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# Amaranthaceae as a Bioindicator of Neotropical Savannah Diversity

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Additional information is available at the end of the chapter

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## 1. Introduction

Brazil is the first in a ranking of 17 countries in megadiversity of plants, having 17,630 endemic species among a total of 31,162 Angiosperm species [1], distributed in five Biomes. One of them is the Cerrado, which is recognized as a World Priority Hotspot for Conservation because it has around 4,400 endemic plants – almost 50% of the total number of species – and consists largely of savannah, woodland/savannah and dry forest ecosystems [2,3]. It is estimated that Brazil has over 60,000 plant species and, due to the climate and other environmental conditions, some tropical representatives of families which also occur in the temperate zone are very different in appearance [4].

The Cerrado Biome is a tropical ecosystem that occupies about 2 million km<sup>2</sup> (from 3-24° Lat. S and from 41-43° Long. W), located mainly on the central Brazilian Plateau, which has a hot, semi-humid and markedly seasonal climate, varying from a dry winter season (from April to September) to a rainy summer (from October to March) [5-7]. The variety of landscape – from tall savannah woodland to low open grassland with no woody plants - supports the richest flora among the world's savannahs (more than 7,000 native species of vascular plants) and a high degree of endemism [6,8]. This Biome is the most extensive savannah region in South America (the Neotropical Savannah) and it includes a mosaic of vegetation types, varying from a closed canopy forest (“cerradão”) to areas with few grasses and more scrub and trees (“cerrado *sensu stricto*”), grassland with scattered scrub and few trees (“campo sujo”) and grassland with little scrub and no trees (“campo limpo”) [3,9]. Among the grassland areas there are some flat areas with rocky soil, called “campos rochosos”, which are considered Cerrado areas because of their flora, especially when located in Chapada Diamantina (Bahia State), a transition area between Cerrado and Caatinga Biomes.

Although the Cerrado is considered a Hotspot for the conservation of global biodiversity, with plant species completely adapted to survive adverse conditions of soil and climate,

only 30% of this Neotropical Savannah biodiversity is reasonably well known [8,10]. Coutinho [11] believes that the frequent occurrence of fire is one of the most important factors to determine this Biome's vegetation, acting as a renewal element that selects structural and physiological characteristics. Nowadays it is believed that more than 40% of the original vegetation has already been converted into human-disturbed areas, due to the expansion of crops [12,13]. This process has accelerated the fragmentation of natural habitat, increasing the pressure on local biodiversity extinction and introducing exotic species, also amplifying soil erosion, water pollution and alterations in vegetation and hydrologic conditions [2,8,14].

The Amaranthaceae family is composed of 2,360 species and here will be listed those that occur in Brazil, emphasizing the Cerrado species and including information on endemism, endangerment and economic or potential use. We also provide a list of the most important bibliographical references for those who are interested in studying the species of this family. Some aspects of morphology, leaf anatomy and ultrastructure will be shown for six species found in the Neotropical Savannah core area (Chapada dos Veadeiros) and some of these aspects, as well as taxonomy and ecology, will be discussed in order to propose the use of this plant family as an indicator of the diversity in open areas of this Biome.

## 2. Methodology

### 2.1. List of the Brazilian Amaranthaceae species and those in RPPN Cara Preta

The Brazilian Amaranthaceae list (Table 1) was based on the research by the Brazilian taxonomists Marchioretto [15-21] and Siqueira [22-25] and on the most important taxonomic references to this Family both from the literature (Table 2) and Brazilian Herbaria (Table 3). All cited Herbaria are listed according to the Index Herbariorum [26,27].

The species to be detailed were collected in a Conservation Unit named Reserva Particular do Patrimônio Natural Cara Preta (RPPN Cara Preta), located in Alto Paraíso, Goiás State, Brazil. After obtaining authorization from the NGO Oca Brasil, random walks were done in order to locate, photograph and mark species with a Global Positioning System device, and to collect and make exsiccates for Herbaria deposits, from September 2006 until March 2009. Although some plant leaves were collected during the vegetative stage, these specimens were visited until flowering to identify them correctly. All exsiccates were deposited in Brazilian Herbaria as standard control material (prioritizing PACA, UnB and IBGE Herbaria) and these species are included in Table 1.

### 2.2. Leaf anatomy and ultrastructure

Completely expanded leaves, from 3<sup>rd</sup> to 5<sup>th</sup> node from the apex, of two to six specimens of each species were collected and sectioned. Part of the leaf medial region was fixed in ethanol, acetic acid and formaldehyde [28] for 24 hours and preserved in ethanol 70% until analysis to describe the anatomy and identify starch and crystal composition [28].

Some pieces of the leaf medial region were immediately submerged in a Karnovsky solution [29] of glutaraldehyde 2%, paraformaldehyde 2% and sucrose 3% in sodium cacodylate 0.05 M buffer for 12 to 24 hours and preserved in sodium cacodylate 0.05 M until processing for analysis under an electron microscope. For the latter analysis, these pieces were post-fixed in 2% osmium tetroxide and 1.6% potassium ferricyanide (1:1 v/v), followed by in-block staining with 0.5% uranyl acetate solution (overnight). These samples were then dehydrated in an acetone ascending series and slowly embedded in Spurr's epoxy resin. Semi-thin and ultra-thin sections were obtained in ultramicrotome with glass and diamond knives. Semi-thin sections were stained with toluidine blue and analysed under the optical Zeiss Axiophot, and ultra-thin sections were analysed under the transmission electron microscope TEM JEOL JEM 1011.

### 3. Results and discussion

The Amaranthaceae family is composed of 2,360 species and 146 of them are found in Brazil (Table 1). Ninety-eight species within the family are found in the Cerrado and 73 spp. are endemic to Brazil, of which 13 are endemic to the Cerrado Biome (Table 1). Twenty Amaranthaceae species are exclusive to the Cerrado (Table 1).

At least 22 Amaranthaceae species are referred to as being used in folk medicine (Table 1). In Brazil, only two of these species are already used as commercial drugs, as capsules containing their powdered roots, with studies to support their medicinal activity: *Hebanthe eriantha* (Poir.) Pedersen and *Pfaffia glomerata* (Spreng.) Pedersen (Table 1), both known as "Brazilian-ginseng". However, there is neither registered success in isolating or synthesizing their components nor any economic studies about the viability of this kind of pharmaceutical procedure.

Although the species *Gomphrena macrocephala* St.-Hil. is not cited as medicinal (Table 1), the fructan content in its roots has been determined [30] because this species was considered synonymous with *G. officinalis* Mart. [31]. Later, it was determined that *G. officinalis* was synonymous with *G. arborescens* L.f. and not with *G. macrocephala* [22]. Studying *G. arborescens*, fructan was also determined as the principal carbohydrate in its subterranean system [32]. This species is used in popular medicine to heal respiratory diseases (asthma and bronchitis), to reduce fever and as a tonic [33-35]. An *in vivo* study (in cats) with the use of fructans isolated from *Arctium lappa* L. (Asteraceae) reported a cough-suppressing activity [36], and the presence of fructan in *G. arborescens* roots can partially justify the use of this species as a medicinal plant.

Most members of Brazilian Amaranthaceae are only known by taxonomists and 42 species are in danger of extinction according to Brazilian regional lists; 14 of them are recognized as endangered by the Brazilian Ministry of the Environment (MMA – "Ministério do Meio Ambiente") (Table 1). Most of the endangered species are classified according to the IUCN Red List of vulnerability categories, some even with the same criteria, and there is a wide range of research still to be done.

