

CECÍLIA TAVARES NEVES BRAGA

**PHOTOSYNTHETIC STRATEGIES AND PHOSPHORUS USE EFFICIENCY OF
ASTERACEAE SPECIES IN FERRUGINOUS *CAMPO RUPESTRE***

Dissertation submitted to the Management and Conservation of Natural and Agrarian Ecosystems Graduate Program of the Universidade Federal de Viçosa in partial fulfillment of the requirements for the degree of *Magister Scientiae*.

Adviser: Eduardo Gusmão Pereira

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To God and to my parents.

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To God, who capacited and brought me so far; I would be nothing without Him.

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*“For we know that the whole creation groaneth and
travaileth in pain together until now – Romans 22:8”*
(Holy Bible, KJV)

ABSTRACT

BRAGA, Cecília Tavares Neves, M.Sc., Universidade Federal de Viçosa, October, 2022. **Photosynthetic strategies and Phosphorus use efficiency of Asteraceae species in *Campo Rupestre***. Adviser: Eduardo Gusmão Pereira.

The threatened Brazilian ferruginous *campo rupestre*, also known as canga is a fire-prone and nutrient-impoor landscape, with high diversity of functional traits. The plant species found in this environment are generally characterized as slow growth, higher leaf mass per area (LMA) and leaf dry mass content (LDMC), limited more by phosphorus (P) than nitrogen (N) when compared to other grasslands. In chapter one, a bibliographical review was conducted in order to establish the current state of P nutrition in those landscapes. In chapter two, the field work, conducted in the *Serra da Calçada* (MG, Brazil) during the rainy season of 2021 and 2022 was described. This work's main goal was to understand the diversity of strategies to acquire and use P in Asteraceae species from ferruginous *campo rupestre*, regarding their functional traits and photosynthetic performance. To accomplish this, five Asteraceae species were selected: *Dasyphyllum velutinum*, *Baccharis reticularia*, *Lychnophora pinaster*, *Trixis vauthieri* and *Hololepis pedunculata*. A high diversity of functional traits, such as LMA, LDMC, foliar area and nutrient concentration was found. Some of which can be directly and indirectly related to the diversity of strategies to better absorb and use P. The species showed, in general, 70% mycorrhizal colonization and retranslocation rates of 77% and 65% for P and N, respectively. Even though these species have a diversity of functional traits, their photosynthetic P-use efficiency and P fractions (metabolic, nucleic acid, lipidic and residual) showed no significant difference. Finally, it was concluded that the diversity of functional traits found in Asteraceae species is most likely related by its phylogeny and the history of evolution of Asteraceae in the ferruginous *campo rupestre*. The ecological filters imposed by nutrient availability pressure during their evolutionary path have converged into a similar physiological performance.

Keywords: P-fractions. Canga. Nutritional strategies. Photosynthetic P-use efficiency. Mycorrhiza. Convergence.

RESUMO

BRAGA, Cecília Tavares Neves, M.Sc., Universidade Federal de Viçosa, outubro de 2022. **Photosynthetic strategies and phosphorus use efficiency of Asteraceae species in *Campo Rupestre***. Orientador: Eduardo Gusmão Pereira.

O campo rupestre ferruginoso ameaçado de extinção, também conhecido como canga, é uma paisagem propensa ao fogo e pobre em nutrientes, com alta diversidade de características funcionais. As espécies vegetais encontradas neste ambiente são geralmente caracterizadas como de crescimento lento, maior massa foliar por área (LMA) e teor de massa seca foliar (LDMC), limitadas mais pelo fósforo (P) do que pelo nitrogênio (N) quando comparadas a outros ambientes savânicos. No capítulo um, foi realizada uma revisão bibliográfica a fim de estabelecer o estado atual da nutrição fosfatada nessas paisagens. No capítulo dois, foi descrito o trabalho de campo, realizado na Serra da Calçada (MG, Brasil) durante o período chuvoso de 2021 e 2022. Este trabalho teve como objetivo principal compreender a diversidade de estratégias de aquisição e uso de P em espécies de Asteraceae de campo rupestre ferruginoso, quanto às suas características funcionais e desempenho fotossintético. Para tanto, cinco espécies de Asteraceae foram selecionadas: *Dasyphyllum velutinum*, *Baccharis reticularia*, *Lychnophora pinaster*, *Trixis vauthieri* e *Hololepis pedunculata*. Foi encontrada uma alta diversidade de características funcionais, como LMA, LDMC, área foliar e concentração de nutrientes. Algumas delas podem estar direta e indiretamente relacionadas à diversidade de estratégias para melhor absorção e utilização do P. As espécies apresentaram, em geral, 70% de colonização micorrízica e taxas de retranslocação de 77% e 65% para P e N, respectivamente. Apesar dessas espécies apresentarem uma diversidade de características funcionais, sua eficiência fotossintética de uso de P e frações de P (metabólico, ácido nucléico, lipídico e residual) não apresentaram diferença significativa. Por fim, concluiu-se que a diversidade de características funcionais encontradas nas espécies de Asteraceae está provavelmente relacionada à sua filogenia e à história de evolução das Asteraceae no campo rupestre ferruginoso. Os filtros ecológicos impostos pela pressão de disponibilidade de nutrientes ao longo do seu caminho evolutivo convergiram para um desempenho fisiológico semelhante.

Palavras-chave: Frações de P. Canga. Estratégias nutricionais. Uso fotossintético eficaz de P. Micorriza. Convergência.

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1. GENERAL INTRODUCTION

Several ecosystems worldwide are considered to be nutrient-impoverished and yet be landscapes of high biodiversity and endemism rates (Hopper, Silveira & Fiedler, 2016; Lambers et al., 2011). This description fits the concept of biodiversity hotspots (Myers et al., 2000): ecosystems that comprise a rich and endemic biodiversity heavily threatened by a variety of anthropic activities and their consequences (such as climatic change), with a high risk of extinction. Over 30% of biodiversity hotspots are considered to be old, climatically buffered, infertile landscapes (OCBILs), such as the Cape region in South Africa and southwestern Australia (Hopper et al., 2016).

Although this type of landscape is scattered along the globe, there are some similarities in both functional and morphological traits in the plants that inhabit them. Nutrient deficient soils are one of the environmental filters that show a pattern of strategies regarding the nutrient use and acquisition in OCBILs; especially P deficiency (Hopper et al., 2021).

Because of the particularities that brought these ecosystems to be in a delicate balance, the impacts of anthropogenic pressures and climate change brought upon them is too great (Fernandes et al., 2020). The urban expansion, plants' extraction and mining activities are a few of the many examples of how humans directly impact landscapes such as these. Indirectly, the industrial revolution and all the events that unfolded after, contributed to catastrophic changes in environments close and near to the change starting point, particularly over the temperature of the planet (Heilmeier, 2019). With that, the ecosystems face changes that fundamentally threaten their integrity regarding not only their biodiversity, but also the ecosystem services produced by them.

One of the biodiversity hotspots in Brazil is the Cerrado Domain (Batalha, 2011). Within Cerrado areas (but not only there), there is another type of phytophysionomy called *campo rupestre*, a grassland landscape very similar to the *cerrado rupestre*. However, differently from the *cerrado rupestre*, the *campo rupestre* is mostly characterized by sparse shrubs with few trees (Pereira & Fernandes, 2022). Within the threatened *campo rupestre* area, there is in Minas Gerais an economic region called Iron Quadrangle which hosts the ferruginous *campo rupestre*, also called *canga*. It is a landscape of high economic interest and heavily threatened by the mining activities conducted in the area. Despite the fact that ferruginous are often considered an inauspicious environment for many plants to thrive, there is a great biodiversity rate of

both plants and animals. More than that, there is a high rate of endemic species in ferruginous *campo rupestre*, that present morphophysiological and functional adjustments to thrive in these stress-prone conditions (Carmo & Jacobi, 2013; Rios et al., 2022), even though they occupy the same ecological niche of low P in the soil.

Accompanying the high biodiversity rate, there is a high diversity of functional traits in plants species in the *canga* vegetation. Because the soil is nutrient impoverished, there are morphological and physiological adaptations displayed such as narrow leaves, greater sclerophylly, greater amplitude of foliar areas, slow growth, accumulation of nutrients in the tissues, among others (Negreiros et al., 2014; Rios et al., 2022). There are many botanical families that inhabit this landscape, the richest ones being the Poaceae, Asteraceae and Fabaceae (Viana & Lombardi, 2007). The Asteraceae family is representative of this environment and also presents a wide variation of functional traits.

Little is known about the physiological strategies of Asteraceae species in response to the poor nutrient availability in the soil in nutrient-impooverished landscapes. We do know, however, that mycorrhizal association, nutrient allocation and even translocation (Jacobi & Carmo, 2008; Mo et al., 2019) are among them. It is crucial to understand these strategies in order to better select native species to be used in rehabilitation and restoration techniques in mining degraded areas, such as the *canga* (Jacobi & Carmo, 2008).

This present work aims to contribute in this regard, offering literature ground to future projects in the areas of biodiversity conservation, ecosystems' ecology and restoration of degraded areas.

REFERENCES:

Batalha, M. A. (2011). The Brazilian Cerrado is not a biome. *Biota Neotropica*, 11, 21-24.

Fernandes, G. W., Arantes-Garcia, L., Barbosa, M., Barbosa, N. P., Batista, E. K., Beiroz, W., ... & Silveira, F. A. (2020). Biodiversity and ecosystem services in the Campo Rupestre: A road map for the sustainability of the hottest Brazilian biodiversity hotspot. *Perspectives in Ecology and Conservation*, 18(4), 213-222.

Hopper, S. D., Silveira, F. A., & Fiedler, P. L. (2016). Biodiversity hotspots and Ocbil theory. *Plant and Soil*, 403(1), 167-216.

Hopper, S. D., Lambers, H., Silveira, F. A., & Fiedler, P. L. (2021). OCBIL theory examined: reassessing evolution, ecology and conservation in the world's ancient, climatically buffered and infertile landscapes. *Biological Journal of the Linnean Society*, 133(2), 266-296.

Jacobi, C. M., Carmo, F. F. D., & Vincent, R. D. C. (2008). Estudo fitossociológico de uma comunidade vegetal sobre canga como subsídio para a reabilitação de áreas mineradas no Quadrilátero Ferrífero, MG. *Revista Árvore*, 32, 345-353.

Mo, Q., Li, Z. A., Sayer, E. J., Lambers, H., Li, Y., Zou, B. I., ... & Wang, F. (2019). Foliar phosphorus fractions reveal how tropical plants maintain photosynthetic rates despite low soil phosphorus availability. *Functional Ecology*, 33(3), 503-513.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., Da Fonseca, G. A., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature*, 403(6772), 853-858.

Negreiros D, Le Stradic S, Fernandes GW, Rennó HC (2014) CSR analysis of plant functional types in highly diverse tropical grasslands of harsh environments. *Plant Ecol* 215:379–388.

Lambers, H., Brundrett, M. C., Raven, J. A., & Hopper, S. D. (2011). Plant mineral nutrition in ancient landscapes: high plant species diversity on infertile soils is linked to functional diversity for nutritional strategies. *Plant and Soil*, 348(1), 7-27.

Pereira, C. C., & Fernandes, G. W. (2022). Cerrado Rupestre is not campo rupestre: The unknown and threatened savannah on rocky outcrops. *Nature Conservation*, 49, 131-136.

Rios, C. O., Pimentel, P. A., Živčák, M., Brestič, M., & Pereira, E. G. (2022). Can ecological strategies be explained by photochemical efficiency in ironstone outcrops vegetation?. *Plant and Soil*, 1-16.

2. CHAPTER 1: Phosphorus use efficiency and acquisition strategies of plants in nutrient-impooverished landscapes: a bibliographic review

In nutrient impoverished environments there's a great diversity of plants, both physiologically and morphologically adapted. Regarding functional traits, there's a great amplitude in leaves' shape, area, thickness, leaf mass per area (LMA), leaf dry mass content (LDMC), and presence of trichomes (Guilherme Pereira et al., 2019).

Some nutrient-impooverished environments, when combined with other climatic and anthropogenic factors, are classified as "biodiversity hotspots" (Myers et al., 2000; Hopper et al., 2021). These are ecosystems and landscapes that are both rich in endemic biodiversity and constantly threatened by anthropic pressures and climatic change

This review will focus on how we can relate plants' diversity with functional traits and physiological strategies to better photosynthetic performances in environments of poor nutrient availability.

Functional traits and the leaf economic spectrum in plants from nutrient-impooverished landscapes

Plant functional traits are related to strategies and adaptations to environments with nutrient impoverished soils. Some plants in those environments with low P and N availability would present longer leaf lifespans (LLS), slower growth and higher nutrient retranslocation, especially of phosphorus (P) and nitrogen (N) (Osnas et al., 2013). Lower leaf mass per area (LMA) has also been proven to have direct positive relationships with the photosynthetic phosphorus use efficiency (PPUE) (Hiadaka & Kitayama, 2013). Native plants can also display these as a consequence of their evolution in environments that provide limited concentration of nutrients in the soil (Walker et al., 2017; Reich., 2014; Wright et al., 2004). Other functional traits related to nutrient-impooverished landscapes include lower N and P concentration on tissues, especially on leaves with high LLS, as well as higher N:P ratios in P impoverished soils (Wright et al., 2004, Mo et al., 2019).

Several of these strategies presented by endemic native plants to nutrient impoverished landscapes have deep relations with the whole ecosystem ecology (Westoby & Wright, 2006). These strategies may converge into a pool of adaptations that have ensured the survival and thrivingness of this plants in stress-prone

environments without coming to strain conditions (Veneklaas, et al., 2022; Westoby & Wright, 2006; Heilmeier, 2019).

One consequence of those deep relations between plants and landscapes is the provision of ecosystem services, such as recharge and water regulation, pollinators and natural pest controllers (Fernandes et al., 2020). However, there is a dramatic change in the interaction with environmental factors such as temperature and radiation due to the climatic changes (Heilmeier, 2019). This change may cause a severe imbalance in the ecosystems.

It has been suggested by Heilmeier (2019), that the better we understand the relationship between stress factors and functional traits of native endemic species, the better are the chances of preserving ecosystem services, areas and vegetation as climate change advances all over the world.

