Titte	Reference			
Amazon fruits inhibit growth and promote pro-apoptotic effects on human ovarian carcinoma cell lines	42			
Amazonian palm Oenocarpus bataua ("patawa"): Chemical and biological antioxidant activity — Phytochemical composition	52			
Aniba canelilla (Kunth) Mez (Lauraceae): A Review of Ethnobotany, Phytochemical, Antioxidant, Anti-Inflammatory, Cardiovascular, and Neurological Properties.				
Aniba rosaeodora (Var. amazonica Ducke) Essential oil: Chemical Composition, Antibacterial, Antioxidant and Antitrypanosomal Activity	39			
Antioxidant activity and characterization of phenolic compounds from bacaba (Oenocarpus bacaba Mart.) Fruit by HPLC-DAD-MSn	53			
Antioxidant activity and peroxidase inhibition of Amazonian plants extracts traditionally used as anti-inflammatory	11			
Antioxidant activity and potential photoprotective from amazon native flora extracts	20			
Antioxidant and Antiulcerogenic Activity of the Dry Extract of Pods of Libidibia ferrea Mart. Ex Tul. (Fabaceae)	22			
Anti-inflammatory and antioxidant activities of cat's claw (Uncaria tomentosa and Uncaria guianensis) are independent of their alkaloid content	26			
Bioactive Constituents, Radical Scavenging, and Antibacterial Properties of the Leaves and Stem Essential Oils from Peperomia pellucida (L.) Kunth	32			
Calycophyllum spruceanum (Benth.), the Amazonian "tree of youth" prolongs longevity and enhances stress resistance in Caenorhabditis elegans	28			
Chemical composition and antioxidant capacity of açaí (Euterpe oleracea) genotypes and commercial pulps	54			
Determination of carotenoids, total phenolic content, and antioxidant activity of Arazá (Eugenia mazonian McVaugh), an mazonian fruit	58			
Evaluation of Amazon fruits: chemical and nutritional studies on Borojoa sorbilis	27			
Fruit-based drink sensory, physicochemical, and antioxidant properties in the Amazon region: Murici (Byrsonima crassifolia (L.) Kunth and verbascifolia (L.) DC) and tapereba (Spondia mombin)	66			
GC-MS profiling of the phytochemical constituents of the oleoresin from Copaifera langsdorffii Desf. And a preliminary in vivo evaluation of its antipsoriatic effect	21			
Myrcia paivae O.Berg (Myrtaceae) Essential Oil, First Study of the Chemical Composition and Antioxidant Potential	67			
Pedra-ume caá fruit: An Amazon cherry rich in phenolic compounds with antiglycant and antioxidant properties	43			
Physicochemical properties and effects of fruit pulps from the amazon biome on physiological parameters in rats	63			
Ripe and unripe inajá (Maximilia maripa) fruit: A new high source of added value bioactive compounds	55			
Guayusa (Ilex guayusa L.) new tea: phenolic and carotenoid composition and antioxidant capacity	69			

DISCUSSION

The antioxidant potential of plants is used as an important reducer of ROS and is used for the treatment of degenerative diseases, such as Parkinson's, Alzheimer's and diabetes (4). The scientific evidence of their bioactivity in pharmacological research was indispensable for the use of these plants as a medicinal resource and for the development of techniques for analyzing antioxidant activity (5).

The selected papers were based on direct and indirect methods of antioxidant capacity analysis. Indirect methods are made by a redox reaction between the antioxidant and the oxidant, while direct methods are characterized by a dispute between the radicals generated from a ROS source and an oxidizable probe. Some papers used the 1,1-diphenyl-2-picrylhydrazyl (DPPH) method of evaluating the antioxidant activity of Amazon plants. This technique measures the antioxidant capacity from the neutralization or reduction of the compound, through the transfer of an electron or transfer of an atom (13).

Other techniques used by the analyzed papers were the oxygen radical absorbance capacity (ORAC) method, which is based on the measurement of the protective factor of the antioxidant compound to the proteins that lost their conformational structure when suffering oxidative damage, and the method of 2,29-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) which is the most used to evaluate the total antioxidant activity through the capture of the radical 2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) in lipophilic and hydrophilic compounds (14,15).

The results of the selected papers were expressed in IC50, which is a parameter of measure for evaluating the inhibitory activity of a substance in relation to another, thus, the lower the IC50, the more active a compound is (12). Another form of the results of the selected papers was in trolox equivalent, which expresses the antioxidant capacity of a mass of sample from its equivalent in trolox concentration (16).