Species	Bioma and level of endemism	Species Threat Level	Habit, popular name and species knowledge
<i>Achyranthes aspera</i> L.	Cerrado		Herb; plant used as indigenous medicine in Ethiopia with chemistry study [37]
<i>Achyranthes indica</i> (L.) Mill.	Cerrado		Herb
<i>Alternanthera adscendens</i> Suess.	Cerrado exclusive		Shrub
<i>Alternanthera albida</i> (Moq.) Griseb.			Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Alternanthera aquatica</i> (D.Parodi) Chodat			Herb
<i>Alternanthera bahiensis</i> Pedersen	Cerrado, endemic to Brazil		Herb or subshrub
<i>Alternanthera bettzichiana</i> (Regel) G.Nicholson			Herb; popularly named "anador"; folk medicinal plant, used as analgesic and antipyretic [39]
<i>Alternanthera brasiliana</i> (L.) Kuntze	Cerrado, endemic to Brazil		Herb; called "perpétua-do-mato, periquito-gigante, penicilina" or Brazilian joyweed; folk medicinal plant, used as diuretic, digestive, depurative, bequic, astringent and antidiarrhoeal; ornamental plant; C <sub>3</sub> photosynthesis structure [40-42]
<i>Alternanthera decurrens</i> J. C. Siqueira	Brazilian Cerrado endemic (Januária - MG)	CR [43]	Subshrub
<i>Alternanthera dendrotricha</i> C.C.Towns.	Cerrado, endemic to Brazil		Shrub
<i>Alternanthera flavida</i> Suess.			Subshrub
<i>Alternanthera hirtula</i> (Mart.) R.E.Fr.		EN [44]	Herb
<i>Alternanthera januariensi</i> J. C. Siqueira	Brazilian Cerrado endemic (Januária - MG)	CR [43]	Subshrub
<i>Alternanthera kurtzii</i> Schinz			Herb
<i>Alternanthera littoralis</i> P.Beauv.			Herb; called "periquito-da-praia" [45]
<i>Alternanthera malmeana</i> R.E.Fr.		EN [44]	Herb
<i>Alternanthera markgrafii</i> Suess.	Brazilian Cerrado endemic (Serra de Grão Mogol - MG)		Herb

<i>Alternanthera martii</i> (Moq.) R.E. Fries	Cerrado, endemic to Brazil		Subshrub
<i>Alternanthera micrantha</i> R.E.Fr.	Endemic to Brazil	VU [44]	Herb; called "periquito-da-serra" [45]
<i>Alternanthera minutiflora</i> Suess.	Endemic to Brazil		Herb
<i>Alternanthera multicaulis</i> Kuntze	Endemic to Brazil		Herb
<i>Alternanthera paronychioides</i> A.St.-Hil.	Cerrado	VU [44,46]	Herb; called "periquito-roseta, periquito"; C <sub>3</sub> -C <sub>4</sub> intermediary photosynthesis structure; C <sub>4</sub> photosynthesis physiology; ornamental plant [38,41,42,45,47]
<i>Alternanthera philoxeroides</i> (Mart.) Griseb.	Cerrado		Herb; called "perna-de-saracura, carrapicho-de-brejo" and alligatorweed [45]
<i>Alternanthera pilosa</i> Moq.			Herb
<i>Alternanthera praelonga</i> A.St.-Hil.		CR [44]	Herb
<i>Alternanthera puberula</i> D.Dietr.	Cerrado exclusive		Herb
<i>Alternanthera pulchella</i> Kunth			Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Alternanthera pungens</i> Kunth	Cerrado		Herb; called "erva-de-pinto"; folk medicinal plant, used to treat syphilis and skin diseases; C <sub>4</sub> photosynthesis physiology [38,39]
<i>Alternanthera ramosissima</i> (Mart.) Chodat	Cerrado		Herb
<i>Alternanthera regelii</i> (Seub.) Schinz	Cerrado exclusive, endemic to Brazil		Herb
<i>Alternanthera reineckii</i> Briq.	Cerrado	VU [44]	Herb; called "periquito-de-reineck" [45]
<i>Alternanthera rufa</i> (Mart.) D.Dietr.	Cerrado, endemic to Brazil		Herb
<i>Alternanthera sessilis</i> (L.) R.Br.	Cerrado	LC [48]	Herb
<i>Alternanthera tenella</i> Colla	Cerrado	VU [44]	Herb; called "apaga-fogo, carrapichinho, corrente, folha-de- papagaio, periquito, periquito-figueira, perpétua-do-mato, sempre-viva" and joyweed; folk medicinal plant, used as diuretic; this species is naturally infected by a potyvirus; C <sub>3</sub> -C <sub>4</sub> photosynthesis physiology and structure [38,39,40,45,47,49,50]

<i>Alternanthera tetramera</i> R.E.Fr.			Herb
<i>Amaranthus blitum</i> L.			Herb; called "caruru"; folk medicinal plant, used to fight anemia; C <sub>4</sub> photosynthesis physiology [38,39,51]
<i>Amaranthus caudatus</i> L.	Cerrado		Herb; called "rabo-de-gato, cauda-de-raposa, disciplina-de-freira, rabo-de-raposa"; folk medicinal plant, used to treat pulmonary diseases and as emoliente; C <sub>4</sub> photosynthesis physiology; ornamental plant [33,38,39,41]
<i>Amaranthus cruentus</i> L.	Cerrado		Herb; called "caruru-vermelho, veludo, bredo-de-jardim, crista-de-galo"; folk medicinal plant, used as emollient and laxative; C <sub>4</sub> photosynthesis physiology [33,38,39,51]
<i>Amaranthus deflexus</i> L.			Herb; called "caruru-rasteiro"; C <sub>4</sub> photosynthesis physiology [38,51]
<i>Amaranthus hybridus</i> L.	Cerrado		Herb; called "caruru, bredo" and smooth pigweed; C <sub>4</sub> photosynthesis physiology [38,51]
<i>Amaranthus muricatus</i> (Moq.) Hieron.			Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Amaranthus retroflexus</i> L.	Cerrado		Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Amaranthus rosengurtii</i> Hunz.		EN [44]	Herb
<i>Amaranthus spinosus</i> L.	Cerrado		Herb; called "caruru-bravo, caruru-de-espinho, bredo-de-espinho, caruru-de-porco"; folk medicinal plant, used to combat eczema and as emollient, laxative and antibleorrhagic; C <sub>4</sub> photosynthesis physiology [33,38,39]
<i>Amaranthus viridis</i> L.	Cerrado		Herb; called "caruru-bravo, caruru-verdadeiro, cururu, caruru-de-soldado, caruru-de-folha-miúda, amaranto-verde"; folk medicinal plant, used as emollient and diuretic desobstruente; C <sub>4</sub> photosynthesis physiology [33,38-40,51]
<i>Blutaparon portulacoides</i> (A.St.-Hil.) Mears	Cerrado	VU [44]	Herb; called "capotiraguá"; folk medicinal plant, used to combat leukorrhea; C <sub>4</sub> photosynthesis physiology [38,39]
<i>Blutaparon vermiculare</i> (L.) Mears	Cerrado		Herb; C <sub>4</sub> photosynthesis physiology [38]