Nutritional strategies in plants from nutrient-impooverished landscapes

Mycorrhiza is a positively symbiotic relation between *fungi* and terrestrial plants, and is a very common strategy to improve the absorption of nutrients where their availability may be scarce. There is a variety of mycorrhiza: ectomycorrhiza (ECM), ericoid and orchid mycorrhizas and arbuscular mycorrhizal associations (AM) (Brundrett, 2004). The most cosmopolitan type of mycorrhiza is the AM, which occurs mostly as a result of *embryophyta* in association with *Glomeromycota fungi*, found in both cultivated and native species (Brundrett et al., 1996).

Particularly in severely P-impooverished soils, there is a higher proportion of non mycorrhizal (NM) species (Lambers & Teste, 2013). As pointed out by Oliveira et al (2015), the prevalence of NM strategies indicates a strong global functional convergence in plant mineral nutrition strategies among severely P-impooverished ecosystems worldwide, including the *campo rupestre*. In those environments, there are radicular specializations that provide the needed nutrients (Lambers et al., 2011).

NM species include carnivores, parasites and cluster rooted plants (Lambers et al., 2011). The latter, also called proteoid roots, which release organic anions, or carboxylates, in exudative bursts. This release of carboxylates into the rhizosphere allows the plant to access soil P that is usually inaccessible. For this reason, cluster roots are considered most effective in P-poor landscapes rather than mycorrhiza (Lambers et al., 2015).

As described by Abrahao et al (2020), there are strong relations between certain types of roots, such as vellozioid ones, and microhabitats. Vellozioid roots are a type of radicular specialization recently described and found in *campo rupestre*. In their case specifically, there is a direct relation between the rocky substrate and the release of carboxylates which interact with it. Similarly to the cluster roots, this release and interaction of carboxylates and substrate allows inaccessible P to become accessible by plants' roots (Abrahao et al., 2020). Nutrient impoverishment is one of the strongest environmental filters in *campo rupestre*. This release of carboxylates can also be found in relation to the soil 's nutritional content. The high concentration of manganese (Mn), for instance, is caused when the plants release carboxylates which allows P to be better absorbed, similarly to the effects of the carboxylates of cluster roots.

Nutrient retranslocation is another strategy that plants may display in environments that are poor in nutrients; not to better acquire them, but to more efficiently use them (Chuyong et al., 2000). This is a physiological way to ensure that the nutrients can be "recycled" within the plants' tissues. It has been indicated by literature (Santos et al., 2020) that the highest percentages of retranslocation are found in plants that have fewer *fungi* colonizations in comparison to the ones that are more colonized.

Adjustments in phosphorus fractions

Cellular P concentration is divided into two major groups: inorganic P (Pi) and organic P (Hidaka & Kitayama, 2011; Mo, et al., 2019). The organic P is subdivided into four smaller groups: metabolic (which is used in metabolic reactions), residual (which is scattered in the cytosol), lipidic (takes part in the formation of the cell) and nucleic acid (takes part in the formation of DNA and RNA). The metabolic fraction is also responsible for the fulfillment of the Calvin-Benson cycle, which means that the low concentrations of phosphorus in the leaves can directly diminish the photosynthetic process. It was often assumed that a lack of availability of P in the soil would result in relatively low photosynthetic rates (Hidaka & Kitayama, 2013), however, this has been disproved (Mo et al., 2019). Despite the low availability in the soil, the plants may display adaptations, such as changes in P allocation, that maintains their photosynthetic rates. Because of this, it is thought that these plants must have metabolic P as its highest fraction. Gao et al. (2022), indicated that the allocation patterns can vary between species and that some species, particularly endemic ones,

have higher allocation of nucleic acid P first and then metabolic P. Endorsing this, Yuan and Chen (2009) and Han et al. (2021) have disclosed that fast growing species show this behavior more than slow growing ones, since nucleic acid P is related to reproduction because of its role in the replication and formation of DNA and RNA.

Phosphorus use efficiency and nutritional traits

Not much is known about how exactly native plants from nutrient impoverished environments maintain their photosynthetic rate high enough. There has been, however, an indication that there is a relation between this and the accumulation of other nutrients, along with nutrient use efficiency (Hidaka & Kitayama, 2009).

Photosynthetic nutrient use efficiency can be defined as the rate of photosynthesis per unit leaf nutrient (Pons & Westbeek, 2004; Warren & Adams, 2004). Therefore, there is a relationship between the nutrient leaf concentration and how efficiently the photosynthetic *apparatus* uses specific nutrients in its processes. This happens specifically for P and N with PPUE (photosynthetic phosphorus use efficiency) and PNUE (photosynthetic nitrogen use efficiency) in the Calvin-Benson cycle. However, not only the foliar concentration of nutrients affects the photosynthetic nutrient use efficiency (Warren & Adams, 2004).

One particular example of the Brazilian ferruginous *campo rupestre* is that its native species, differently from cultivated ones, are tolerant to high levels of toxic nutrients, such as Mn. Some species not only tolerate Mn, but also absorb this nutrient in high concentrations. The presence of Mn in the tissues is caused by an exudation of carboxylates that, when released in the rhizosphere, make P inaccessible in the soil, accessible (Lambers et al., 2011).

Nutrients that have higher availability typically present lower use efficiency (Lambers et al., 2011; Liu et al., 2017). Because of this, the accumulation of some nutrients presents as an indirect proportion to efficient nutrient use (Lambers et al., 2022).

Transient OJIP Fluorescence: JIP-test and its response to nutritional limitations

Chlorophyll fluorescence is a useful tool in plant physiology studies and has been proven to detect early abiotic stress' effects on photosynthetic efficiency (Thach et al., 2007; Tsimilli-Michael., 2020). It allows the evaluation of plant development and

responses to environmental stresses, since we may collect the information about changes in the efficiency of photochemistry and heat dissipation by measuring the yield of chlorophyll fluorescence. This can be quantified by exposing a leaf to light of defined wavelength and measuring the intensity of light re-emitted at longer wavelengths (Maxwell & Johnson, 2000; El-Mejjaouy et al., 2022).

Chlorophyll *a* fluorescence, in general, can provide information about the state of photosystem II (PSII) by showing to what extent PSII is using the energy absorbed by chlorophyll and the extent to which it is being damaged by excess light, which is important to the physiology of the plant. Also, the electrons' flow through PSII is usually indicative of the overall rate of photosynthesis, which provides us with the possibility of estimating the photosynthetic performance under different conditions (Tsimilli-Michael, 2020; Bussotti et al., 2010).

Usually, nutrient deficiency causes a decrease in the photochemical efficiency, increases the non-photochemical dissipation and decreases the PSIIs that are active (Kalaji et al., 2014). P, specifically, has been proven to decrease the photosynthetic electron transport capacity due to the impairment of the electron transport chain. Because of this, there is an enhancement in the energy dissipation to prevent photo-oxidative damage in the leaves (Lin et al., 2009). N also takes part in similar processes. In crop plants, N deficiency diminishes the amount of electron acceptors and limits the CO₂ assimilation of plants, resulting in inactivation of PSIIs (Redillas et al., 2011).

The diversity of functional traits in Asteraceae species from ferruginous substrate

Asteraceae (Bercht. & J. Presl), part of the Asteralis order is one of the most diverse and distinct families within the angiosperms. It's composed by several *genus* and over 24,000 species. It's a family composed by an abundant number of economically relevant species, being native or cultivated ones (Roque, Magalhães & Nakajima, 2017).

There's been a lot of discussion on how to approach the taxonomy of Asteraceae with new molecular and micromorphological tools (Janačković, Susanna & Marin, 2019). However, there's been a consensus that the global characteristics of the Asteraceae family include inflorescence in capitulum, anthers, inferior ovary, bicarpelar, with a basal placentation ovum; all of which justify its taxation as a monophyletic group (Roque, Magalhães & Nakajima, 2017). The Asteraceae family is

also considered to be a generalist regarding its pollinators (Pruski & Sancho, 2004). However, there's been indications in literature that the most common relation is entomological. The insects most commonly found in these relations are butterflies and bees (Mani & Saravan, 1999).

According to Viana & Lombardi (2007), the Asteraceae family is distributed in both types of *canga*: *nodular* and *couraçada* (Dario et al., 2022). The Asteraceae species are characterized mainly by shrubs and sub-shrubs. This family's foliar traits are most diverse, having great ranges of foliar area and weight. Some species also present specific foliar characteristics, such as the presence of trichomes and thorns. The leaf's shape also displays great diversity, with some being large and long; large and short; small and short; small and long (Mucina, 2018). The species generally are of slow growth and display long leaf lifespans (Mucina, 2018).

Shields et al. (2016) have described endemic Asteraceae species in New Zealand, also characterized as slow growing plants, as having great potential as Ecosystem Service Providers (ESPs) or Service Providing Units (SPUs). Some of the listed ecosystemic services provided by them were weed suppression, conservation of beneficial invertebrates, soil moisture retention and microbial activity. Muños & Cavieres (2008), have also listed polinar services as part of the ES provided by Asteraceae species.

REFERENCES:

- Abrahamo, A., de Britto Costa, P., Teodoro, G. S., Lambers, H., Nascimento, D. L., Adrián López de Andrade, S., ... & Silva Oliveira, R. (2020). Vellozioid roots allow for habitat specialization among rock-and soil-dwelling Velloziaceae in campos rupestres. *Functional Ecology*, *34*(2), 442-457.
- Brundrett, M., Bougher, N., Dell, B., Grove, T., Malajczuk, N. (1996). Working with mycorrhizas in forestry and agriculture (Vol. 32, p. 374). Canberra: Australian Centre for International Agricultural Research.
- Brundrett, M. (2004). Diversity and classification of mycorrhizal associations. *Biological reviews*, *79*(3), 473-495.
- Chuyong, G. B., Newbery, D. M., & Songwe, N. C. (2000). Litter nutrients and retranslocation in a central African rain forest dominated by ectomycorrhizal trees. *New Phytologist*, *148*(3), 493-510.
- Dario, C. P., Jessica, C. B., JP, A. M., CP, R. D., Daniel, N., & Wilson, F. G. (2022). Floristic mosaics of the threatened brazilian campo rupestre. *Nature Conservation Research. Заповедная наука*, *7*(1), 10-18.
- El-Mejjaouy, Y., Lahrir, M., Naciri, R., Zeroual, Y., Mercatoris, B., Dumont, B., & Ouakroum, A. (2022). How far can chlorophyll a fluorescence detect phosphorus status in wheat leaves (*Triticum durum* L.). *Environmental and Experimental Botany*, *194*, 104762.
- Fernandes, G. W., Arantes-Garcia, L., Barbosa, M., Barbosa, N. P., Batista, E. K., Beiroz, W., ... & Silveira, F. A. (2020). Biodiversity and ecosystem services in the Campo Rupestre: A road map for the sustainability of the hottest Brazilian biodiversity hotspot. *Perspectives in Ecology and Conservation*, *18*(4), 213-222.
- Gao, Y., Zhang, Z., Zhang, B., Yin, H., Chai, X., Xu, M., ... & Zeng, F. (2022). Foliar P-Fractions Allocation of *Karelinia caspia* and *Tamarix ramosissima* Are Driven by Soil

and Groundwater Properties in a Hyper-Arid Desert Ecosystem. *Frontiers in plant science*, 13, 833869.

Guilherme Pereira, C., Hayes, P. E., O'Sullivan, O. S., Weerasinghe, L. K., Clode, P. L., Atkin, O. K., & Lambers, H. (2019). Trait convergence in photosynthetic nutrient-use efficiency along a 2-million year dune chronosequence in a global biodiversity hotspot. *Journal of Ecology*, 107(4), 2006-2023.

Han, Z., Shi, J., Pang, J., Yan, L., Finnegan, P. M., & Lambers, H. (2021). Foliar nutrient allocation patterns in *Banksia attenuata* and *Banksia sessilis* differing in growth rate and adaptation to low-phosphorus habitats. *Annals of Botany*, 128(4), 419-430.

Heilmeyer, H. (2019). Functional traits explaining plant responses to past and future climate changes. *Flora*, 254, 1-11.

Hidaka, A., & Kitayama, K. (2009). Divergent patterns of photosynthetic phosphorus-use efficiency versus nitrogen-use efficiency of tree leaves along nutrient-availability gradients. *Journal of Ecology*, 97(5), 984-991.

Hidaka, A., & Kitayama, K. (2013). Relationship between photosynthetic phosphorus-use efficiency and foliar phosphorus fractions in tropical tree species. *Ecology and evolution*, 3(15), 4872-4880.

Hopper, S. D., Lambers, H., Silveira, F. A., & Fiedler, P. L. (2021). OCBIL theory examined: reassessing evolution, ecology and conservation in the world's ancient, climatically buffered and infertile landscapes. *Biological Journal of the Linnean Society*, 133(2), 266-296.

Janačković, P., Susanna, A., & Marin, P. D. (2019). Micromorphology and anatomy in systematics of Asteraceae. An old-fashioned approach?. *Biologica Nyssana*, 10(2), 77-85.

Kalaji, H. M., Oukarroum, A., Alexandrov, V., Kouzmanova, M., Brestic, M., Zivcak, M., ... & Goltsev, V. (2014). Identification of nutrient deficiency in maize and tomato plants

by in vivo chlorophyll a fluorescence measurements. *Plant physiology and biochemistry*, 81, 16-25.

Pruski, J. F. & Sancho, G. (2004). Asteraceae. In: N. Smith et al. (eds.) Flowering plants systematics. Harper & Row, New York.

Redillas, M. C. F. R., Jeong, J. S., Strasser, R. J., Kim, Y. S., & Kim, J. K. (2011). JIP analysis on rice (*Oryza sativa* cv Nipponbare) grown under limited nitrogen conditions. *Journal of the Korean Society for Applied Biological Chemistry*, 54(5), 827-832.

Roque, N.; Magalhães T. A.; Nakajima, N. J. (2017). A Família Asteraceae no Brasil Clasificação e Diversidade; Editora da Universidade Federal da Bahia: Salvador, Brazil.

Lambers, H., Brundrett, M. C., Raven, J. A., & Hopper, S. D. (2011). Plant mineral nutrition in ancient landscapes: high plant species diversity on infertile soils is linked to functional diversity for nutritional strategies. *Plant and Soil*, 348(1), 7-27.

Lambers, H., & Teste, F. P. (2013). Interactions between arbuscular mycorrhizal and non-mycorrhizal plants: do non-mycorrhizal species at both extremes of nutrient availability play the same game. *Plant Cell Environ*, 36(11), 1911-1915.

Lambers, H., Martinoia, E., & Renton, M. (2015). Plant adaptations to severely phosphorus-impooverished soils. *Current opinion in plant biology*, 25, 23-31.