Antioxidant potentials expressed in IC50

The Fabaceae family, also known as legume, has about 18000 species and is able to develop in varied types of soil and climate. In the Amazon region, plants of the Fabaceae family are used in medicine practiced by traditional peoples mainly against infectious diseases

(17-19). Among the components of this family, the species *Dalbergia monetaria* was the one that presented the highest antioxidant capacity (table 2) (20-22). This can be justified by the presence of flavonoids in the analyzed sample, which is defined as a secondary metabolite associated with the reduction of reactive oxygen species and the protection against oxidative stress. These substances are phenolic compounds that besides acting as antioxidants also have antimicrobial and photoreceptor function (23,24).

Another family of Amazon plants with antioxidant capacity was the Rubiaceae, which is characterized by the tropical worldwide location and the production of metabolites with great pharmacological potential. In Brazil, the Rubiaceae has approximately 1400 species and has plants of great ornamental, economic and phytotherapeutic importance for traditional medicine (25). The species *Calycophyllum spruceanum* was the one that obtained the best results of antioxidant capacity (table 2) (26-28). This can be explained by the presence of tannin in the chemical constitution of the plant, a flavanone substance characterized as a type of flavonoid with high antioxidant and antibacterial power (29,30).

The species *C. spruceanum* presented divergent results of antioxidant capacity in studies done by different papers with the same analysis technique (11,28) This can be explained by methodological divergence between the studies, mainly regarding the collection of samples, which can modify the results of the antioxidant capacity due to impacts on the conditions of the analyzed substrate that should be as similar as possible to the form found in nature (31).

The diversified constituents of the same plant can present different antioxidant capacities. This can be perceived by evaluating the DPPH data of *Peperomia pellucida*, the only plant identified from the Piperaceae family in this review. The researchers' results showed that the antioxidant potential of the leaf oil is much lower than the stem oil (Table 2) (32). Aebisher et al. (33) noticed, from the study of the chemical components of the essential oils (EOs) of species from the Lamiaceae family, that non-phenolic components can interfere with the action of the phenolic components, which can justify this difference and suggest the relevance of evaluating the capacities of the constituents of the Amazon plants in an isolated way.

In addition, the EOs have high antioxidant capacity because they have a high concentration of phenolic compounds in their chemical structure (34). However, when comparing the antioxidant action of these oils from *P. pellucida* with the plant that presented the lowest potential of the Rubiaceae family (the *Borojoa sorbilis*, with DPPH equal to 68.09 ppm), these oils extracted from the leaf and stem of *P. pellucida* are much lower.

Plants can present benefits that add up to the antioxidant activity, since studies with EOs of species from the traditional medicine of Mexico identified chemical components in these oils that have antimicrobial activity in Gram-positive and Gram-negative bacteria (35). However, there is also the possibility of them having compounds that induce cardiovascular, hepatic and central nervous system and digestive toxicity in humans (36) and, although many plants already have this toxic potential known, several Amazon plant species do not have this chemical characterization, and may be composed of harmful compounds already known or others not yet discovered.

The Lauraceae family has about 70 genera and 2500 species spread across the continents of Asia and South America and is rich in benzyltetrahydroisoquinoline alkaloids and its largest genus is *Cinnamomum*. The genus *Aniba* is found throughout America and has 92 species (37). The analyzed papers showed that *Aniba canelilla* is the species that has the highest antioxidant potential within the Lauraceae family (table 2) and has as its main chemical constituents 1-nitro-2-phenylethane, which has antinociceptive and anti-inflammatory action; methyl eugenol, which is a substance found in the essential oils of several aromatic plants and has antioxidant activity; eugenol, safrole, anabasine, tannin, a-pinene, b-pinene, b-phellandrene, b-caryophyllene, b-sesquiphellandrene, p-cymene, linalool, a-copaene and spathulenol. Its antioxidant potential is related to phenolic antioxidants and coumarins, and can be used for the development of pharmaceutical products (20,38).

Another species of the Lauraceae family with antioxidant capacity is *Aniba* rosaeodora (table 2), used as a medicinal plant for having antimicrobial and antioxidant potential. Its main chemical constituents are: α-terpinolene, linalool cis-oxide and linalool (39). *A. canelilla* has more antioxidant compounds than *A. rosaeodora*, which corroborates the results presented in the literature.

The Malpighiaceae is a family of plants that is part of the Brazilian flora with more than 500 species grouped in 45 genera, among the most numerous are *Byrsonima Rich, Ex Kunth, Heteropterys Kunth* and *Banisteriopsis C.B.Rob. ex Small* (40). According to the literature, the species *Byrsonima japurensis* has IC50 of 8.4 ppm revealing considerable antioxidant capacity, this can be explained by the large amount of flavonoids that it has (11,41,42).