<i>Celosia argentea</i> L.	Cerrado		Herb; called "celosia-branca, celósia-plumosa, crista-de-galo, crista-de-galo-plumosa, suspiro, veludo-branco"; folk medicinal plant, used to combat diarrhea and as anthelmintic and astringent; ornamental plant [39,41,52]
<i>Celosia corymbifera</i> Didr.	Endemic to Brazil		Subshrub
<i>Celosia grandifolia</i> Moq.		EN [44]	Herb, subshrub; called "bredo-domato" [45]
<i>Chamissoa acuminata</i> Mart.		VU [44]	Subshrub; called "mofungo-rabudo" [45]
<i>Chamissoa altissima</i> (Jacq.) Kunth	Cerrado	VU [44]	Subshrub; called "mofungo-gigante" [45]
<i>Chenopodium album</i> L.	Cerrado		Herb
<i>Chenopodium ambrosioides</i> L.	Cerrado		Herb; called "erva-de-santa-maria, erva-santa, quenopódio" [40]
<i>Chenopodium murale</i> L.	Cerrado		Herb
<i>Cyathula achyranthoides</i> (Kunth) Moq.			Herb
<i>Cyathula prostrata</i> Blume	Cerrado		Herb
<i>Froelichia humboldtiana</i> (Roem. & Schult.) Seub.	Cerrado		Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Froelichia interrupta</i> (L.) Moq.			Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Froelichia procera</i> (Seub.) Pedersen	Cerrado		Herb; called "ervaço"; C <sub>4</sub> photosynthesis physiology [38,41]
<i>Froelichia sericea</i> (Roem. & Schult.) Moq.			Herb
<i>Froelichia tomentosa</i> (Mart.) Moq.	Cerrado		Herb; C <sub>4</sub> photosynthesis physiology [38]
<i>Froelichiella grisea</i> R.E. Fries	Braziliann Cerrado endemic (Chapada dos Veadeiros - GO)	VU [43]	Herb; C <sub>3</sub> photosynthesis structure [42]
<i>Gomphrena agrestis</i> Mart.	Cerrado, endemic to Brazil	EN [46]	Herb
<i>Gomphrena arborescens</i> L.f.	Cerrado exclusive		Herb, subshrub; called "perpétua, perpétua-do-campo, perpétua-domato, paratudo-do-campo, paratudo-erva, raiz-do-padre"; folk medicinal plant, used as tonic, to reduce fever and against respiratory diseases; potential use as ornamental plant; roots are fructan-rich; C <sub>4</sub> photosynthesis physiology/structure [32,35,38,40,42,49,53,54]



<i>Gomphrena basilanata</i> Suess.	Endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena celosoides</i> Mart.	Cerrado		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena centrota</i> E.Holzh.	Endemic to Brazil	VU [43]	Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena chrestoides</i> C.C.Towns.	Brazilian Cerrado endemic (Chapada Diamantina - BA)	VU [43]	Subshrub
<i>Gomphrena claussenii</i> Moq.	Cerrado, endemic to Brazil		Subshrub
<i>Gomphrena debilis</i> Mart.	Endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena demissa</i> Mart.	Cerrado, endemic to Brazil		Subshrub; folk medicinal plant, used to combat the flu; C <sub>4</sub> photosynthesis physiology [38,49]
<i>Gomphrena desertorum</i> Mart.	Cerrado, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena duriuscula</i> Moq.	Endemic to Brazil	EN [43]	Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena elegans</i> Mart.	Cerrado, endemic to Brazil	VU [46]	Subshrub
<i>Gomphrena gardnerii</i> Moq.	Cerrado, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena globosa</i> L.	Cerrado		Subshrub; called "gonfrena, perpétua, perpétua-roxa, sempre-viva, suspiro, suspiro-roxo"; folk medicinal plant used to fight respiratory diseases; C <sub>4</sub> photosynthesis physiology; ornamental plant [38,40,45,55,56]
<i>Gomphrena graminea</i> Moq.	Cerrado	VU [44]	Subshrub; called "perpétua-gramínea"; C <sub>4</sub> photosynthesis physiology [38,45]
<i>Gomphrena hatschbachiana</i> Pedersen	Cerrado, endemic to Brazil	VU [43]	Subshrub
<i>Gomphrena hermogenesii</i> J.C. Siqueira	Brazilian Cerrado endemic (Chapada dos Veadeiros - GO)		Subshrub; C <sub>3</sub> photosynthesis physiology; C <sub>4</sub> photosynthesis structure [38,42]
<i>Gomphrena hillii</i> Suess.	Brazilian Cerrado endemic (Paraíso do Norte - TO)		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena incana</i> Mart.	Cerrado exclusive, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena lanigera</i> Pohl ex Moq.	Cerrado exclusive, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology and structure [38,42]
<i>Gomphrena leucocephala</i> Mart.	Endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena macrocephala</i> A.St.-Hil.	Cerrado exclusive, endemic to Brazil		Subshrub; roots are fructan-rich; C <sub>4</sub> photosynthesis physiology [30,38]

<i>Gomphrena marginata</i> Seub.	Brazilian Cerrado endemic (Diamantina - MG)		Subshrub
<i>Gomphrena matogrossensis</i> Suess.	Cerrado exclusive, endemic to Brazil		Subshrub
<i>Gomphrena microcephala</i> Moq.	endemic to Brazil		Subshrub
<i>Gomphrena mollis</i> Mart.	Cerrado, endemic to Brazil		Subshrub; called "erva-mole, erva-rosa"; folk medicinal plant, used as tonic and carminative [39]
<i>Gomphrena moquini</i> Seub.	Brazilian Cerrado endemic (Serra do Cipó - MG)		Subshrub
<i>Gomphrena nigricans</i> Mart.	Cerrado, endemic to Brazil	VU [43]	Subshrub
<i>Gomphrena paranensis</i> R.E.Fr.	Cerrado exclusive, endemic to Brazil		Subshrub, C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena perennis</i> L.		VU [44]	Subshrub; called "perpétua-sempreviva"; C <sub>4</sub> photosynthesis physiology [38,45]
<i>Gomphrena pohlii</i> Moq.	Cerrado exclusive		Subshrub; called "infalível, paratudo, paratudinho, paratudo-amarelinho"; roots are used in folk medicine against respiratory diseases; C <sub>4</sub> photosynthesis physiology and structure [38,39,42,49]
<i>Gomphrena prostrata</i> Mart.	Cerrado, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology and structure [38,42]
<i>Gomphrena pulchella</i> Mart.		EN [44]	Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena pulvinata</i> Suess.	Endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena regeliana</i> Seub.	Cerrado exclusive, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena riparia</i> Pedersen	Endemic to Brazil	CR [43]	Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena rudis</i> Moq.	Cerrado exclusive, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena rupestris</i> Nees	Cerrado, endemic to Brazil		Subshrub
<i>Gomphrena scandens</i> (R.E.Fr.) J.C.Siqueira	endemic to Brazil	VU [43]	Subshrub
<i>Gomphrena scapigera</i> Mart.	Cerrado, endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena schlechtendaliana</i> Mart.		EN [44]	Subshrub; called "perpétua-schlechtendal"; C <sub>4</sub> photosynthesis physiology [38,45]