Lin, Z. H., Chen, L. S., Chen, R. B., Zhang, F. Z., Jiang, H. X., & Tang, N. (2009). CO₂ assimilation, ribulose-1, 5-bisphosphate carboxylase/oxygenase, carbohydrates and photosynthetic electron transport probee by the JIP-test, of tea leaves in response to phosphorus supply. *BMC Plant Biology*, 9(1), 1-12.

Liu, M. C., Kong, D. L., Lu, X. R., Huang, K., Wang, S., Wang, W. B., ... & Feng, Y. L. (2017). Higher photosynthesis, nutrient-and energy-use efficiencies contribute to invasiveness of exotic plants in a nutrient poor habitat in northeast China. *Physiologia plantarum*, 160(4), 373-382.

Mani, M. S. & Saravan, J. M. (1999). Pollination ecology and evolution in Compositae (Asteraceae). Science Publishers. New Hampshire.

Mattos, E. (2006). O estudo da fluorescência da clorofila e a detecção de estresses ambientais em plantas. PRADO, CHBA; CASALI, CA Fisiologia Vegetal: práticas em relações hídricas, fotossíntese e nutrição mineral. São Paulo: Editora Manole, 255-266.

Maxwell, K., & Johnson, G. N. (2000). Chlorophyll fluorescence—a practical guide. *Journal of experimental botany*, 51(345), 659-668.

Mo, Q., Li, Z. A., Sayer, E. J., Lambers, H., Li, Y., Zou, B. I., ... & Wang, F. (2019). Foliar phosphorus fractions reveal how tropical plants maintain photosynthetic rates despite low soil phosphorus availability. *Functional Ecology*, 33(3), 503-513.

Mucina, L. (2018). Vegetation of Brazilian campos rupestres on siliceous substrates and their global analogues. *Flora*, 238, 11-23.

Muñoz, A. A., & Cavieres, L. A. (2008). The presence of a showy invasive plant disrupts pollinator service and reproductive output in native alpine species only at high densities. *Journal of Ecology*, 96(3), 459-467.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., Da Fonseca, G. A., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature*, 403(6772), 853-858.

Oliveira, R. S., Galvão, H. C., de Campos, M. C., Eller, C. B., Pearse, S. J., & Lambers, H. (2015). Mineral nutrition of campos rupestres plant species on contrasting nutrient-impooverished soil types. *New Phytologist*, 205(3), 1183-1194.

Pons, T. L., & Westbeek, M. H. (2004). Analysis of differences in photosynthetic nitrogen-use efficiency between four contrasting species. *Physiologia Plantarum*, 122(1), 68-78.

Reich, P. B. (2014). The world-wide 'fast–slow' plant economics spectrum: a traits manifesto. *Journal of Ecology*, 102, 275-301.

Santos, L. J. S., ARANTES, A. D., Donato, S. L. R., Brito, C. F. B., LIMA, M. A. C. D., & Rodrigues Filho, V. A. (2020). Leaf contents and biochemical cycling of nutrients in accessions of umbu and umbu-caja. *Revista Caatinga*, 33, 690-701.

Shields, M. W., Tompkins, J. M., Saville, D. J., Meurk, C. D., & Wratten, S. (2016). Potential ecosystem service delivery by endemic plants in New Zealand vineyards: successes and prospects. *PeerJ*, 4, e2042.

Thach, L. B., Shapcott, A., Schmidt, S., & Critchley, C. (2007). The OJIP fast fluorescence rise characterizes Graptophyllum species and their stress responses. *Photosynthesis Research*, 94(2-3), 423–436. doi:10.1007/s11120-007-9207-8.

Tsimilli-Michael, M. (2020). Revisiting JIP-test: An educative review on concepts, assumptions, approximations, definitions and terminology. *Photosynthetica*, 58, 275-292.

Veneklaas, E. J. (2022). Phosphorus resorption and tissue longevity of roots and leaves—importance for phosphorus use efficiency and ecosystem phosphorus cycles. *Plant and Soil*, 1-11.

Viana, P. L., & Lombardi, J. A. (2007). Florística e caracterização dos campos rupestres sobre canga na Serra da Calçada, Minas Gerais, Brasil. *Rodriguésia*, 58, 157-177.

Walker, A. P., McCormack, M. L., Messier, J., Myers-Smith, I. H., Wullschleger, S. D. (2017). Trait covariance: the functional warp of plant diversity. *New Phytologist*, 216, 976-980.

Warren, C. R., & Adams, M. A. (2004). Evergreen trees do not maximize instantaneous photosynthesis. *Trends in plant science*, 9(6), 270-274.

Westoby, M., & Wright, I. J. (2006). Land-plant ecology on the basis of functional traits. *Trends in ecology & evolution*, 21(5), 261-268.

Wright, I. J., Reich, P. B., ... Villar, R. (2004). The worldwide leaf economics spectrum. *Nature*, 428, 821-827.

Yuan, Z., & Chen, H. Y. (2009). Global trends in senesced-leaf nitrogen and phosphorus. *Global Ecology and Biogeography*, 18(5), 532-542.

3. CHAPTER 2: The high functional diversity of Asteraceae species converges to similar strategies in nutrient-impooverished soils at ferruginous *campo rupestre*

Abstract

Plants in nutrient impoverished landscapes display a variety of strategies that allows better acquisition and use of phosphorus (P), such as changes in foliar P allocation, mycorrhizal associations, high P resorption rate and high photosynthetic P-use efficiency. The ferruginous *campo rupestre*, also known as *canga*, is a grassland landscape with great diversity of plant species with high functional diversity. The *canga* plant species are generally characterized as of slow growth, higher leaf mass per area (LMA) and leaf dry mass content (LDMC) when compared to other grasslands. Our main goal was to understand the diversity of strategies to acquire and use P in Asteraceae species from ferruginous *campo rupestre*, regarding their functional traits and photosynthetic performance. Field surveys were conducted at *Serra da Calçada* (MG, Brazil) during the rainy season of 2021 and 2022. We selected five Asteraceae species: *Dasyphyllum velutinum*, *Baccharis reticularia*, *Lychnophora pinaster*, *Trixis vauthieri* and *Hololepis pedunculata*. We found a high diversity of functional traits, such as LMA, LDMC, foliar area and nutrient concentration. Some of which can be directly and indirectly related to the diversity of strategies to better absorb and use P. The species showed, in general, 70% mycorrhizal colonization and retranslocation rates of 77% and 65% for P and N, respectively. Even though these species have a diversity of functional traits, their photosynthetic P-use efficiency and P fractions (metabolic, nucleic acid, lipidic and residual) showed no significant difference. We have concluded that the diversity of functional traits found in Asteraceae species is most likely related by its phylogeny and the history of evolution of Asteraceae in the ferruginous *campo rupestre*. The ecological filters imposed by nutrient availability pressure during their evolutionary path have converged into a similar physiological performance.

Keywords: P-fractions; *canga*; nutritional strategies; photosynthetic P-use efficiency; mycorrhiza.

3.1 INTRODUCTION

Nutrient-impoverished landscapes are usually global hotspots of biodiversity (Oliveira et al, 2015) and, therefore, are the ones that are most likely to suffer from climatic changes and anthropic pressures, such as urban expansion and industrial activities (Silveira et al., 2016). Phosphorus-impoverished soils may limit the growth of plants in different ecosystems all over the globe (Hernández & Munné-Bosch, 2015; Zhongming et al, 2021).

The low availability of phosphorus in the soil influences the foliar P concentration and its fractions, which contains both organic and inorganic compounds (Zhongming et al, 2021). Often, plant species adapted to P-impoverished soils, present adjustments in the foliar allocation of P and its P-fractions (Zhongming et al. 2021). Cellular organic P is subdivided into four smaller groups: metabolic (which is used in metabolic reactions), residual (which is scattered in the cytosol), lipidic (takes part in the formation of the cell) and nucleic acid (takes part in the formation of DNA and RNA) (Hidaka & Kitayama, 2011 and Mo, et al., 2019). The changes in foliar P-fractions are functionally related to the stress-tolerance capacity in plants from severely impoverished soils, as well as influencing their growth and reproduction (Zhongming et al. 2021).

The P limitation in the soil would also affect the functional traits of plants. It is expected that in P-impoverished environments the plant species possess leaves with longer life spans (LLS), higher leaf dry mass contents (LDMC) and leaf mass per area (LMA). Also would be present some morphological leaf adaptations such as thinner shaped and thicker leaves and even presence of trichomes (Medeiros et al., 2008; Negreiros et al., 2014; Rios et al., 2022). These adaptations directly and indirectly affect how the species respond to stressful conditions (Wright et al., 2004). Even so, there is still a gap as to how these functional traits relate to strategies to better use and acquire nutrients such as P and N in nutrient impoverished landscapes (Westoby & Wright, 2006; Reich, 2014; Heilmeyer, 2019).

The *campo rupestre* is an old and nutrient-impoverished grassland ecosystem (Abrahão et al, 2019) in Brazil, which has been appointed as a conservation priority (Oliveira et al, 2015). The *campo rupestre* has been threatened by mining (Silveira et al, 2016), particularly in the iron quadrangle in *Minas Gerais*, which harbors the ferruginous *campo rupestre*, known as *canga*, rich in iron ore and other minerals of

high economic interest (Carmo & Jacobi, 2013). The ferruginous *campo rupestre*, is a place with high botanical and functional diversity, and high rates of endemism, which provides us with an opportunity to further study and understand the relation of functional traits and the photosynthetic and nutrient-acquisition strategies that native plants display in this impoverished reference ecosystem (Abrahão et al, 2019).

The Asteraceae family is one of the most representative in *campo rupestre* landscapes, and hosts a high diversity of species with functional traits. In the ferruginous *campo rupestre*, its species vary greatly in niches occupied. This is especially true regarding the leaf traits such as shape, mass, area, thickness and foliar specializations (Caminha-Paiva, et al., 2021; De Oliveira et al., 2018). This could give ground to new restoration techniques and strategies for the rehabilitation of mining-impacted landscapes.

Given that studies such as Mo et al (2019) show that species from nutrient impoverished landscapes can display allocation and retranslocation of nutrients in order to maintain their photosynthetic rates, specially when it comes to their metabolic P fraction, our hypothesis is that the diversity of functional traits in Asteraceae species from *canga*, will reflect in a diverse range of strategies to better acquire, absorb, allocate and efficiently use P in its photosynthetic processes. Those adjustments would include higher foliar P allocation, higher mycorrhizal associations, and high P resorption rate that would sustain high efficiency in the use of P and photosynthetic metabolism despite the low nutrient availability in this ecosystem. Our main goal was to understand the diversity of strategies to acquire and use P in Asteraceae species from ferruginous *campo rupestre*, regarding their functional traits and photosynthetic performance.

3.2 MATERIAL AND METHODS

Study area and sampled species

The study was conducted in *Serra da Calçada*, which is located in the Brazilian's iron quadrangle, located in the *Serra do Espinhaço* chain (20°05'35"S, 43°59'01"W), at 1,5 m height (Viana & Lombardi, 2007). The local climate is classified as tropical with a dry winter (Kottek et al., 2006), according to the classification of Köppen and Geige (2006). It is characterized by an average temperature of 20,7 °C, low annual thermal variation, dry winters from May to October, rainy summers from November to

April, and an average annual rainfall of 2041.6 mm. The soil in the area was characterized with low availability of nutrients, with a pH of 4.56 ± 0.13 ; P of 0.92 ± 0.08 ; potassium (K) of 30.9 ± 2.18 ; iron (Fe) of 172.48 ± 8.18 ; manganese (Mn) of 27.24 ± 3.41 ; zinc (Zn) of 3.15 ± 0.29 (Rios et al., 2022).

The Asteraceae family was pointed as the one of the most rich families in the *Serra da Calçada* area, with 42 species (Viana & Lombardi, 2007). We have chosen five species from the Asteraceae family representative from the *canga* phytophysionomy for analysis: *Hololepis pedunculata* (DC. ex Pers.) DC.; *Trixis vauthieri* (DC); *Dasyphyllum velutinum* (Baker) Cabrera; *Baccharis reticularia* (DC); *Lychnophora pinaster* Mart. (Fig. 1). Individuals from the same species were at least two meters apart so we could make sure they were different individuals instead of ramifications.

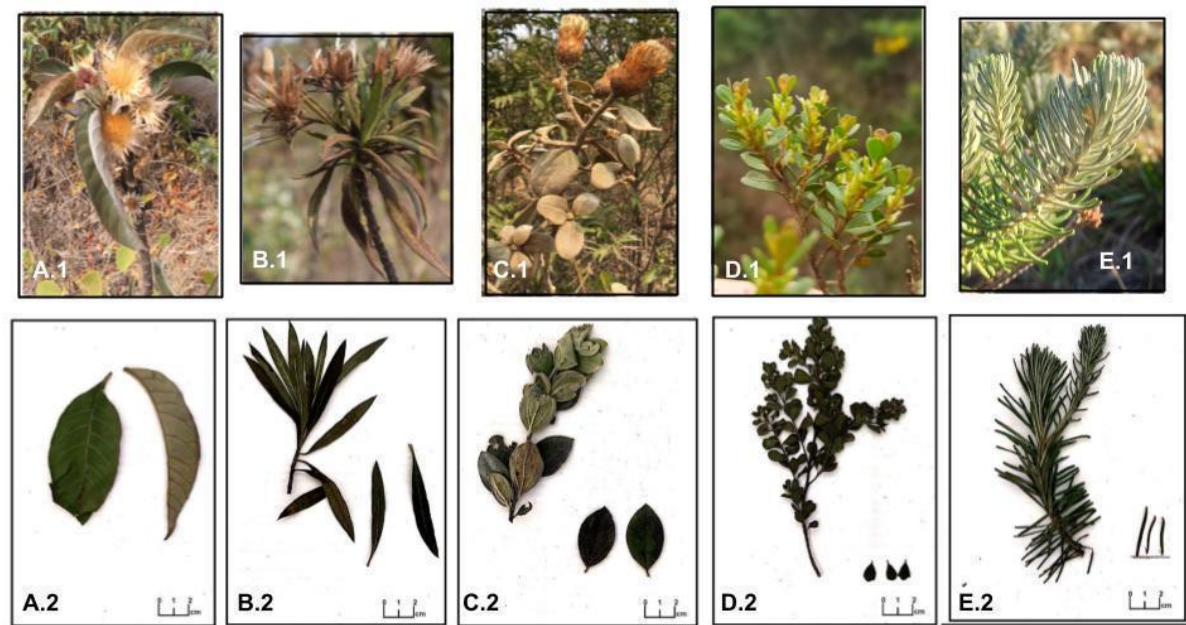


Figure 1: The five Asteraceae species selected for the study - A: *Hololepis pedunculata*; B: *Trixis vauthieri*; C: *Dasyphyllum velutinum*; D: *Baccharis reticularia*; E: *Lychnophora pinaster*. Image source: A.1, A.2, B.1, B.2, C.1, C.2, D.2 and E.2 (personal archive). D.1 and E.1 (gently provided by Paulo Antônio Pimentel). (Scale = 2cm)

Soil and mycorrhizal colonization analysis

Soil samples in contact with the roots of each individual (rhizosphere soil at 5 centimeters of depth), have been collected to perform a more detailed analysis of the

plants' microhabitats. We measured the P, Mn and Fe concentration in the soil according to EMBRAPA (Silva et al., 1998).