Other Amazon plants analyzed by the selected papers were those of the Apocynaceae family, with the species *Aspidosperma nitidum*, Myrtaceae, with the species *Eugenia punicifolia*, Passifloraceae, with *Passiflora nitida*, Olacaceae, with the species *Ptychopetalum olacoides*, Bignoniaceae, with *Arrabidaea chica*, and Moraceae, with the species *Brosimum acutifolium* (table 2) (11,20,43). Among these plants, the species of the Moraceae family was the one that presented the highest antioxidant activity. A possible explanation for this result is the presence of 4'-Hydroxy-7,8-[2-(2-hydroxy isopropyl)dihydrofuran] and 4',7-dihydroxy-8-(3,3-dimethylallyl) in its chemical constitution, which is one of the classes of flavonoids, have several biological properties inherent to the group, such as antioxidant, anti-inflammatory, antitumor (44).

Table 2. Antioxidant potentials expressed in IC50.

Family	Species	Antioxidant potentials (IC50)	Reference
Fabaceae	Dalbergia monetaria	5,46ppm	20
	Caesalpinia pyramidalis	6,45 ppm	20
	Libidibia férrea	53,20 ppm	22
	Copaifera langsdorffii	8710 ppm	21
	Calycophyllum spruceanum	3 ppm	28
Rubiaceae	Uncaria guianensis	12,6 ppm	26
	Uncaria tomentosa	20,8 ppm	26
	Borojoa sorbilis	66,19 ppm	27
Piperaceae	Peperomia pellucida	1.670 ppm - óleo da folha 2.830 ppm - óleo do caule	32
Lauraceae	Aniba canelilla	1,8 ppm	20
	Aniba rosaeodora	15,46 ppm	39
Malpighiaceae	Byrsonima japurensis	8,4 ppm	11
Apocynaceae	Aspidosperma nitidum	99,14 ppm	20
Myrtaceae	Psidium guajava	89,5 ppm	43
Passifloraceae	Eugenia punicifolia	49,9 ppm	11
Olacaceae	Ptychopetalum olacoides	29,7 ppm	11
Bignoniaceae	Arrabidaea chica	15,36 ppm	20
Moraceae	Brosimum acutifolium	2,84 ppm	20

Antioxidant potentials expressed in trolox equivalent

Only one eligible paper was found with the ORAC method of the Fabaceae family, which determined the antioxidant capacity of *Chamaecrista diphylla* (L.) (Table 3). The authors identified for the first time 35 chemical compounds, of which 32 had not yet been characterized in the genus of this species (45). Among them, aloesol (C13H14O4), two isomers of carviolin (C16H12O6) and 7-hydroxy-2,5-dimethyl-4H-chromen-4-one (C11H10O3) have chromones in the chemical skeleton, which justifies the antioxidant potential of the plant, since they have affinity with ROS, decreasing oxidative stress (46).

The highest concentration of these substances was found in the enriched fraction of ethyl acetate (FAE) of the ethanolic extract studied by the researchers, which justifies the antioxidant capacity of it isolated (9440 µmol TE/g) being superior to the potential of the extract, since several studies prove the antioxidant effect of FAE of plants (47-49). In addition, In vivo studies with FAE of *Trichilia catigua* indicate improvement in the oxidative control of the brain in induced ischemic state (50).

The Arecaceae family, also known as the palm family, is characterized by being adapted to various types of soil and climate, which explains its worldwide distribution. However, the Aracaceae develops more abundantly in regions of equatorial and humid climate like the Amazonian region. This family has approximately 3919 catalogued species, and in Brazil they have been registered 270 species. In the country, the Arecaceae has medicinal and economic importance for local populations (51).

The *Maximilia maripa* (Inajá) species was the one that showed the highest antioxidant activity (Table 3) (52–55). This can be explained by the presence of carotenoids in its chemical composition, such as licopeno and β -carotene, which have great antioxidant and anticancer capacity. In addition, the *M. maripa* also has a high content of phenolic compounds that have the ability to combat oxidative stress, which also contributes to its antioxidant capacity (56).

Another plant family from the Amazon with antioxidant capacity is Myrtaceae, which is characterized by being typical of tropical and subtropical climates. This family has about 5800 plant species and presents chemical compounds associated with prevention of diseases such as cancer and diabetes mellitus (57). The *Psidium guajava* species was the one that showed the highest antioxidant capacity in the analyzed papers (Table 3) (53,58). A possible explanation for this is the presence of high levels of phenolic compounds such as quercetin in the plant, as well as other flavonoids, substances that combat oxidative stress. In addition, this plant species has a high content of polysaccharides in its composition, which have antioxidant properties, anti-inflammatory and antitumor effects (59).