<i>Gomphrena sellowiana</i> Mart.	Endemic to Brazil	VU [44]	Subshrub
<i>Gomphrena serturneroides</i> Suess.	Endemic to Brazil		Subshrub; C <sub>4</sub> photosynthesis physiology [38]
<i>Gomphrena vaga</i> Mart.	Cerrado, endemic to Brazil	VU [44]	Subshrub; called "thoronoé"; folk medicinal plant, used as analgesic [57]
<i>Gomphrena virgata</i> Mart.	Cerrado, endemic to Brazil		Subshrub; called "cangussú-branco, vergateza"; folk medicinal plant, antileptargic; C <sub>4</sub> photosynthesis physiology and structure [33,38,42]
<i>Hebanthe eriantha</i> (Poir.) Pedersen	Cerrado	EN [44], VU [58]	Subshrub, shrub; called "corango-açu, ginseng-brasileiro, picão-de-tropeiro, solidonia, suma"; folk medicinal plant, used to combat colic and enteritis; most of its chemical constituents are known and roots of this plant are already used by pharmaceutical companies [40,59]
<i>Hebanthe grandiflora</i> (Hook.) Borsch & Pedersen	Cerrado		Bush scandentia
<i>Hebanthe occidentalis</i> (R.E.Fr.) Borsch & Pedersen	Cerrado		Subshrub scandentia
<i>Hebanthe pulverulenta</i> Mart.	Cerrado	VU [58]	Subshrub scandentia; called "corango-veludo" [45]
<i>Hebanthe reticulata</i> (Seub.) Borsch & Pedersen			Subshrub, shrub scandentia
<i>Hebanthe spicata</i> Mart.			Shrub erect or scandentia
<i>Herbstia brasiliiana</i> (Moq.) Sohmer		EX [46]	Subshrub
<i>Iresine diffusa</i> Humb. & Bonpl. ex Willd.	Cerrado		Subshrub; called "bredinho-difuso" [45]
<i>Iresine poeppigiana</i> Klotzsch			Subshrub
<i>Lecosia formicarum</i> Pedersen	Endemic to Brazil		Subshrub
<i>Lecosia oppositifolia</i> Pedersen	Endemic to Brazil	CR [43]	Herb or subshrub
<i>Pedersenianthus argentata</i> (Mart.) Holub			Herb
<i>Pfaffia acutifolia</i> (Moq.) O.Stützer	Cerrado		Herb or subshrub
<i>Pfaffia aphylla</i> Suess.	Brazilian Cerrado endemic (Gouveia - MG)		Subshrub
<i>Pfaffia argyrea</i> Pedersen	Cerrado exclusive, endemic to Brazil	VU [43]	Herb or subshrub
<i>Pfaffia cipoana</i> Marchior. et al.	Brazilian Cerrado endemic (Itambé do Mato Dentro - MG)		Subshrub

<i>Pfaffia denudata</i> (Moq.) Kuntze	Cerrado exclusive, endemic to Brazil		Herb, subshrub, shrub
<i>Pfaffia elata</i> R.E.Fr.	Cerrado exclusive, endemic to Brazil		Subshrub
<i>Pfaffia glabrata</i> Mart.	Cerrado exclusive		Herb, subshrub; called "corango- sempreviva" [45]
<i>Pfaffia glomerata</i> (Spreng.) Pedersen	Cerrado	VU [44]	Herb, subshrub; called "anador, canela-velha, ginseng-brasileiro, finseng, páfia, paratudo, corango- sempreviva"; folk medicinal plant, most of its chemical constituents are known and roots of this plant are already used by pharmaceutical companies; butanolic extract showed antihyperglycemic potential in vivo; C <sub>3</sub> photosynthesis physiology and structure [38,40,42,45,60]
<i>Pfaffia gnaphaloides</i> (L.f.) Mart.	Cerrado	VU [44]	Herb, subshrub, called "corango-de- seda", C <sub>3</sub> photosynthesis physiology and structure [38,42,45]
<i>Pfaffia hirtula</i> Mart.	Cerrado exclusive, endemic to Brazil		Herb, subshrub
<i>Pfaffia jubata</i> Mart.	Cerrado, endemic to Brazil		Herb, subshrub; called "marcela- branca, marcela-do-campo, marcela- do-cerrado" and kytertenim; roots are used in folk medicine against intestinal problems [39,49]
<i>Pfaffia minarum</i> Pedersen	Cerrado exclusive, endemic to Brazil	VU [43]	Subshrub
<i>Pfaffia rupestris</i> Marchior. <i>et</i> <i>al.</i>	Brazilian Cerrado endemic (Rio Pardo de Minas - MG)		Subshrub
<i>Pfaffia sarcophylla</i> Pedersen	Brazilian Cerrado endemic (Niquelândia - GO)		Subshrub; nickel hyperaccumulator, it is one of the first species to recolonize the ground with high concentrations of total Ni in the soil (>1%) [61]
<i>Pfaffia sericantha</i> (Mart.) Pedersen	Cerrado, endemic to Brazil		
<i>Pfaffia siqueiriana</i> Marchior. & Miotto	Cerrado, endemic to Brazil		Subshrub
<i>Pfaffia townsendii</i> Pedersen	Cerrado, endemic to Brazil	VU [43]	Subshrub; C <sub>3</sub> photosynthesis physiology and structure [38,42]
<i>Pfaffia tuberculosa</i> Pedersen	Cerrado, endemic to Brazil		Herb, subshrub
<i>Pfaffia tuberosa</i> (Spreng.) Hicken	Cerrado		Herb, subshrub; called "corango-de- batata" [45]
<i>Pfaffia velutina</i> Mart.	Cerrado exclusive, endemic to Brazil		Subshrub

<i>Pseudoplantago friesii</i> Suess		PE [44]	Popular name is "caruru-açu" [45]
<i>Quaternella confusa</i> Pedersen	Cerrado exclusive, endemic to Brazil		Shrub
<i>Quaternella ephedroides</i> Pedersen	Cerrado, endemic to Brazil		Shrub
<i>Quaternella glabratoides</i> (Suess.) Pedersen	Endemic to Brazil	EN [44]	Subshrub; called "corangão" [45]
<i>Xerosiphon angustiflorus</i> (Mart.) Pedersen	Cerrado, endemic to Brazil		Subshrub
<i>Xerosiphon aphyllus</i> (Pohl ex Moq.) Pedersen	Cerrado, endemic to Brazil		Subshrub

**Notes:** The species threat level category is the same as that used in the IUCN Red List: CR (critically endangered), EN (endangered), EX (extinct), LC (Least Concern) and VU (vulnerable).

**Table 1.** Amaranthaceae species found in Brazil, identifying those endemics to Brazil and the ones found in the Neotropical Savannah (Cerrado), level of threat, habit, popular name (mostly in Portuguese) and some of the knowledge about the species.

### 3.1. Morphology, taxonomy challenge and species list of Brazilian Amaranthaceae

Taxonomy is the science that aims to identify and characterize species. It includes the study of the plant's behaviour in nature and is based on plant morphology. The use of other data, such as anatomy studies, genetic characters, ecology and geographic pattern, aims to include and define affinities and parental relations among plant groups. Only by knowing the species is it possible for Botany to contribute to other scientific areas, including to the conservation of species *in situ*, not only of plants but also of animals.

It is not easy to correctly identify Brazilian Amaranthaceae species. Different species can be very alike in habit and vegetative morphology. The correct identification depends almost exclusively on some flower details, whose small dimensions make it especially difficult to work in the field, demanding a highly specialized work, only partially carried out for this family (15-22).

Brazilian species of this family are predominantly herbs, shrubs or climbing plants. They can be annual or perennial, with erect, prostrate, decumbent or scandent stem. In species from the Neotropical Savannah or from rocky fields, the underground organ is thickened and composed of roots and a xylopodium – a portion of the subterranean system which is responsible for the re-sprouting after a fire or other environmental stress [62]. The leaf arrangement can be opposite, alternate or with a basal aggregation of leaves. Leaves are exstipulate, glabrous or pubescent, with entire lamina and margins. Inflorescences can be cymoses, in spikes, in heads, corymboses or paniculates, axillary or axial. Flowers are bisexual or monoecious and small. The perianth is undifferentiated, actinomorphic, with five distinct or partially connated sepals. Flowers are associated with dry and papery bracts. Fruits are dry, usually a single-seeded achene or capsules with few seeds [15-22]. A short list of the most important Brazilian Herbaria to visit in order to study Amaranthaceae taxonomy is presented on Table 2 and the literature used to identify the species of this family is presented in Table 3.