After collecting the soil, we collected root samples for both P concentration (using dry material) analysis and a mycorrhizal-association inspection (using preserved material). During the inspection, we checked whether there was mycorrhizal presence and calculated the percentage of the colonizations for each individual, using the coverslip method (Brundrett et al., 1996) and a microscope, for the samples were too small for petri dishes.

Leaf Mass per Area, Leaf Dry Mass Content and foliar area

In March, 2021, we collected branches with leaves (enough to provide us with 1g of foliar material) and stored them in moist paper bags for further analysis, which was conducted in the lab with the last pair of fully and healthy expanded leaves. This collection was performed during the morning. While the leaves were still fresh, they were weighted and photographed, with a scale and the leaf area was calculated using the program ImageJ. The leaves were then put in a drying oven, where they remained at 65°C until the mass was stabilized. The leaf dry mass content (LDMC) was calculated using the values of the fresh mass and the dry mass. Using the dry mass weight and the foliar area, we calculated the leaf mass per area (LMA).

Nutrient concentration and foliar P fractionation

In March, 2021, the concentration of Ca, Mn, Fe and N in the leaves and P in the leaves and roots was measured. We weighted 0,5 g material of both tissues after it was dried and grinded. N was sulfuric acid-biodigested. P, Fe and Mn were nitric-perchloric acid-biodigested. The absorbance of Ca, Mn, Fe and P was measured using the spectrophotometer (a Shimadzu UV-1800 model) and N using an elemental analyser (Perkin-Elmer CHNS/OPEZ 400 series) (Carmo et al., 2000).

The P-fractions (lipidic, metabolic, nucleic acid and residual) were measured using fresh leaf material (50 mg), which was collected and stored in liquid nitrogen in the field, being kept in it until the fractionation started, remaining cool until the end. The extraction and fractionation was performed using CMF (chloromethylfurfural), CMW (3-[[4-methylphenyl)sulfonyl]amino}propyl pyridine-4-yl carbamate), CWW (chloroform water washed), TCA (trichloroacetic acid) and methanol. The samples were centrifuged, evaporated and sulfuric acid-biodigested. Since the fractions contain small

concentrations of P, we used malachite green dye (De Lima Campos, personal communication) and measured the absorbance of 650 nm using the spectrophotometer (UV-1800, Shimadzu, Japan).

Evaluation of nutrient cycling

In March, 2022, we collected foliar material once more in order to perform a relative magnitude of biogeochemical cycling (RMBC) analysis. Leaves from each individual plant were collected in two different developmental stages (young and old). The young leaves were chosen as the last pair of fully expanded leaves of each branch, and the first, mostly dried senescent pair of leaves of each branch for the older ones.

We measured the concentration of P and N, which are mobile in plants, as well as Ca, which is immobile (Carmo et al., 2000). The latter is used as a marker to determine the cycling of mobile nutrients in each species based upon the following equation:

$$\text{Re Nut} = 1 - \left[\frac{\text{Mobile nut. (senescent leaves)/Ca (senescent leaves)}}{\text{Mobile nut. (young leaves)/Ca (young leaves)}} \right] \times 100$$

in which:

%Re Nut = percentage of the translocated nutrient

Mobile Nut (senescent leaves)/Ca (senescent leaves) = relationship between the concentrations of mobile nutrients and Ca concentration in senescent leaves

Mobile Nut (young leaves)/Ca (young leaves) = relationship between the concentration of mobile nutrients and Ca concentration in young leaves (Chuyong et al., 2000, Santos et al., 2020).

Photosynthetic parameters and PPUE

In March, 2021, we measured, in the field, the gas exchange (net photosynthetic rate, stomatal conductance, transpiration and internal CO₂ concentration) in 10 healthy adult individuals of each species (a total of 50 individuals) using an infrared gas analyser (IRGA, LI-6400XT, LICOR, NE, USA), using a 2x3 cm chamber. The measurements were made in the morning, from 7:00 to 11:00 am at an irradiance of 1500 μmol m⁻² s⁻¹, at 28°C, with a CO₂ concentration of 400 ppm.

The adjustment of the foliar area, for the species that had leaves smaller than the chamber area (*B. reticularia* and *L. pinaster*), was made for correct determination of gas exchange variables, using the program ImageJ. In their case, the leaves had to

be detached from the branch; this was possible because the leaves kept their photosynthetic rate for approximately 14 minutes before it decayed from the detachment, which was enough time to conduct the analysis.

Chlorophyll fluorescence transient was measured using a chlorophyll fluorometer (FluorPen FP 110 model) after an hour acclimation to the dark. We performed the JIP test (Bussotti et al., 2010), which provided us with data such as the specific energy fluxes (per reaction center) for absorption (ABS/RC), trapping (TR_0/RC), electron transport (ET_0/RC) and energy dissipation flow (DI_0/RC) (Bussotti et al., 2010). We also evaluated the maximum quantum efficiency of PSII (F_v/F_m).

We calculated the Photosynthetic Phosphorus Use Efficiency (PPUE) values based upon the first nutritional concentration, which was performed in the same leaves we conducted the physiological analysis.

Experimental design and statistical analysis

In the field, we chose 10 healthy and adult individuals of each species (replicates) to conduct the analysis, in a total of 50 individuals. First, we performed the Shapiro-Wilk test, followed by the Bartlett test and the Levene test in order to assess the data's normal distribution and its variance and independence. The statistical analysis compared species conducted by the multivariate statistics test ANOVA (Analysis of Variance) using $\alpha = 0,05$ for the normally distributed data (C_i , F_v/F_m , mycorrhizal colonization, ET_0/RC , ABS/RC , TR_0/RC and DI_0/RC); we used Duncan as its post-hoc test. The non-parametric equivalent Kruskal-wallis test for the data which didn't fit the ANOVA assumptions (leaf area, LDMC, LMA, net photosynthesis, stomatal conductance, transpiration, P concentrations (total and fractions), Mn and Fe in both leaves and roots, PPUE and chlorophyll index); we used Dunnett as its post-hoc test. We also performed a Principal Component Analysis (PCA) using all of the measured parameters. All tests were performed using the software R 4.2.1 version.

3.3 RESULTS

High concentrations of Fe and Mn differ in the rhizospheric soil of Asteraceae species

For both Mn and P concentration in rhizospheric soil, *B. reticularia* had the highest means. For Fe, it was *H. pedunculata*. The highest Fe leaf concentration was found in *T. vauthieri*; for Mn, it was *L. pinaster* (Table S1).

There's a high diversity of functional traits in Asteraceae species from *canga*, with higher mycorrhizal colonization and nutrient concentration in leaves and roots

H. pedunculata had the highest leaf area and leaf dry matter content (LDMC) among the Asteraceae species evaluated, while *L. pinaster* had the highest leaf mass per area (LMA) (Table 1). *T. vauthieri* had the highest leaf concentrations of both P and N. *D. velutinum* had the highest N:P ratios and P concentration on roots. *L. pinaster* had the highest mycorrhizal colonization percentage (Table 1).

We also calculated the concentration of P, N and the N:P ratios in senescent leaves. The highest concentration of N was found in senescent leaves of *T. vauthieri*; for P, it was *B. reticularia* and for the N:P ratios, it was *L. pinaster* (Table S2).

Table 1: Functional traits (leaf area, LMA, LDMC, N and P concentration on leaves, N:P ratios and P concentration on roots) and mycorrhizal colonization of *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. P, phosphorus; N, nitrogen; LMA, leaf mass per area; LDMC, Leaf Dry Mass Content. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$). These values are the means of the ten repetitions per species. Mean \pm standard error.

Species	<i>H. pedunculata</i>	<i>T. vauthieri</i>	<i>D. velutinum</i>	<i>B. reticularia</i>	<i>L. pinaster</i>
Leaf area (cm ²)	40.78 \pm 11.01a	16.44 \pm 5.27 b	9.62 \pm 2.21 b	0.61 \pm 0.21 b	0.44 \pm 0.16 b
LDMC (g g ⁻¹)	0.829 \pm 0.16 a	0.654 \pm 0.17 b	0.677 \pm 0.30 b	0.433 \pm 0.07 b	0.416 \pm 0.11 b
LMA (g m ⁻²)	380.03 \pm 79.95 b	293.15 \pm 77.90 b	416.28 \pm 123.67 b	389.47 \pm 53.97 b	830.42 \pm 240.29 a
[N] on leaves (g Kg ⁻¹)	18.50 \pm 3.32 b	32.78 \pm 4.85 a	21.98 \pm 2.53 b	24.38 \pm 3.58 b	16.59 \pm 2.17 b
[P] on leaves (g Kg ⁻¹)	1.23 \pm 0.41 b	1.98 \pm 0.52 a	1.07 \pm 0.05 b	1.53 \pm 0.25 b	1.04 \pm 0.32 b
N:P ratios	15.09 \pm 2.18 b	17.15 \pm 3.02 b	20.57 \pm 2.01 a	15.73 \pm 1.60 b	16.85 \pm 3.66 b
[P] on roots (mg Kg ⁻¹)	18.19 \pm 0.56 b	19.03 \pm 0.51 b	23.47 \pm 1.67 a	16.43 \pm 0.65 b	19.25 \pm 1.02 b
Mycorrhizal colonization (%)	58.98 \pm 6.10 b	58.65 \pm 5.88 b	61.29 \pm 5.47 b	51.69 \pm 4.77 b	70.98 \pm 12.97 a

Gas exchange, chlorophyll a fluorescence: different traits in *canga* environment, yet same net photosynthesis in Asteraceae species

There was no statistical difference detected for the net photosynthesis (A_n), transpiration rate (E) and stomatal conductance (g_s) in the evaluated Asteraceae species. The internal concentration of CO_2 (C_i) was the only data which was significant and *B. reticularia* had the highest mean (Fig. 2).

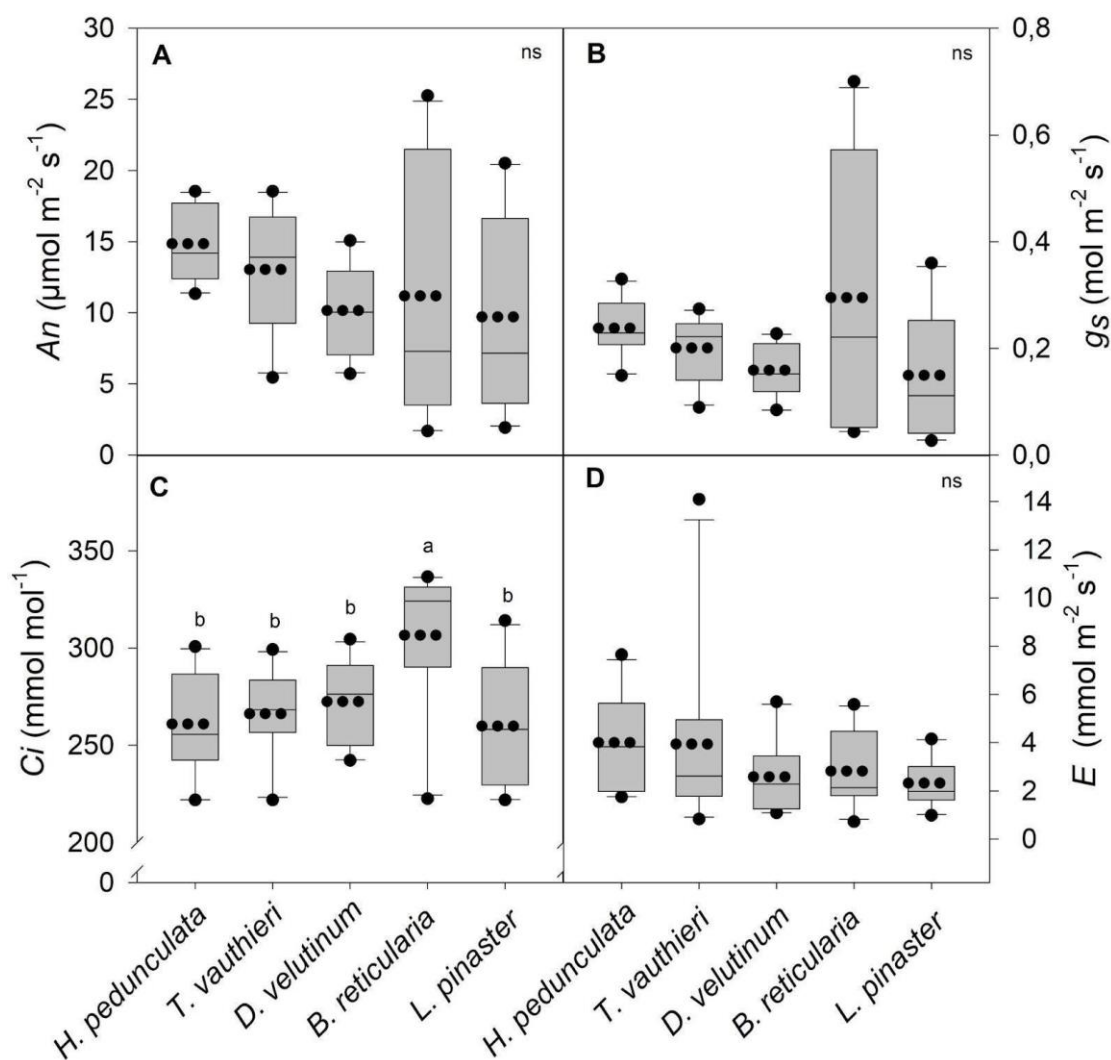


Figure 2: Boxplot of the net photosynthesis (A_n ; A), stomatal conductance (g_s ; B), internal CO_2 concentration (C_i ; C) and transpiration rate (E ; D) in ten individuals of *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. The median is shown by a straight line, the mean by the dotted line and the outliers are shown as black dots. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$).