The families Sapotaceae, Oxalidaceae and Clusiaceae presented plant species with antioxidant capacity (Table 3). The Sapotaceae family is represented in Brazil by 11 genera and about 230 species; it belongs to the group of woody plants; *Pouteria caimito* revealed

antioxidant capacity; this value can be explained by the presence of chemical components that offer antioxidant properties; such as riboflavin, vitamin A, niacin and vitamin C (60-61). The *Averrhoa bilimbi* L. species belongs to Oxalidaceae; a family composed mainly of shrubs and trees distributed in temperate and tropical zones; it has medicinal importance for traditional communities for its characteristic antioxidant and anti-inflammatory property (62-63).

The only plant from Clusiaceae found in this review was *Garcinia xanthochymus*; this plant presents various phytochemicals that contribute to this value of potential antioxidant capacity; such as xanthones, benzofenones, flavonoids, depsidones and isocumarines. These phytochemicals contribute to pharmacological activities of this plant as an antioxidant and antidiabetic agent (63-64).

Tabel 3. Antioxidant potentials expressed in trolox equivalent

Family	Species	Antioxidant potentials (Trolox Equivalent)	Reference
Fabaceae	Chamaecrista diphylla (L.)	4290 μmol TE/g	45
Aracaceae	Maximilia maripa	6820 μmol TE/g	55
	Euterpe oleracea	1572,42 μmol TE/g	54
	Oenocarpus bataua	2292,5 μmol TE/g	52
	Oenocarpus bacaba	0,3425 μmol TE/g	53
Myrtaceae	Psidium guajava	32000 μmol TE/g	53
	Eugenia stipitata	2 μmol TE/g	58
Sapotaceae	Pouteria caimito	0,0289 μmol TE/g	63
Oxalidaceae	Averrhoa bilimbi L.	0,0062 μmol TE/g	63
Clusiaceae	Garcinia xanthochymus	0,0429 μmol TE/g	63

The literature presents evidence that test results for analysis of antioxidant capacity may vary depending on the method used for analysis. The chemical conditions during tests such as pH of sample are factors that must be considered during selection of test used for measurement of antioxidant capacity; because these determinants can modify real capacities of samples analyzed. Techniques used must be rigorously selected according to parameters most favorable for each method analysis (65).

The results found corroborate with the literature, since there is divergence in the antioxidant capacity of the samples when analyzed by different techniques. The study done by Vargas et al. (11), shows differences in the antioxidant capacity of the samples analyzed by the DPPH and ABTS assays. The *Calycophyllum spruceanum* and *Byrsonima japurensis* samples, when analyzed by DPPH, have the best results. However, when analyzed by ABTS, the best results were from the *C. spruceanum, Maytenus guyanensis* and *Ptychopetalum olacoides* samples. This divergence of results was also found in other studies analyzed (11,27,42,66,67).

It is noted that many studies of measurement of the antioxidant capacity of native plants of the Amazon use different methodologies that are adapted from another study previously carried out (27,28,43,55,67-69). This limits the comparison of the results between studies, since the procedures generate results in units of measure that are not convertible into a standard type, making it impossible to rank the plants according to their antioxidant potentials in a single listing (31). In this review, for example, the works were divided into two groups, not being possible to determine which Amazonian plant, of the works analyzed, has the highest antioxidant capacity among the others.

Alam et al. (70) proved that the assays performed in vitro from the DPPH method present superior results compared to the in vivo potential. This is because the method uses alcoholic solvents, mainly ethanol, which increases the solubility and transfer of hydrogens to the DPPH radical. In aqueous media, this solubility decreases, also declining the antioxidation capacity of the compounds of these plants.

CONCLUSION

A diversity of Amazonian plants with antioxidant potential was observed in different botanical families. The analysis of studies points to phenolic compounds, such as flavonoids, tannins, chromones, FAE and quercetin, as principal determinants for these species to have this antioxidant capacity. Furthermore, some species also present additional benefits, such as antimicrobial activity, but there is still a lack of studies that evaluate the constituents of these plants in isolation to determine their efficacy and their mechanisms of action. The study was limited by the use of different techniques, since this can cause divergence of results and

makes research analysis difficult. Future developments in this line of research include more studies with the plants with the highest antioxidant capacity with a focus on the analysis of the chemical constituents that make up the samples of the Amazonian plants, since non-phenolic substances can interfere with the action of the phenolic constituents and compromise the antioxidant capacity of the Amazonian plants.

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