Index	Herbarium Name	Institution and municipality
ALCB	Herbário da Universidade Federal da Bahia	UFBA/Campus de Ondina, Salvador, Bahia, Brazil
BHCB	Herbário da Universidade Federal de Minas Gerais	UFMG, Belo Horizonte, Minas Gerais, Brazil
BOTU	Herbário da Universidade Estadual Paulista	UNESP, Botucatu, São Paulo, Brazil
CEN	Herbário da EMBRAPA Recursos Genéticos e Biotecnologia	EMBRAPA/CENARGEN, Brasília, Distrito Federal, Brazil
CEPEC	Herbário do Centro de Pesquisas do Cacau	CEPEC, Itabuna, Bahia, Brazil
CESJ	Herbário da Universidade Federal de Juiz de Fora	UFJF, Juiz de Fora, Minas Gerais, Brazil
CPAP	Herbário do Centro de Pesquisas Agropecuárias do Pantanal	CPAP, Corumbá, Mato Grosso do Sul, Brazil
ESA	Herbário da Universidade de São Paulo	ESALQ/USP, Piracicaba, São Paulo, Brazil
GUA	Herbário Alberto Castellanos	FEEMA/INEA, Rio de Janeiro, Rio de Janeiro, Brazil
HTO	Herbário da Universidade Federal do Tocantins	UFTO, Porto Nacional, Tocantins, Brazil
HUEFS	Herbário da Universidade Estadual de Feira de Santana	UFES, Feira de Santana, Bahia, Brazil
IAC	Herbário do Instituto Agrônomo de Campinas	IAC, Campinas, São Paulo, Brazil
IBGE	Herbário da Reserva Ecológica do IBGE	IBGE/RECOR, Brasília, Distrito Federal, Brazil
JPB	Herbário da Universidade Federal da Paraíba	UFPB, Cidade Universitária, João Pessoa, Paraíba, Brazil
MBM	Herbário do Museu Botânico Municipal	Prefeitura Municipal/SMA, Curitiba, Paraná, Brazil
PACA	Herbarium Anchieta	Instituto Anchietano de Pesquisas/UNISINOS, São Leopoldo, Rio Grande do Sul, Brazil
RB	Herbário do Jardim Botânico do Rio de Janeiro	JBRJ, Rio de Janeiro, Rio de Janeiro, Brazil
SP	Herbário do Instituto de Botânica	Secretaria de Meio Ambiente, São Paulo, São Paulo, Brazil
SPF	Herbário da Universidade de São Paulo	USP, São Paulo, São Paulo, Brazil
UB	Herbário da Universidade de Brasília	UnB, Brasília, Distrito Federal, Brazil
UEC	Herbário da Universidade Estadual de Campinas	UNICAMP, Campinas, São Paulo, Brazil
UFG	Herbário da Universidade Federal de Goiás	UFG, Goiânia, Goiás, Brazil
VIC	Herbário da Universidade Federal de Viçosa	UFV, Viçosa, Minas Gerais, Brazil

**Notes:** Herbaria are cited according to the Index Herbariorum and all of them have good collections of Amaranthaceae. The Institution/city where the Herbaria are located is also referred.

**Table 2.** List of the most important Herbaria references for researchers interested in studying the Brazilian Amaranthaceae

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- [15-21] Revisions of Brazilian *Froelichia*, *Froelichiella*, *Hebanthe* and *Pfaffia*; species list and phytogeography
  - [22-25] Revision of Brazilian *Gomphrena*; species list na phytogeography
  - [45] Amaranthaceae from Santa Catarina State, Brazil
  - [63] Restoring the *Hebanthe* genera
  - [64,65] Brazilian Amaranthaceae species and the Family in the World
  - [66] Revision of Amaranthaceae in the World
  - [67-71] Studies in South American Amaranthaceae
  - [72] Amaranthaceae in Flora Brasiliensis
  - [73] Studies of *Pfaffia* and *Alternanthera* genera
  - [74,75] Amaranthaceae in Central and South America
  - [51,52,56,76] Amaranthaceae from Rio Grande do Sul State, Brazil
- 

**Note:** These references are ordered by author and should be consulted in order to identify Brazilian species correctly.

**Table 3.** List of the most important bibliographical references for researchers interested in studying the Brazilian Amaranthaceae

The Reserva Particular do Patrimônio Natural (RPPN) Cara Preta, in Alto Paraíso, Goiás State, is a good representative of Neotropical Savannah vegetation, at about 1,500 meters of altitude and showing rocky slopes with Cerrado *sensu stricto* (Figure 1), grassland with scattered scrubs and few trees and grassland with few scrubby plants and no trees (Figure 2). The *Pfaffia* genus was restricted to a rocky slope and the other species were found in a level field of sandy soils, usually covered by Poaceae and Cyperaceae. It was very difficult to find all the species. It was only possible because of frequent visits to RPPN Cara Preta, using GPS to mark the local after finding any probable member of the family in order to be able to accompany them until the flowering stage. The area was monitored for one and a half year and only *Gomphrena hermogenesii* J.C. Siqueira and *Pfaffia townsendii* Pedersen (Figure 3) were localized, the first one always in vegetative stage. A key event to help finding all six species was a fire that burned out the vegetation in August of the year 2008: without the competition of the grasses, the Amaranthaceae species regrew and flowered rapidly, in order to spread their seeds before the grasses could fully recover (Figures 4-8).

*Pfaffia townsendii* is a shrub species with persistent aerial portions that flowers throughout the year (Figure 3). The herb *G. hermogenesii* is endemic to Chapada dos Veadeiros and also has permanent aerial portions (about 10-20 cm high), but it was commonly found in vegetative stage under the grass leaves; its flowering stage was stimulated by fire (Figure 4). *Froelichiella grisea* R.E.Fr. (Figure 5), *G. lanigera* Pohl. ex Moq. (Figure 6), *G. prostrata* Mart. (Figure 7) and *P. gnaphaloides* (L.f.) Mart. (Figure 8) species were recorded in the flowering stage at RPPN Cara Preta around 20 days after a fire that burned out all the vegetation in the area, which is evidence of the pirophytic behaviour of most Neotropical Savannah Amaranthaceae. Five of these species had never been recorded in this RPPN before and one of them was last recorded in 1966 (*F. grisea*), according to Herbaria data. Figures 1-8 are reproduced [77] with the authorization of the Biota Neotropica Editor, Dr. Carlos Joly.



**Figure 1.** (Figures 1-8) Photographs of the environment and of the studied species at Reserva Particular do Patrimônio Natural (RPPN) Cara Preta, Alto Paraíso, Goiás State, Brazil. **Fig. 1.** Rocky slope where were found the species *Pfaffia townsendii* Pedersen and *P. gnaphaloides* (L. f.) Mart. **Fig. 2.** Humid rocky grassland where were found the species *Froelichiella grisea* R.E.Fr., *Gomphrena hermogenesii* J.C. Siqueira, *G. lanigera* Pohl. ex Moq. and *G. prostrata* Mart. **Fig. 3.** *P. townsendii*. **Fig. 4.** *G. hermogenesii*. **Fig. 5.** *F. grisea*. **Fig. 6.** *G. lanigera*. **Fig. 7.** *G. prostrata*. **Fig. 8.** *P. gnaphaloides*.