D. velutinum had the highest mean for chlorophyll *a*, *b* and total (Fig. 3 and Fig. S1). On the other hand, *B. reticularia* had the highest values for the ratio chlorophyll *a/b* (Fig. S1). No difference was found between the species for Fv/Fm (Fig. 3)

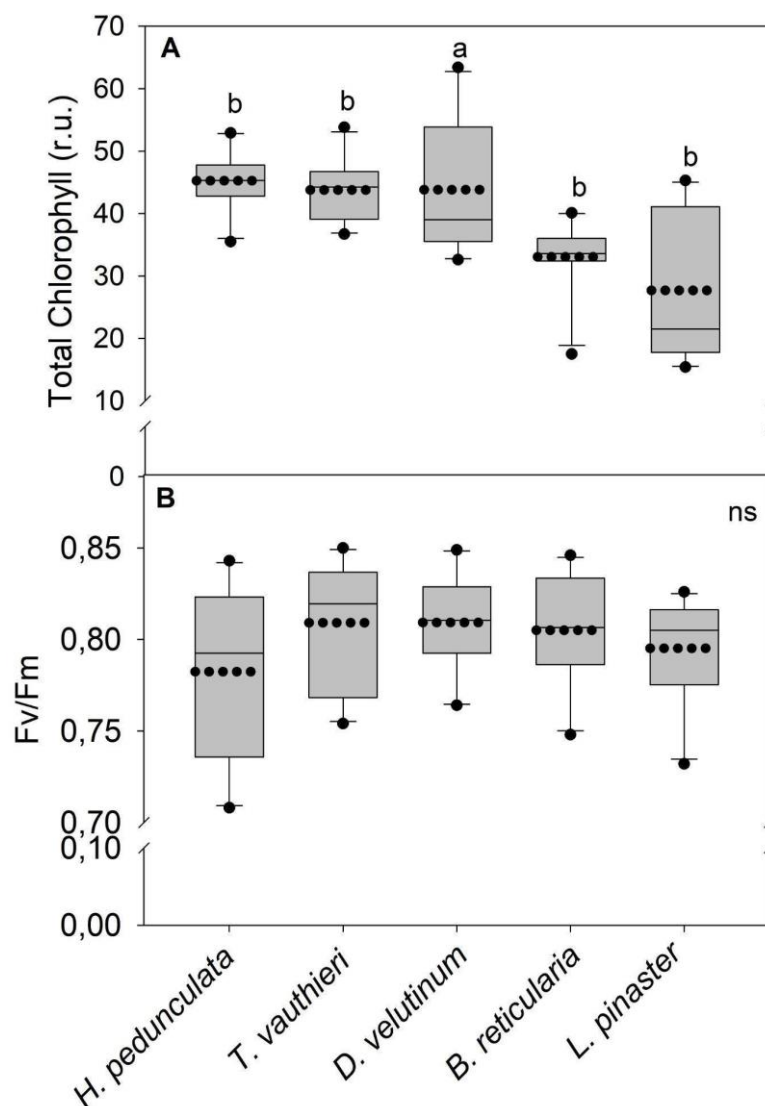


Figure 3: BoxPlot of total chlorophyll (A) and maximum quantum efficiency of Photosystem II (Fv/Fm; B) in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster* using ten individuals. The median is shown by a straight line, the mean by the dotted line and the outliers are shown as black dots. Different letters indicate different means according to statistical test (Duncan or Dunent) at the 5% confidence level ($p \leq 0,05$).

The different stages of the fluorescence transient - O, J, I, P - showed similar responses for all species in the induction curve (Figure S2). There was a significant difference in the ET₀/RC, ABS/RC and TR₀/RC among the species, with *B. reticularia*

the highest for all three variables (Fig. 4). However, there was no significant statistical difference for DI_o/RC .

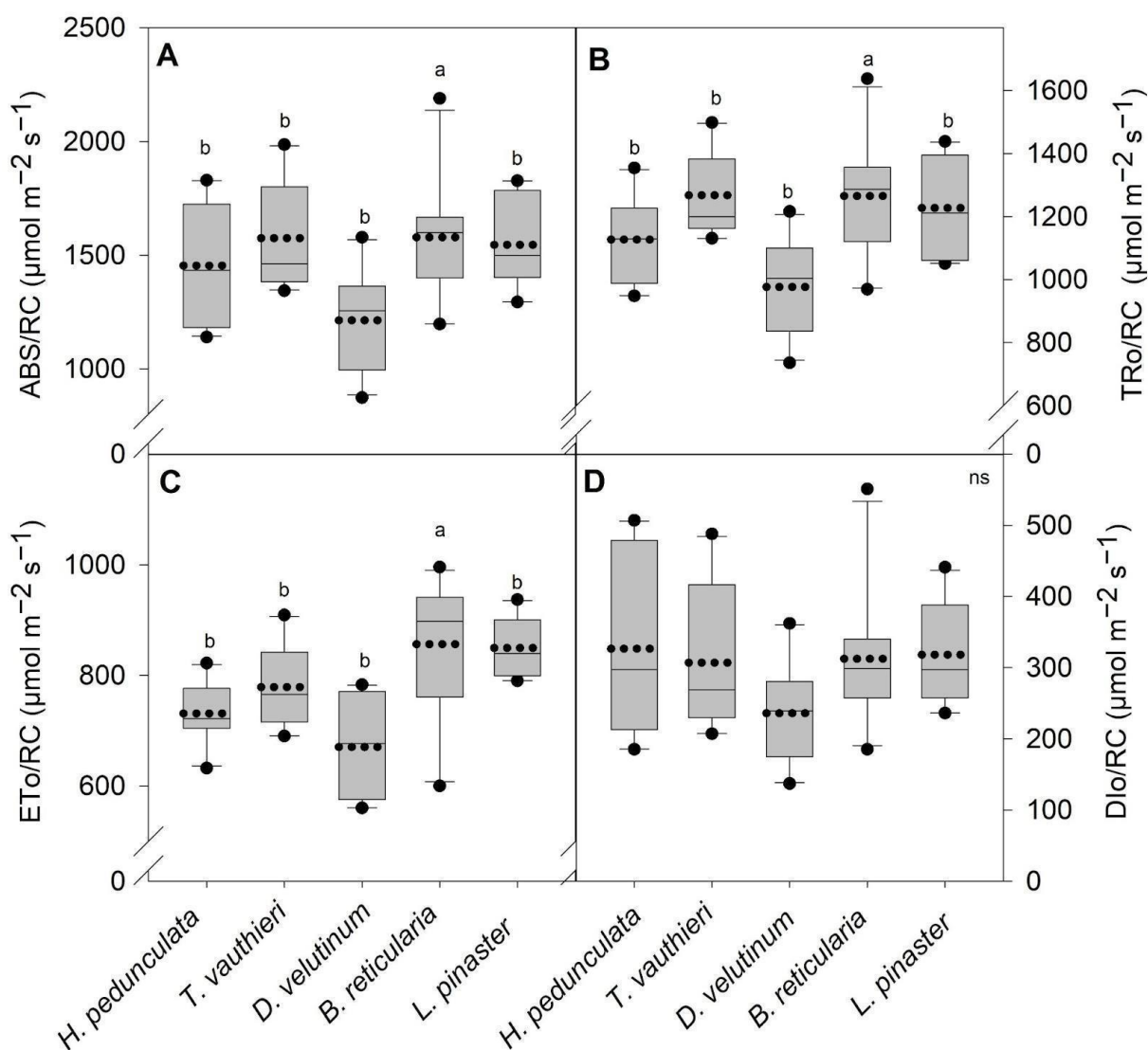


Figure 4: Boxplot of JIP's test flux variables: energy absorption by active reactive center (ABS/RC; A); energy trapping by active reactive center (TR_o/RC; B); electron transport by active reactive center (ET_o/RC; C) and energy dissipation by active reactive center (DI_o/RC) in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. The median is shown by a straight line, the mean by the dotted line and the outliers are shown as black dots. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$).

For photosynthetic P-use efficiency (PPUE) and retranslocation of P and N, *L. pinaster* had the highest mean compared to the other species (Fig. 5).

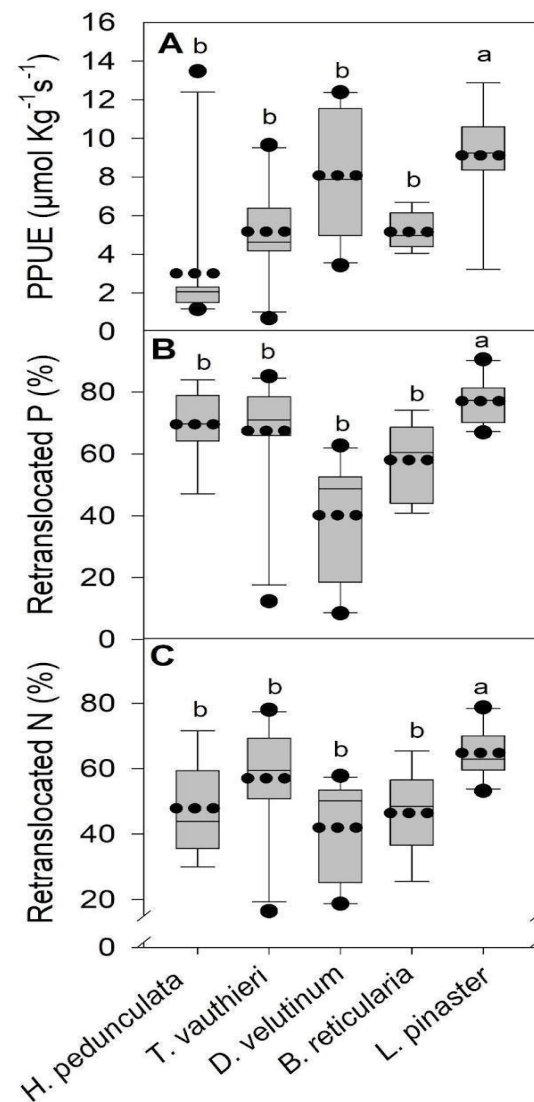


Figure 5: Boxplot of photosynthetic phosphorus-use efficiency (PPUE; A); retranslocation percentage of phosphorus (B); and nitrogen (C) in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. The median is shown by a straight line, the mean by the dotted line and the outliers are shown as black dots. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$).

Regarding the P fractions (metabolic, nucleic acid, residual and lipidic), no statistical differences were found between the five species. Among the four fractions' absolute values' averages, the highest and second highest ones were, respectively, Nucleic Acid and Metabolic P (Fig. 6).

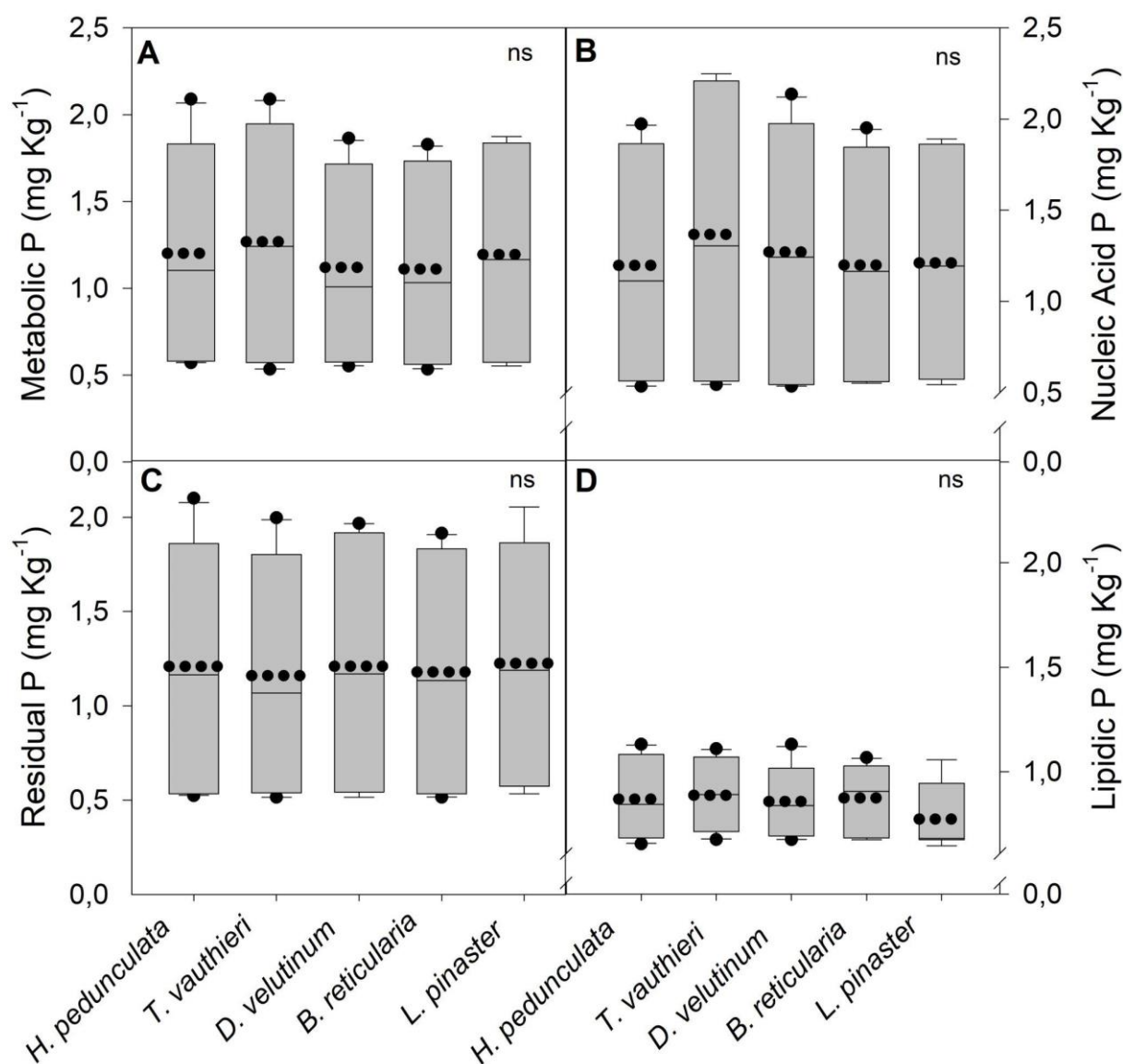


Figure 6: Boxplot of phosphorus fractions: metabolic P (A), nucleic acid P (B), residual P (C), lipidic P (D) in leaves of *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. The median is shown by a straight line, the mean by the dotted line and the outliers are shown as black dots. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$).