Five species are herb to subshrub, and only *P. townsendii* is a shrub (Figure 3). Well-developed tuberous subterranean systems were found in *F. grisea* (Figure 5) and *G. hermogenesii*, while in *G. lanigera* and *P. gnaphaloides* the underground organ was less developed, also tuberous. *G. prostrata* and *P. townsendii* presented a well-developed and lignified underground organ. Leaves of *F. grisea* and *G. hermogenesii* are opposite and alternate in the other studied species. *F. grisea* and *G. lanigera* can present a basal aggregation of leaves. Leaves are always tomentose, with exception of the adaxial face of *F.*



*grisea*, which can be glabrous. Inflorescence is axial in all these species, spikes in *F. grisea* and *G. lanigera* and heads in the other studied species. Flowers are yellowish in *F. grisea*, *G. hermogenesii* and *G. lanigera*, with a tendency to turn red in the first and last one. In *G. prostrata*, *P. gnaphaloides* and *P. townsendii* flowers are white, turning beige in the last species. All the species have flowers associated with dry and papery bracts that persist alongside their dry fruits, usually a single-seeded achene, favouring anemocoric dispersion.

The fastest lifespan was observed in *G. lanigera*, which took around 20 days to regrowth and finish the flowering phase. In Figure 6, *G. lanigera* was about 20 days old and fruits were almost mature, indicating proximity to the seed dispersal phase. *Pfaffia townsendii* alone showed behaviour that was independent of fire, since even *G. hermogenesii* only flowered after being burned to the ground and regrowing from its xylopodium. The other four species were found after the occurrence of fire, all of them in the flowering stage.

In the Taxonomy and Morphology areas, studies of the genera *Achyranthes*, *Alternanthera*, *Amaranthus*, *Blutaparon*, *Celosia*, *Chamissoa*, *Chenopodium*, *Cyathula*, *Iresine*, *Lecosia*, *Pederseniana*, *Pseudoplantago*, *Quaternella* and *Xerosiphon* still need to be done, not only covering the revision of the Brazilian species, biogeography and morphological evolution, but also molecular biology to establish synonyms and to delimit variations among individuals of each species.

### 3.2. Leaf anatomy of Amaranthaceae species

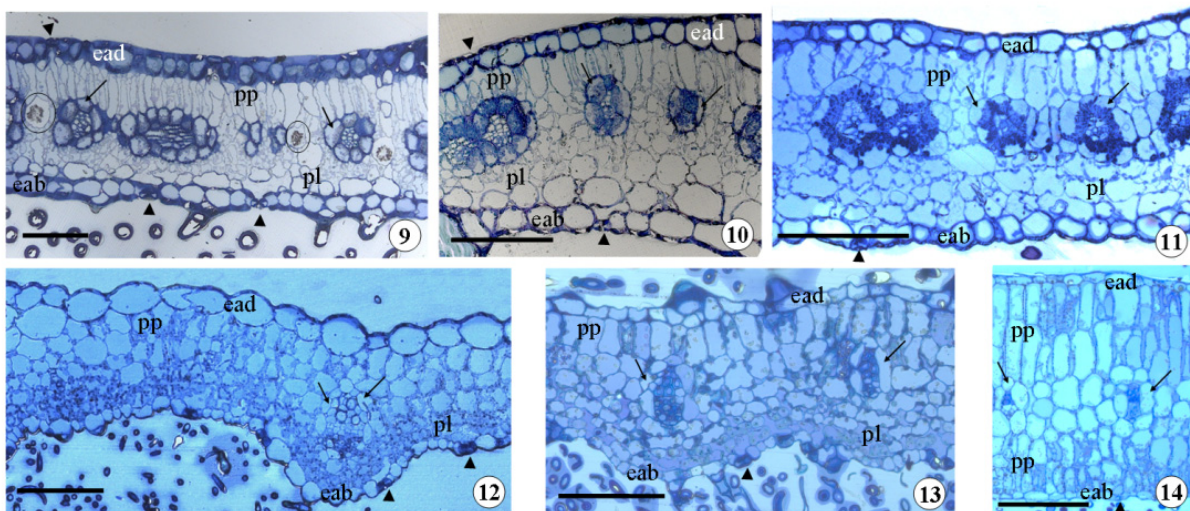
Leaves of the six studied species have anatomical variation among the genera and are more similar between species of the same genus. Transverse sections show that *G. hermogenesii* (Figure 9), *G. lanigera* (Figure 10) and *G. prostrata* (Figure 11) have large nonglandular trichomes covering the single layered epidermis, dorsiventral mesophyll with upper palisade parenchyma and spongy parenchyma near the lower epidermis. All these three species are amphistomatic, and a complete well-developed parenchymatous sheath with thicker cell walls surrounds the vascular bundles (Kranz cells), in which starch accumulates. Calcium oxalate druses were found in the mesophyll. The leaf anatomy of the three *Gomphrena* spp. is compatible with the C<sub>4</sub> photosynthesis pathway.

*Pfaffia gnaphaloides* (Figure 12) and *P. townsendii* (Figure 13) have more undulating surfaces and a thinner leaf blade in relation to the *Gomphrena* species. Trichomes are also more frequent and thinner and the mesophyll is dorsiventral. The parenchymatous sheath has thinner walls than the neighbouring cells in *Pfaffia* species. Both species had elevated stomata on the lower epidermis and only *P. gnaphaloides* had few stomata on the upper epidermis. Starch was distributed in all mesophyll cells and calcium oxalate druses were rare. The anatomy of *Pfaffia* spp. leaves is compatible with C<sub>3</sub> photosynthesis metabolism.

*Froelichiella grisea* (Figure 14) has the only isobilateral mesophyll among the studied species, with palisade parenchyma near both upper and lower epidermis. Palisade cells are shorter near the lower epidermis. The parenchymatic vascular bundle is not conspicuous and organelles in these cells are positioned towards the outer cell walls, in

the same way as they are found in the other mesophyll cells. Calcium oxalate druses were more common near the midrib, and the reaction to starch was similar to that of all the mesophyll cells. Its leaf anatomy is compatible with  $C_3$  photosynthesis metabolism. Figures 9-14 [77] were reproduced with the authorization of the Biota Neotropica Editor, Dr. Carlos Joly.

*Gomphrena* trichomes are similar to the ones described for *G. arborescens* [32,54,78]. Although it is expected that stomata are reduced on the upper surface of land plants, the Cerrado *Gomphrena* species *G. arborescens*, *G. pohlii* and *G. virgata* have a similar number of stomata on both surfaces [78], subjecting them to a greater water loss, which is compensated by the well-developed subterranean systems that guarantee water supply during the lifespan of their leaves. The size and number of stomata on both leaf surfaces of *G. hermogenesii*, *G. lanigera* and *G. prostrata* is still to be verified, but simple observation indicates that it should be similar to the phenomena observed in the first cited species, since they also have a relatively well-developed subterranean system.



**Figure 2.** (Figures 9-14) Micrographies of the middle leaf transversal sections of the studied Amaranthaceae species. **Fig. 9.** *Gomphrena hermogenesii* - leaf blade thickness from medium to thick, dorsiventral mesophyll and complete parenchymatous bundle sheath, with thick cell walls and collateral vascular bundles. **Fig. 10.** *G. lanigera* - medium leaf blade, dorsiventral mesophyll and complete parenchymatous bundle sheath, with thick cell walls and collateral vascular bundles. **Fig. 11.** *G. prostrata* - thin to medium leaf blade, dorsiventral mesophyll and complete parenchymatous bundle sheath, with thick cell walls and collateral vascular bundles. **Fig. 12.** *Pfaffia gnaphaloides* - thin leaf blade, dorsiventral mesophyll, less defined parenchymatous bundle sheath and collateral vascular bundles. **Fig. 13.** *P. townsendii* - only hypostomatous leaf species, thin leaf blade, dorsiventral mesophyll, less defined parenchymatous bundle sheath and collateral vascular bundles. **Fig. 14.** *Froelichiella grisea* - thick leaf blade, isobilateral mesophyll with elongated palisade parenchyma under the adaxial epidermis, less defined bundle sheath and collateral vascular bundles. **Legend:** eab = abaxial epidermis; ead = adaxial epidermis; pl = spongy parenchyma; pp = palisade parenchyma; arrowhead = stoma; circle = druse; arrow = parenchymatous bundle sheath. Bar = 100  $\mu$ m.