Principal component analysis (PCA) of functional traits, photosynthesis, JIP test, chlorophyll of Asteraceae: 4 main groups out of five species

We performed a PCA using all of the variables previously described and this analysis divided the five Asteraceae species into four groups: the first is composed by *D. velutinum*, the second by *B. reticularia*, the third by *L. pinaster* and the fourth by both *H. pedunculata* and *T. vauthieri* (Figure S3).

The direct relations between the species and variables are, per group: first, LMA, N:P ratios and P concentration in the roots. Second, PPUE and P concentration in the soil. Third, Mycorrhizal colonization, concentration of Mn in the leaves and ET_o/RC . And fourth, Fe concentration in the soil and leaves, A , E , LDMC, leaf area, Metabolic P and P concentration in the leaves (Figure S3).

The most interesting indirect relations between species and variables are in the first group: *D. velutinum* had the lowest relation with N and P retranslocation, ABS/RC and TR_o/RC (Figure S3).

3.4 DISCUSSION

The five Asteraceae species, despite seemingly presenting different functional traits, presented a convergence of these traits, including the photosynthetic and nutritional strategies into similar performance. Studies have indicated that species from nutrient impoverished environments, throughout the generations, display different roots (Abrahamo et al., 2014) and leaves (Guilherme Pereira et al., 2019) traits that culminate into a functional convergence of acquisition and efficient use of P in the photosynthetic process as observed in our work. The literature appoints this performance as an ecosystem-based common convergence (Veneklaas, 2022; Lambers et al., 2022). This is particularly relevant in the ecosystem's balance, since this physiological functional convergence consequently enhances trade off relations between the various botanic and animal species, with relations such as herbivory, inhabiting that environment and maintaining their ecosystemic services (Guilherme Pereira et al., 2019).

In the face of the climatic and biodiversity crisis, this balance is crucial for the maintenance of these services; not just for animals and plants to thrive, but also for humans. The great representativeness of the Asteraceae family in the *campo rupestre* allows us to comprehend the relations between diversity of functional traits and the nutrient acquisition strategies. For instance, the P concentration in the roots may suggest that *D. velutinum* stores P in its root tissues, since it was not in the P richest microhabitat (Nkrumah et al., 2021). The N and P concentrations in the leaves (Table 1) showed an interesting result: *T. vauthieri* was the species that had the highest leaf concentration of both nutrients, despite not being the richest P-soil species. This suggests that *T. vauthieri* has a better N and P acquisition NM strategy than, for

instance, *B. reticularia*, which had the highest soil concentration of P (Hayes et al., 2014). These values gave ground to the N:P ratios. Its average value was of 17,07, which shows that the analyzed species, as a whole, are nutrient limited by both Nitrogen and Phosphorus (Güsewell, 2004).

Campo rupestre plant species are more limited by P than N (Abrahamo et al, 2019; Oliveira et al, 2015). Studies performed with some Asteraceae species in the quartzite *campo rupestre* have presented N:P values of 24 (Oliveira et al., 2015). With other families, higher than 25 and up to 65 for species from amazonian *canga* (Mitre et al., 2018). This difference from literature can be explained, however, by the fact that most studies were conducted in different locations of a different subtype, the quartzite *campo rupestre* (Oliveira et al, 2014).

The results show a high diversity of functional, morphological and physiological traits in the five species that, in the grand picture, converge. The difference in chlorophyll concentration and PSII efficiency is most likely due to the fact that even though *B. reticularia* doesn't have as much chlorophyll as *D. velutinum*, its use of chlorophyll in photosynthetic process must be more efficient, given that both species share an equal photosynthetic performance and similar LMA. Despite the high leaf functional diversity the five species have varied seemingly shared adaptations that increase their photosynthetic performance since they share the same habitat and have shared it for many generations (Godoy et al., 2020). Also, the nutritional limitations imposed by the *canga* such as low N and P availability influence the physiological responses of the species (slow growth, longer leaf life spans, higher mycorrhizal colonizations, higher retranslocation of nutrients), despite the difference in the functional traits (Morellato & Silveira, 2018; Caminha-Paiva et al., 2021).

The results on photosynthetic P-use efficiency (PPUE) and nutrient retranslocation (Fig. 5) highlighted *L. pinaster*, as the highest means among all of the species. This may be explained by the fact that this species also presented higher mycorrhizal colonization (Table 1). Mycorrhiza are expected to be effective at enhancing P acquisition in environments where the only pathways to absorb P are through the roots and mycorrhizal associations (Lambers et al., 2022) but are supposed to be ineffective in *campo rupestre*, for the costs of maintaining this colonization are too high in comparison to its benefits. However, in the studied *canga*, all species had colonization means higher than 50% (51 to 70%), in contrast with the quartzite *campo rupestre*, where the colonization is lower than 50% (Oliveira et al., 2015).

Another study, conducted in a different portion of *canga* environment (*Serra do Rola Moça*) in the *Espinhaço* chain found, in 3 Asteraceae species, a 40% colonization (Baião et al., 2021). One possibility to explain the differences found in the quartzitic and *canga* environments is the P availability in the soil, which could be a factor that makes the mycorrhizal association advantageous. However, the P values in the soil presented in the quartzite are not superior to justify this contrast (Oliveira et al., 2015); although the P values in *Serra do Rola Moça's canga* are higher enough to explain the differences found in the species of the same family (Baião et al., 2021). Another, and more plausible, possibility is that the high concentration of metals in the soil contributes to the association of plants and *fungi* and consequently confers more tolerance to the this particular *canga* species (Oliveira et al., 2016; Folli-Pereira et al., 2012; Canton et al., 2016; Ramos et al., 2016).

L. pinaster is one of the main species of the *Lychnophora* genus, endemic to the *Minas Gerais* state, mainly distributed along the *Espinhaço* chain (Silva, Vieira & Marques, 2018). It is found in different types of *campo rupestre* (Mourão & Stehmann, 2007), which may explain why it differs from the other four species in mycorrhizal colonization, PPUE and nutrient retranslocation; all three contributing to the plant's successful establishment in the *canga*.

In order to ensure the high PPUE in Asteraceae species from *canga*, some mechanisms are involved, such as enhancing the acquisition of P through the mycorrhizal association and its more efficient use through its higher retranslocation of P. It can also be amended by long leaf longevity, which has been presented by literature as a characteristic of *canga* species (Lambers et al., 2022). Although the concentration of Mn in the soil could be considered high, or toxic even, for a great number of cultivated species, it is not for native ones (Lambers et al., 2015). The Mn concentrations were also too low to hypothesize the exudation of carboxylates and accessiblize P in the soil and, therefore, were considered unlikely as a strategy (Lambers et al., 2015). This is especially true since *L. pinaster* had the highest concentration of Mn in its leaves and highest mycorrhizal colonization, opposed to what the literature appointed for species from other plant families, such as Proteaceae, Cyperaceae, Anarthriaceae, Xanthorrhoeaceae, Dasypogonoceae, Malpighiaceae, Cactaceae and Poaceae (Lambers et al., 2015; Lambers et al., 2022).

Another endorsement of this is the relation between the translocation of N and P and LMA. The higher the nutrient retranslocation and LMA, the lower the P concentration (Veneklaas, 2022; Lambers 2022). Interestingly, the difference found in the total concentration of P among the species did not result in differences after P fractionation. There was no significant difference between the five species for both the fraction related to the photosynthetic process (metabolic P) (Fig. 6) and the net photosynthesis values (Fig. 2). Therefore, these results endorse previous research (Mo et al., 2019) that indicated that the plants like these studied Asteraceae species that inhabit P-poor soils have adaptations to keep the metabolic P absolute values higher than other fractions in order to maintain an optimum photosynthetic performance.

Many authors (Epstein and Bloom, 2005; Oliveira et al., 2015; Lambers et al., 2011) points out that the mechanisms that keep the metabolic P higher than other fractions are most likely strategies to increase the amount of absorbed P. Despite this, other studies with P fractions-related evaluations (Mo et al., 2019, Gao et al., 2022) have shown that there are also strategies that increase nutrient use efficiency rather than to just increase the absorption.

Tropical tree species react to low P availability by diminishing the concentration of foliar P (Hidaka & Kitayama, 2011), which are also a common response in plants from arid ecosystems (Gao et al., 2022). Indeed, some studies (Hidaka & Kitayama, 2011; Gao et al., 2022) showed lower foliar P values, coupled with lower concentration of total foliar P, allocation in metabolic P fraction higher than the others, as the strategy to thrive in nutrient impoverished environments. The allocation patterns can vary between species. Most species that allocate P are coupled with higher absolute values of nucleic acid P first and then metabolic P (Gao et al., 2022). This relation endorses that our results may also indicate P allocation as a strategy to increase the use efficiency of this nutrient in all five Asteraceae native species.

Fast growing species commonly show higher nucleic acid P more than slow growing ones (Chen, 2009; Han et al., 2021). Nucleic acid P is also related to the reproductive process in plants. However, it is known that *canga*'s native species are not in the fast growing spectra (Wright et al., 2004), but the Asteraceae family hosts many fast growing species (Smith et al. 1993, Mateo et al., 2001, Bartolome et al., 2013), which could also endorse our results. Complementary to that, other works (Zhang et al., 2021; Sun et al., 2022) have also shown interesting results regarding P

fractions in slow- and fast-grown Asteraceae, native and invasive species, respectively. Despite the absolute values in all the four P fractions were higher than ours, their results indicate that invasive species enhances *fungi* root colonization as well as the P availability in the soil (Zhang et al., 2021; Sun et al., 2022). Native and invasive Asteraceae species also had the same net photosynthetic values and Metabolic P fraction (Sun et al., 2022). The nucleic acid P-fractions were higher than the metabolic fractions when there was no addition of nutrients (Zhang et al., 2021; Sun et al., 2022) in both invasive and native species. Therefore, it is safe to conclude that not only the speed growth directly relates to the higher values of nucleic acid than metabolic P fractions. It is also known that fast-grown species commonly also reduce P retranslocation effectiveness (Veneklaas, 2022). Our results are complementary to that, since all five species are effective in the retranslocation of both N and P.

The percentage of P and N retranslocation found in the Asteraceae species is, averagely, 75%, superior to the 65% average of most species (Veneklaas, 2022; Vergutz et al., 2012), being able to reach levels as high as 80%, especially in *L. pinaster*. Recycling P from senescencing tissues in P-poor environments is a process that is of higher complexity in roots than in leaves (for these two tissues are both considered to have high metabolic activities). Similar net photosynthesis and metabolic P fraction also indicate that these strategies make all five Asteraceae species thrive in the same environment, maintaining their high performance, reducing competition or impacting their fitness (Godoy et al., 2020).

The strategies to acquire and use P are crucial factors to understand promotion of the species co-existence in hyperdiverse in P-impoverished and fire-prone landscapes (Lambers et al., 2022), including the ferruginous *campo rupestre*. Another question we must ask is why is Asteraceae so representative in *canga*. Low P is related to landscapes with high diversity (Veneklaas, 2022; Li et al., 2007; Lambers et al., 2011) and was confirmed by Rios et al (2022) for 22 plant species in the same sampled *canga* area. Though it is not what we expected to find according to our hypothesis, the statistically equal photosynthetic rates as well as metabolic P fraction in all five species are a confirmation that despite the differences in morphology, the plants seem to share one main physiological mechanism through different adaptations that guarantee it. Their various adaptations have converged into an efficient physiological performance (Fig. 7).

D. velutinum was the species with higher chlorophyll index and the presence of whitish trichomes affect the photosynthesis greatly by enhancing the reflectance and, therefore, playing a morphological contribution in excess light dissipation. *B. reticularia* with higher chlorophyll *a/b* ratio, and highest means of absorbed, trapped and transported electrons indicates another mechanism to regulate the photosynthetic rates; instead of heat dissipation or leaf reflectance, its physical strategy is to effectively absorb the light energy and maximize transport of electrons. This difference found in the chlorophyll index (Fig. 3) and the chlorophyll *a* fluorescence (Fig. 4) can be attributed to the structure of antenna complexes and energy dissipation more than the level of pigments.

Greater leaf areas potentially display higher mass fluxes, which, in turn, bring higher nutrient concentrations as a consequence (Donovan et al., 2011; Wright et al., 2004). Metabolic mechanisms such as photosynthetic nutrient use efficiency increase the energy spent in the photosynthetic process and keeping it high may be damaging rather than beneficial in the long run. This may be the cause of the nutrient efficiency decrease, as observed in *L. pinaster* that had the smallest area and highest PPUE. This is likely explained by the fact that metabolic mechanisms tend to increase the cost of energy.

It is also relevant to remark on the fact that even though two different measurements were made in the same month of two different years, we found differences in the nutritional concentration in the foliar tissues. This, coupled with the high levels of N and P we found indicate that the speed of nutrient cycling in these species may be faster than what is anticipated by the literature.

All of these characteristics and convergence of the functional traits in five different species indicate that there's a strong relation between the ecological filters and their successful establishment in the *campo rupestre*. This most likely shows that this particular environment is crucial in the process of selecting species and adaptations throughout generations and time. Therefore, we can assume that it's very likely that the environment's pressure is one of the most impactful factors in the pool of ecological filters.

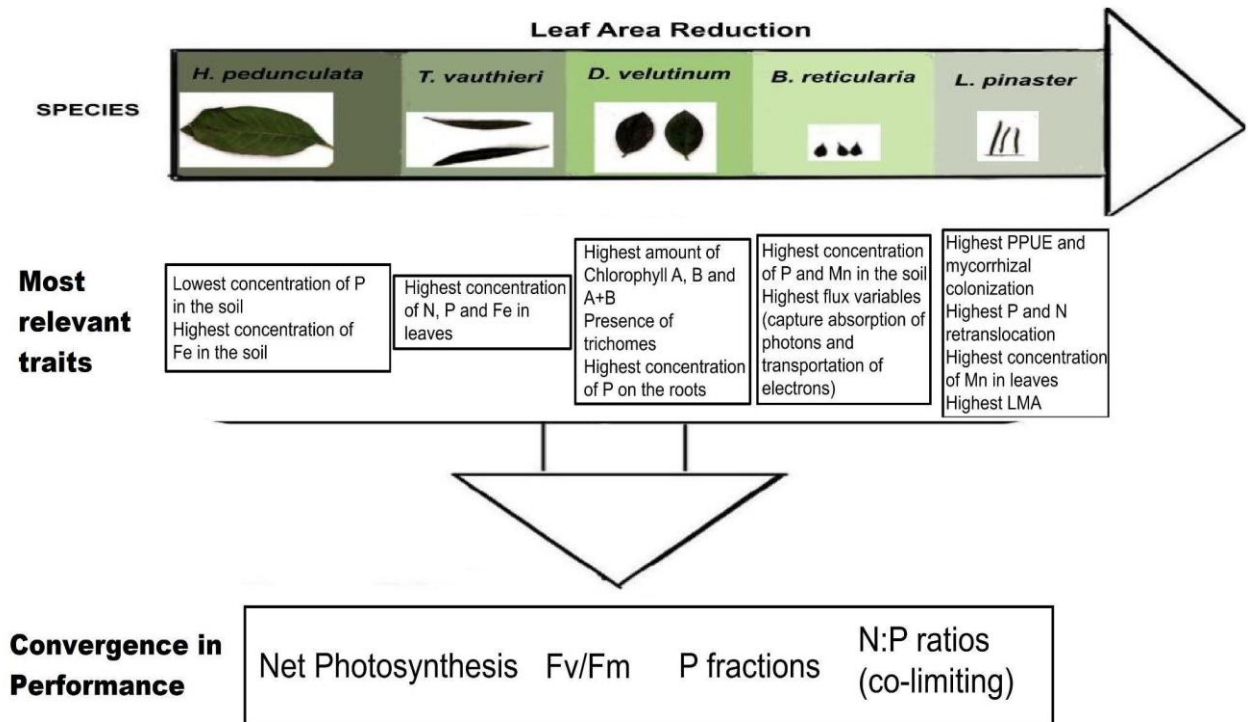


Figure 7: Summary on how functional traits diversity per species converges to the physiological performance in Asteraceae species from ferruginous *campo rupestre*. P, phosphorus; N, nitrogen; Mn, manganese; Fe, iron. LMA, leaf mass per area; PPUE, photosynthetic P use efficiency; Fv/Fm, maximum potential quantum efficiency of Photosystem II.

3.5 CONCLUSION

Our results indicate that there are no direct proportional relations between the phosphorus concentrations (both total and metabolic P fraction), other functional parameters and the photosynthetic rates of the five species. However, we have seen that biologically, these factors impact in very different ways the performance of the species, resulting in a broad diversity of functional attributes converging to a similar photosynthetic performance in the nutrient impoverished ferruginous *canga*.

The species' physiological performance, though there is little to none data to compare to our own, was very enlightening. All five species displayed different functional attributes in specific areas, which makes it even more compelling. For instance, even though AM species are, in literature, described as being inefficient in landscapes such as the *campo rupestre*, *L. pinaster* has shown high rates of mycorrhizal colonization that align with better nutrient photosynthetic use efficiency and even with nutrient retranslocation of P and N. This may indicate that there are

deeper relations between species, environment and ecological filters that are still unknown to us. This is particularly interesting in the regard of giving ground to restoration techniques, since this species cannot only be further studied for this purpose, but can also be used to bring awareness of preservation because it is a flag and symbol species of the state.

Our results show that the species have similar ecophysiological strategies in both photosynthesis and nutrition. The similarities in the variables measured could point to a convergence due to the strong environmental filter that exists in the *campo rupestre*. And while this adaptive convergence is very important in the establishment of the Asteraceae species in the *campo rupestre*, it can also mean vulnerability to them.

Further research is needed to better understand how the species can be used in the future as a part of ecological restoration techniques for degraded areas within environments with endemic biodiversity, therefore diminishing the risk of endangered and extinction of these species.

3.6 REFERENCES

- Abrahão, A., Lambers, H., Sawaya, A. C. H. F., Mazzafera, P., & Oliveira, R. (2014). Convergence of a specialized root trait in plants from nutrient-impooverished soils: phosphorus-acquisition strategy in a non mycorrhizal cactus. *Oecologia*, *176*(2), 345-355.
- Abrahão, A., Costa, P. D. B., Lambers, H., Andrade, S. A. L., Sawaya, A. C. H. F., Ryan, M. H., & Oliveira, R. S. (2019). Soil types select for plants with matching nutrients-acquisition and use traits in hyperdiverse and severely nutrient-impooverished campos rupestres and cerrado in Central Brazil. *Journal of Ecology*, *107*(3), 1302-1316.
- Abrahão, A., de Britto Costa, P., Teodoro, G. S., Lambers, H., Nascimento, D. L., Adrián López de Andrade, S., ... & Silva Oliveira, R. (2020). Vellozioid roots allow for habitat specialization among rock-and soil-dwelling Velloziaceae in campos rupestres. *Functional Ecology*, *34*(2), 442-457.
- Baião, É. E., Santos, C. H. B., Santos, A. H., Marques, G., Lima, J. C., Rigobelo, E. C., & Scotti, M. R. (2021). High C-and N-based soil fertility and microbial associations sustain the plant biodiversity of the campo rupestre in Brazil. *Geoderma Regional*, *25*, e00401.
- Bartolome, A. P., Villaseñor, I. M., & Yang, W. C. (2013). *Bidens pilosa* L.(Asteraceae): botanical properties, traditional uses, phytochemistry, and pharmacology. *Evidence-based complementary and alternative medicine*, 2013.
- Brundrett, M., Bougher, N., Dell, B., Grove, T., & Malajczuk, N. (1996). Working with mycorrhizas in forestry and agriculture (Vol. 32, p. 374). Canberra: Australian Centre for International Agricultural Research.
- Bussotti, F., Desotgiu, R., Pollastrini, M., & Cascio, C. (2010). The JIP test: a tool to screen the capacity of plant adaptation to climate change. *Scandinavian Journal of Forest research*, *25*(S8), 43-50.

Caminha-Paiva, D., Negreiros, D., Barbosa, M., & Fernandes, G. W. (2021). Functional trait coordination in the ancient and nutrient-impooverished campo rupestre: soil properties drive stem, leaf and architectural traits. *Biological Journal of the Linnean Society*, 133(2), 531-545.

Canton G.C., Bertolazzi A.A., Cogo A.J., Eutrópio F.J., Melo J., de Souza S.B., Krohling A. C., Campostrini E., da Silva A.G., Façanha A.R., Sepúlveda N., Cruz C., Ramos A.C. (2016). Biochemical and ecophysiological responses to manganese stress by ectomycorrhizal fungus *Pisolithus tinctorius* and in association with *Eucalyptus grandis*. *Mycorrhiza*. Jul;26(5):475-87. doi: 10.1007/s00572-016-0686-3. Epub 2016 Feb 10. PMID: 26861483.

Carmo, C. D. S., de Araujo, W. S., Bernardi, A. D. C., & Saldanha, M. F. C. (2000). Métodos de análise de tecidos vegetais utilizados na Embrapa Solos.

Carmo, F. F. D., & Jacobi, C. M. (2013). Canga vegetation in the Iron Quadrangle, Minas Gerais: characterization and phytogeographical context. *Rodriguésia*, 64, 527-541.

Chuyong, G. B., Newbery, D. M., & Songwe, N. C. (2000). Litter nutrients and retranslocation in a central African rain forest dominated by ectomycorrhizal trees. *New Phytologist*, 148(3), 493-510.

Cornelissen, J. H. C., Lavorel, S., Garnier, E., Díaz, S., Buchmann, N., Gurvich, D. E., ... & Poorter, H. (2003). A handbook of protocols for standardized and easy measurement of plant functional traits worldwide. *Australian journal of Botany*, 51(4), 335-380.

De Lima, R. L., Severino, L. S., Cazetta, J. O., De Azevedo, C. A., Sofiatti, V., & Arriel, N. H. (2011). Redistribuição de nutrientes em folhas de pinhão-mansão entre estádios fenológicos. *Revista Brasileira de Engenharia Agrícola e Ambiental-Agriambi*, 15(11).

de Oliveira Joaquim, E., Silva, T. M., Figueiredo-Ribeiro, R. D. C. L., de Moraes, M. G., & de Carvalho, M. A. M. (2018). Diversity of reserve carbohydrates in herbaceous species from Brazilian campo rupestre reveals similar functional traits to endure environmental stresses. *Flora*, 238, 201-209.

Donovan, L. A., Maherali, H., Caruso, C. M., Huber, H., & de Kroon, H. (2011). The evolution of the worldwide leaf economics spectrum. *Trends in Ecology & Evolution*, 26(2), 88-95.

Epstein E. & Bloom A.J. (2005) Mineral Nutrition of Plants: Principles and Perspectives. Sinauer, Sunderland, MA.

Folli-Pereira, M. D. S., Meira-Haddad, L. S. A., Bazzolli, D. M. S., & Kasuya, M. C. M. (2012). Micorriza arbuscular e a tolerância das plantas ao estresse. *Revista Brasileira de Ciência do Solo*, 36, 1663-1679.

Gao, Y., Zhang, Z., Zhang, B., Yin, H., Chai, X., Xu, M., ... & Zeng, F. (2022). Foliar P-Fractions Allocation of *Karelinia caspia* and *Tamarix ramosissima* Are Driven by Soil and Groundwater Properties in a Hyper-Arid Desert Ecosystem. *Frontiers in plant science*, 13, 833869.

Godoy, O., Gómez-Aparicio, L., Matías, L., Pérez-Ramos, I.M. & Allan, E. (2020) An excess of niche differences maximizes ecosystem functioning. *Nature Communications*, 11, 4180. <https://doi.org/10.1038/s41467-020-17960-5>.

Gomes, V. M., Assis, I. R., Hobbs, R. J., & Fernandes, G. W. (2021). Glomalin-Related Soil Protein Reflects the Heterogeneity of Substrate and Vegetation in the campo rupestre Ecosystem. *Journal of Soil Science and Plant Nutrition*, 1-11.

Guilherme Pereira, C., Hayes, P. E., O'Sullivan, O. S., Weerasinghe, L. K., Clode, P. L., Atkin, O. K., & Lambers, H. (2019). Trait convergence in photosynthetic nutrient-use efficiency along a 2-million year dune chronosequence in a global biodiversity hotspot. *Journal of Ecology*, 107(4), 2006-2023.

Güsewell, S. (2004). N: P ratios in terrestrial plants: variation and functional significance. *New phytologist*, 164(2), 243-266.

Han, Z., Shi, J., Pang, J., Yan, L., Finnegan, P. M., & Lambers, H. (2021). Foliar nutrient allocation patterns in *Banksia attenuata* and *Banksia sessilis* differing in growth rate and adaptation to low-phosphorus habitats. *Annals of Botany*, 128(4), 419-430.

Hayes, P., Turner, B. L., Lambers, H., & Laliberté, E. (2014). Foliar nutrient concentrations and resorption efficiency in plants of contrasting nutrient-acquisition strategies along a 2-million-year dune chronosequence. *Journal of Ecology*, 102(2), 396-410.

Heilmeyer, H. (2019). Functional traits explaining plant responses to past and future climate changes. *Flora*, 254, 1-11.

Hernández, I., & Munné-Bosch, S. (2015). Linking phosphorus availability with photo-oxidative stress in plants. *Journal of experimental botany*, 66(10), 2889-2900.

Hidaka, A., & Kitayama, K. (2011). Allocation of foliar phosphorus fractions and leaf traits of tropical tree species in response to decreased soil phosphorus availability on Mount Kinabalu, Borneo. *Journal of Ecology*, 99(3), 849-857.

Kottek, M., Grieser, J., Beck, C., Rudolf, B., & Rubel, F. (2006). World map of the Köppen-Geiger climate classification updated.

Lambers, H., Hayes, P. E., Laliberte, E., Oliveira, R. S., & Turner, B. L. (2015). Leaf manganese accumulation and phosphorus-acquisition efficiency. *Trends in plant science*, 20(2), 83-90.

Lambers, H., de Britto Costa, P., Cawthray, G. R., Denton, M. D., Finnegan, P. M., Hayes, P. E., ... & Zhong, H. (2022). Strategies to acquire and use phosphorus in phosphorus-impooverished and fire-prone environments. *Plant and Soil*, 1-28.

Li, L., Li, S. M., Sun, J. H., Zhou, L. L., Bao, X. G., Zhang, H. G., & Zhang, F. S. (2007). Diversity enhances agricultural productivity via rhizosphere phosphorus facilitation on

phosphorus-deficient soils. *Proceedings of the National Academy of Sciences*, 104(27), 11192-11196.

Machado, I. F., & Figueirôa, S. F. D. M. (2001). 500 years of mining in Brazil: a brief review. *Resources Policy*, 27(1), 9-24.

Mateo, N., Nader, W., & Tamayo, G. (2001). Bioprospecting. *Encyclopedia of biodiversity*, 1, 471-487.

Medeiros, D., de Senna Valle, L., & Alves, R. J. V. (2008). Euphorbiaceae nativas de cerrado e campo rupestre da Serra de São José, Minas Gerais, Brasil. *Arquivos do Museu Nacional*, 66(2).

Mo, Q., Li, Z. A., Sayer, E. J., Lambers, H., Li, Y., Zou, B. I., ... & Wang, F. (2019). Foliar phosphorus fractions reveal how tropical plants maintain photosynthetic rates despite low soil phosphorus availability. *Functional Ecology*, 33(3), 503-513.

Morellato, L. P. C., & Silveira, F. A. (2018). Plant life in campo rupestre: new lessons from an ancient biodiversity hotspot. *Flora*, 238, 1-10.

Mourão, A., & Stehmann, J. R. (2007). Levantamento da flora do campo rupestre sobre canga hematítica couraçada remanescente na Mina do Brucutu, Barão de Cocais, Minas Gerais, Brasil. *Rodriguésia*, 58, 775-786.

Mitre, S. K., Mardegan, S. F., Caldeira, C. F., Ramos, S. J., Furtini Neto, A. E., Siqueira, J. O., & Gastauer, M. (2018). Nutrient and water dynamics of Amazonian canga vegetation differ among physiognomies and from those of other neotropical ecosystems. *Plant Ecology*, 219(11), 1341-1353.