The leaf anatomy of the three *Gomphrena* spp. is similar to that of *G. arborescens* L.f. [54], *G. cespitosa*, *G. dispersa*, *G. nitida*, *G. sonora* [79] and *G. conica*, *G. flaccida* [80] among others, most of them arranged in a *Gomphrena* atriplicoid-type of Kranz anatomy [81,82]. As expected, there was no significant variation in these species' leaf anatomy due to the life cycle stage, although older leaves collected during the vegetative stage have a thicker cuticle covering both epidermis surfaces, especially in *G. hermogenesii* species. The leaf anatomy observed in the two *Pfaffia* spp. is similar to that described in *P. jubata* [83], which also lacks the Kranz anatomy. There is no previous study about the anatomy of *F. grisea* leaves and its genus is monoespecific.

There are still a number of studies to be done in the field of anatomy and histology of Brazilian Amaranthaceae plants. Most of the medicinal species need to be analyzed and validated for their use as drugs, including anatomic description and an investigation of the secondary compounds of the used organs, by histology and by chromatography. Due to the difficulties in correctly identifying the species in the field, anatomical and morphological markers should be defined to guarantee these species' identity even during the vegetative stage. Besides that, anatomical studies can improve the taxonomy data and explain some morphological characters of this plant family, like the anatomical variations in the leaf that are connected to photosynthesis, or the secondary thickening and xylopodium development in underground organs, which is a character for the Cerrado species. The anatomy of few Brazilian Amaranthaceae species has been described, with the exception of some from the *Gomphrena* and *Pfaffia* genera [83-86].

### 3.3. Leaf ultrastructure of Amaranthaceae species

Leaves of the six studied species have less ultrastructural variation among the species of the same genus. *Froelichiella grisea* organelles are equally distributed among chlorenchyma tissues, usually near the cell walls. The chloroplasts of this species are always granal (Figure 15), even in the vascular cells, with large starch granules (usually one or two per organelle) in all tissues. Plastoglobuli are small and less numerous in mesophyll chloroplasts (Figure 15), but guard cell chloroplasts usually have just one large plastoglobulus and less conspicuous grana. Mitochondria and peroxisomes (Figure 15) were found in mesophyll and bundle sheath cells. Leaf ultrastructure is compatible with C<sub>3</sub> photosynthesis metabolism.

Mesophyll cell chloroplasts of *Gomphrena* species have conspicuous grana, rare starch granules and variable size of plastoglobuli: *G. hermogenesii* has larger ones in relation to *G. lanigera* and *G. prostrata*. Bundle sheath chloroplasts are completely devoid of grana or have few stacked thylakoids (Figure 16) in all studied *Gomphrena* species, but always have large starch granules and plastoglobuli. The larger the starch granules, the more deformed the chloroplasts' typical lens shape, as shown in *G. hermogenesii* (Figure 16). Mitochondria are usually numerous in bundle sheath cells and are always near chloroplasts, grouped next to the inner cell wall (towards the vascular bundle). Peroxisomes are rare, and a few

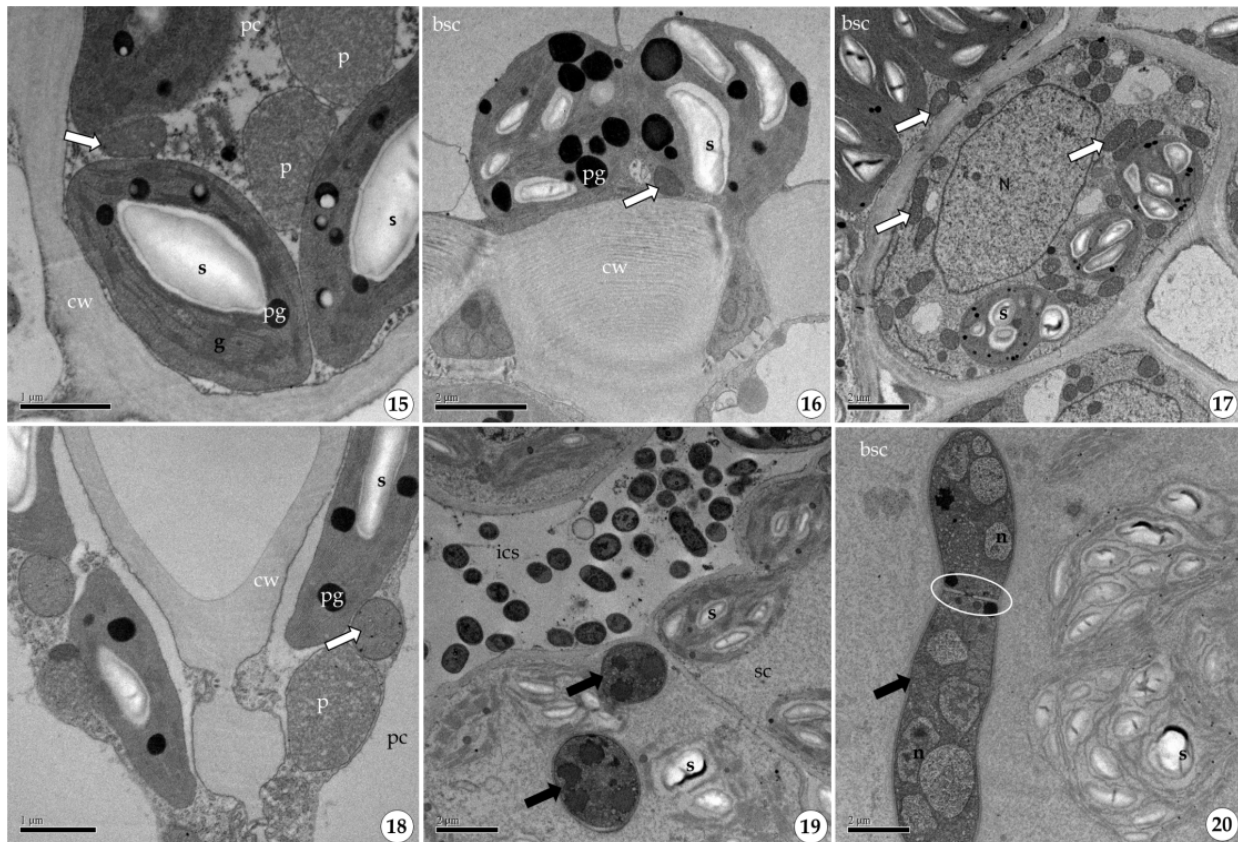
were observed near chloroplasts in palisade and spongy parenchyma cells, but not in the bundle sheath cells. Phloem companion cells are mitochondria-rich in all *Gomphrena* species, as shown in *G. prostrata* (Figure 17). The presence of dimorphic chloroplasts, disposition of the organelles and the occurrence of Kranz syndrome seen in the leaf anatomy indicate that the C<sub>4</sub> photosynthesis pathway operates in the three studied *Gomphrena* spp.

*Pfaffia* species organelles are equally distributed among chlorenchyma tissues, usually near the cell walls. *Pfaffia* chloroplasts are granal even in the vascular cells, showing large starch granules and a similar size in all mesophyll cells, as can be observed in the palisade parenchyma of *P. townsendii* (Figure 18). Mitochondria and peroxisomes are common near chloroplasts (Figure 18). Phloem companion cells are mitochondria-rich and chloroplasts are smaller and granal, as in the other species of this study. Along with the aspects of *Pfaffia* anatomy described previously, their ultrastructure is compatible with the C<sub>3</sub> photosynthesis pathway.

*Pfaffia gnaphaloides* (Figure 19) and *G. hermogenesii* (Figure 20) leaves, collected during the flowering stage, were colonized by two distinct forms of microorganisms: (i) a smaller organism was found in the intercellular spaces (ics) of the spongy parenchyma (Figure 19); (ii) a larger and distinctly eukaryotic organism was found within distinct cells, with some morphological alterations suggesting an infectious process (Figures 19-20).

The external envelopae membranae system of the chloroplasts is disrupted in infected cells (Figure 20) and a size reduction was observed in the chloroplast plastoglobuli. All morphological characteristics observed in the intracellular microorganism suggest that it should be an obligate biotroph endophytic fungus belonging to the Ascomycete division (Figure 20). The invading fungus may be using the plastoglobuli lipids as its primary source of carbon and energy; the reduction of the plastoglobuli could also be due to its mobilization by the host plants in response to the stress caused by these biotic interactions. The complete identification of the fungus and its effect on the plants depends on its isolation from the environment/hosts and complementary studies.

The rare peroxisomes in *Gomphrena* spp. leaf cells and their presence among all chlorenchyma tissues of the *Pfaffia* spp. leaf cells is compatible with their possible photosynthesis metabolisms. Along with the presence of Kranz syndrome and dimorphic chloroplasts, the absence of peroxisome indicates that *Gomphrena* spp. perform photosynthesis via the C<sub>4</sub> pathway. In *Gomphrena* species, CO<sub>2</sub> concentration in the bundle sheath cells must be efficient, leading to a significant reduction in the oxygenase function of its RuBisCO enzyme. This leaves the species virtually free of the photorespiration process, aided by the large walls of the bundle sheath cells. Although a carbon isotope ratio study [38] indicates that *G. hermogenesii* is not a C<sub>4</sub> species, this species also has Kranz anatomy and ultrastructure compatible with C<sub>4</sub> metabolism, as do all the other studied *Gomphrena* spp. [42]. The distribution of its key photosynthetic enzymes will be carried out using immuno-cytochemistry, in our laboratory, in order to complete these data.



**Figure 3.** (Figures 15-20) Citological aspects of Amaranthaceae species as seen through a Transmission Electron Microscope. **Fig.15.** *Froelichiella grisea* palisade parenchyma cell. **Fig. 16.** *Gomphrena hermogenesii* bundle sheath cell. **Fig. 17.** *G. prostrata* phloem companion cell and bundle sheath cell on top. **Fig. 18.** *Pfaffia townsendii* palisade parenchyma cells. **Fig. 19.** *P. gnaphaloides* spongy parenchyma cells and invading microorganisms (black arrows). **Fig. 20.** *G. hermogenesii* bundle sheath cell and the invading Ascomycete fungus (black arrow) and the disrupted chloroplasts with smaller plastoglobuli. **Legend:** black arrow = invading organism; white arrow = mitochondria; ellipsis = septum with a simple pore; bsc = bundle sheath cell; cw = cell wall; ics = intercellular space; n = nucleus of the microorganism; N = nucleus of the plant species; p = peroxisome; pc = palisade parenchyma cell; pg = plastoglobulus in a chloroplast; s = starch granule in a chloroplast; sc = spongy parenchyma cell.

#### 4. Conclusions and perspectives

This chapter presents data on Amaranthaceae species, with no pretension to explain the full potential of this plant family for scientific studies, but rather to provide a basic tool for those interested in amplifying studies on the species of this family. Based on our results, we are convinced of the importance of studying this family further, not only as a tool in the better preservation of endemic species, but also to explore its undoubted economic importance more fully. Basic research is still needed, with the aim of applying knowledge on these species to technological advances, especially in growing crops - since  $C_4$  species have a faster metabolism and growing capacity, as observed in the species found in the RPPN Cara Preta - and to explore medicinal molecules of these plants.  $C_4$  species are also important to balance  $CO_2$  in the atmosphere because of their efficiency in the transformation of carbon into

biomass; in Cerrado Amaranthaceae species, this storage is basically underground in their well-developed subterranean roots and xylopodium.

The number of medicinal plants among the Brazilian Amaranthaceae species may well be higher than already reported (Table 1), because Cerrado inhabitants are particularly interested in the highly developed subterranean systems of some medicinal species [34,49,58] which can be collected at any time of the year, even from species whose aerial portions are not persistent. Due to the morphological similarity among Amaranthaceae species in the Neotropical Savannah, their collectors can easily mistake one species for another during the vegetative stage, which confirms the need for further and more complete studies of the known medicinal and endangered species, at least.

Preparation of plant samples for transmission electron microscopy also proved to be useful in studying the morphology of fungi inside plant cells, as well as aspects of host-parasite interaction. This kind of study could be recommended for plants considered toxic to herbivores and to any medicinal plant consumed by humans, in order to give more information about the real source of poisoning or medicinal effect and for fine quality control. In both studied species (*G. hermogenesii* and *P. gnaphaloides*) the external macro aspects of the plants did not indicate the presence of the endophytic fungus.

RPPN Cara Preta is a small Private Conservation Unit (only 1.5% of the area of the Chapada dos Veadeiros National Park, a government-preserved area of 65,038 hectares). Both Conservation Units are separated only by a road, in Alto Paraíso municipality of Goiás State, Brazil. The latter site is registered by UNESCO as a natural protected Cerrado zone. RPPN Cara Preta has 245 species representing 47 family plants [75,86], which is 9.2% of the 2,661 plant species of Chapada dos Veadeiros [87], a good diversity of plants in relation to the occupied area. There are six Amaranthaceae species in the RPPN – 25% of the 27 species found in the National Park [87]. Considering that the RPPN Cara Preta Utilization Planning Report [86] indicated the presence of three endemic species, plus two Amaranthaceae species not reported initially [75], this Conservation Unit has 2% of endemic species – which is more than expected. According to [2,12], the Cerrado Biome is one of the priority hotspots for conservation because it has, among others, 4,400 endemic plants (1.5% of the Earth's 300,000 species). The Amaranthaceae family in RPPN Cara Preta can be considered a taxon indicator of the good diversity of the Neotropical Savannah. This taxon could be considered a plant diversity indicator in other works on flora in open areas of the Cerrado Biome. Because of the predominant habit (herbs and shrubs) and survival strategies, the presence of species from this family among the collected species clearly indicates a well performed collection effort.

There are a number of important factors indicating that this plant family deserves more studies for a greater understanding by researchers working in Brazil, and we recap them as follows: the Cerrado Biome holds 98 of the 146 Brazilian Amaranthaceae species (almost 70% of the total species) (Table 1); their pyrophytic behavior and survival strategies (fast regrowth and seed dispersal before the complete recovery of grasses after fire) are coherent

with the Biome's characteristics; their morphology shows exceptional adaptation to the seasonal climate and open areas (hairy aerial portions, partial or total loss of the aerial portions during the dry season, well-developed underground system with xylopodium, dry fruit dispersal by wind); their metabolism (evolution of C<sub>4</sub> and intermediary C<sub>3</sub>-C<sub>4</sub> photosynthesis) may have importance for biomass conversion and CO<sub>2</sub> balance; and, finally, many of these plants are already used in medicines by Cerrado inhabitants and there may be much wider medicinal potential in other species of this family.

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