Negreiros D, Le Stradic S, Fernandes GW, Rennó HC (2014) CSR analysis of plant functional types in highly diverse tropical grasslands of harsh environments. *Plant Ecol* 215:379–388.

Nkrumah, P. N., Echevarria, G., Erskine, P. D., Chaney, R. L., Sumail, S., & van der Ent, A. (2021). Contrasting phosphorus (P) accumulation in response to soil P availability in 'metal crops' from P-impooverished soils. *Plant and Soil*, 467(1), 155-164.

Osnas, J. L. D., Lichstein, J. W., Reich, P. B., & Pacala, S. W. (2013). Global Leaf Trait Relationships: Mass, Area, and the Leaf Economics Spectrum. *Science*, 340(6133), 741–744. doi:10.1126/science.1231574.

Oliveira, R. S., Galvão, H. C., de Campos, M. C., Eller, C. B., Pearse, S. J., & Lambers, H. (2015). Mineral nutrition of campos rupestres plant species on contrasting nutrients-impooverished soil types. *New Phytologist*, 205(3), 1183-1194.

Oliveira, R. S., Abrahão, A., Pereira, C., Teodoro, G. S., Brum, M., Alcantara, S., & Lambers, H. (2016). Ecophysiology of campos rupestres plants. In *Ecology and conservation of mountaintop grasslands in Brazil*. Springer, Cham. (p. 227-272).

Ramos, A. C., Façanha, A. R., Palma, L. M., Okorokov, L. A., Cruz, Z., Silva, A. G., ... & Okorokova-Façanha, A. L. (2011). An outlook on ion signaling and ionome of mycorrhizal symbiosis. *Brazilian Journal of Plant Physiology*, 23, 79-89.

Reich, P. B. (2014). The world-wide 'fast–slow' plant economics spectrum: a traits manifesto. *Journal of ecology*, 102(2), 275-301.

Rios, C. O., Pimentel, P. A., Živčák, M., Brestič, M., & Pereira, E. G. (2022). Can ecological strategies be explained by photochemical efficiency in ironstone outcrops vegetation?. *Plant and Soil*, 1-16.

Salomons, W. (1995). Environmental impact of metals derived from mining activities: Processes, predictions, prevention. *Journal of Geochemical Exploration*, 52(1-2), 5–23. doi:10.1016/0375-6742(94)00039-e.

Santos, L. J. S., Arantes, A. D., Donato, S. L. R., Brito, C. F. B., Lima, M. A. C. D., & Rodrigues Filho, V. A. (2020). Leaf contents and biochemical cycling of nutrients in accessions of umbu and umbu-caja. *Revista Caatinga*, 33, 690-701.

Silva, F. D., Eira, P. D., Barreto, W. D. O., Pérez, D. V., & Silva, C. A. (1998). Manual de métodos de análises químicas para avaliação da fertilidade do solo. Rio de Janeiro, Embrapa-CNPQ.

Silva, K. G. D., Silva, E. A. D., Ferreira, M. L., Kanashiro, S., Camargo, P. B. D., & Tavares, A. R. (2017). Gas exchange and chlorophyll a fluorescence parameters of ornamental bromeliads. *Ornamental Horticulture*, 23(4), 400-406.

Silva, P. S. S. D., Vieira, M. A. R., & Marques, M. O. M. (2018). *Lychnophora pinaster* Mart. In *Medicinal and Aromatic Plants of South America* (p. 309-316). Springer, Dordrecht.

Silveira, F. A., Negreiros, D., Barbosa, N. P., Buisson, E., Carmo, F. F., Carstensen, D. W., ... & Lambers, H. (2016). Ecology and evolution of plant diversity in the endangered campo rupestre: a neglected conservation priority. *Plant and soil*, 403(1), 129-152.

Smith, M., Wu, Y., & Green, O. (1993). Effect of light and water-stress on photosynthesis and biomass production in *Boltonia decurrens* (Asteraceae), a threatened species. *American Journal of Botany*, 80(8), 859-864.

Soudzilovskaia, N. A., van Bodegom, P. M., Terrer, C., van't Zelfde, M., McCallum, I., McCormack, M. L., ... & Tedersoo, L. (2019). Global mycorrhizal plant distribution linked to terrestrial carbon stocks. *Nature communications*, 10(1), 1-10.

Sun, F., Zeng, L., Cai, M., Chauvat, M., Forey, E., Tariq, A., ... & Wang, M. (2022). An invasive and native plant differ in their effects on the soil food-web and plant-soil phosphorus cycle. *Geoderma*, 410, 115672.

Sun, J., Rutherford, S., Saif Ullah, M., Ullah, I., Javed, Q., Rasool, G., ... & Du, D. (2022). Plant–soil feedback during biological invasions: effect of litter decomposition from an invasive plant (*Sphagneticola trilobata*) on its native congener (*S. calendulacea*). *Journal of Plant Ecology*, 15(3), 610-624.

Vergutz, L., Manzoni, S., Porporato, A., Novais, R. F., & Jackson, R. B. (2012). Global resorption efficiencies and concentrations of carbon and nutrients in leaves of terrestrial plants. *Ecological Monographs*, 82(2), 205-220.

Veneklaas, E. J. (2022). Phosphorus resorption and tissue longevity of roots and leaves—importance for phosphorus use efficiency and ecosystem phosphorus cycles. *Plant and Soil*, 1-11.

Viana, P. L., & Lombardi, J. A. (2007). Florística e caracterização dos campos rupestres sobre canga na Serra da Calçada, Minas Gerais, Brasil. *Rodriguésia*, 58(1), 157-177.

Westoby, M., & Wright, I. J. (2006). Land-plant ecology on the basis of functional traits. *Trends in ecology & evolution*, 21(5), 261-268.

Wright, I. J., Reich, P. B., Westoby, M., Ackerly, D. D., Baruch, Z., Bongers, F., ... & Villar, R. (2004). The worldwide leaf economics spectrum. *Nature*, 428(6985), 821-827.

Yuan, Z., & Chen, H. Y. (2009). Global trends in senesced-leaf nitrogen and phosphorus. *Global Ecology and Biogeography*, 18(5), 532-542.

Zhang, L., Luo, X., Lambers, H., Zhang, G., Liu, N., Zang, X., ... & Wen, D. (2021). Effects of elevated CO₂ concentration and nitrogen addition on foliar phosphorus fractions of *Mikania micranatha* and *Chromolaena odorata* under low phosphorus availability. *Physiologia Plantarum*, 173(4), 2068-2080.

Zhongming H., Jianmin S., Jiayin P., et al. (2021). Foliar nutrient-allocation patterns in *Banksia attenuata* and *Banksia sessilis* differing in growth rate and adaptation to low-phosphorus habitats. *Authorea*.

3.7 SUPPLEMENTARY MATERIAL

Table S1: Nutrient content in soil and leaves of *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. P, phosphorus; N, nitrogen; Mn, Manganese; Fe, Iron. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$). These values are the means of the ten repetitions per species. Mean \pm standard error.

Species	<i>H. pedunculata</i>	<i>T. vauthieri</i>	<i>D. velutinum</i>	<i>B. reticularia</i>	<i>L. pinaster</i>
P (soil) mg/dm ³	2.26 \pm 0.40 b	2.67 \pm 0.57 b	4.02 \pm 1.59 b	4.65 \pm 1.69 a	2.99 \pm 0.63 b
Mn (soil) mg/dm ³	52.62 \pm 27.32 b	68.57 \pm 42.73 b	64.17 \pm 27.30 b	90.15 \pm 70.85 a	43.43 \pm 25.84 b
Fe (soil) mg/dm ³	315.43 \pm 40.12 a	312.25 \pm 122.26 b	203.71 \pm 43.66 b	201.09 \pm 56.28 b	218.21 \pm 51.05 b
Fe (leaves) mg/Kg	1816.51 \pm 994.71 b	6255.34 \pm 811.82 a	2716.88 \pm 1161.48 b	1261.07 \pm 567.88 b	2232.17 \pm 1071.21 b
Mn (leaves) mg/Kg	436.93 \pm 135.25 b	1008.17 \pm 152.84 b	679.20 \pm 167.07 b	1054.66 \pm 277.54 b	1367.79 \pm 848.39 a

Table S2: Senescent leaves content evaluation and N:P ratios of *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. P, phosphorus; N, nitrogen. Different letters indicate different means according to statistical test (Duncan or Dunnet) at the 5% confidence level ($p \leq 0,05$). These values are the means of the ten repetitions per species. Mean \pm standard error.

Species	<i>H. pedunculata</i>	<i>T. vauthieri</i>	<i>D. velutinum</i>	<i>B. reticularia</i>	<i>L. pinaster</i>
N on old leaves	14.16 \pm 2.01 b	21.57 \pm 3.44 a	17.96 \pm 3,59 b	19.09 \pm 2.86 b	10.94 \pm 1.49 b
P on old leaves	0.59 \pm 0.15 b	0.934 \pm 0.22 b	0.88 \pm 0.12 b	0.98 \pm 0.22 a	0.42 \pm 0.07 b
N:P ratios on old leaves	24.85 \pm 5.41 b	23.59 \pm 2.5 b	20.34 \pm 2.65 b	20.31 \pm 2.003 b	26.31 \pm 2.73 a

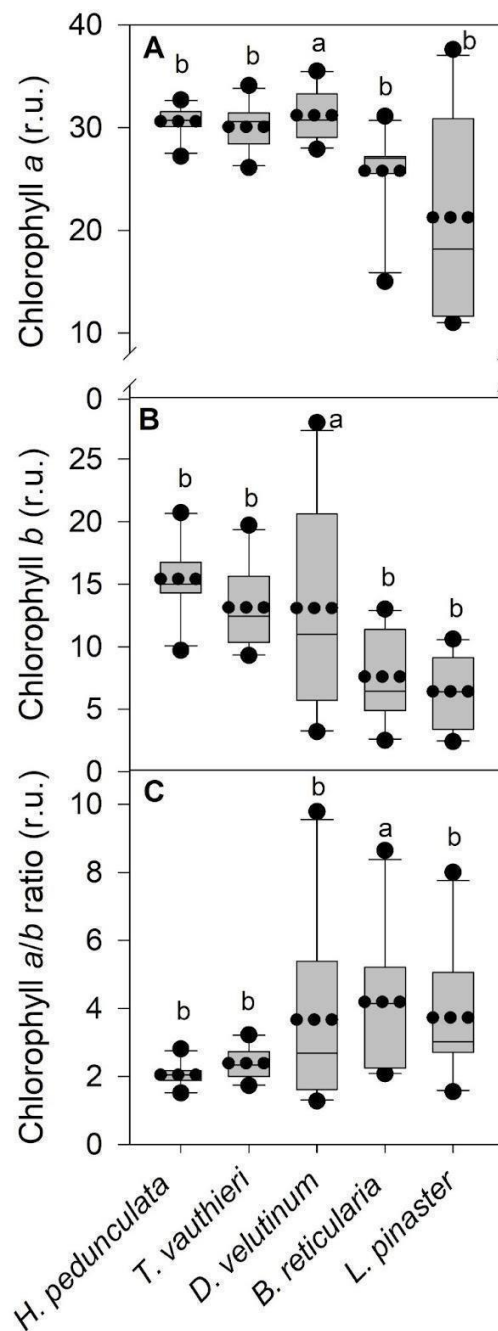


Figure S1: Boxplot of Chlorophyll a (A), Chlorophyll b (B), Chlorophyll a/b ratio (C) in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. The median is shown by a straight line, the mean by the dotted line and the outliers individuals that stand out from the means. Different letters indicate different means according to the performed statistical test at 5% confidence level ($p \leq 0,05$).

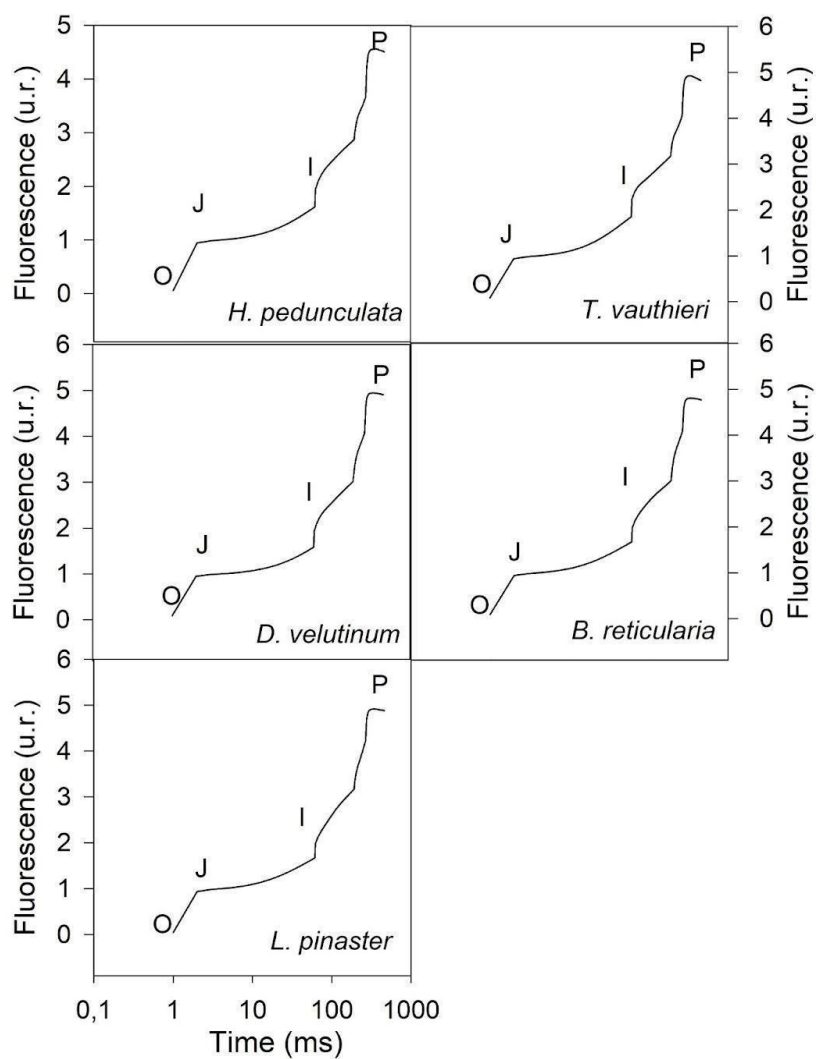


Figure S2: Curve lines graphic of OJIP (different steps of the polyphasic fluorescence transient) curves generated during the JIP test measurements in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*.

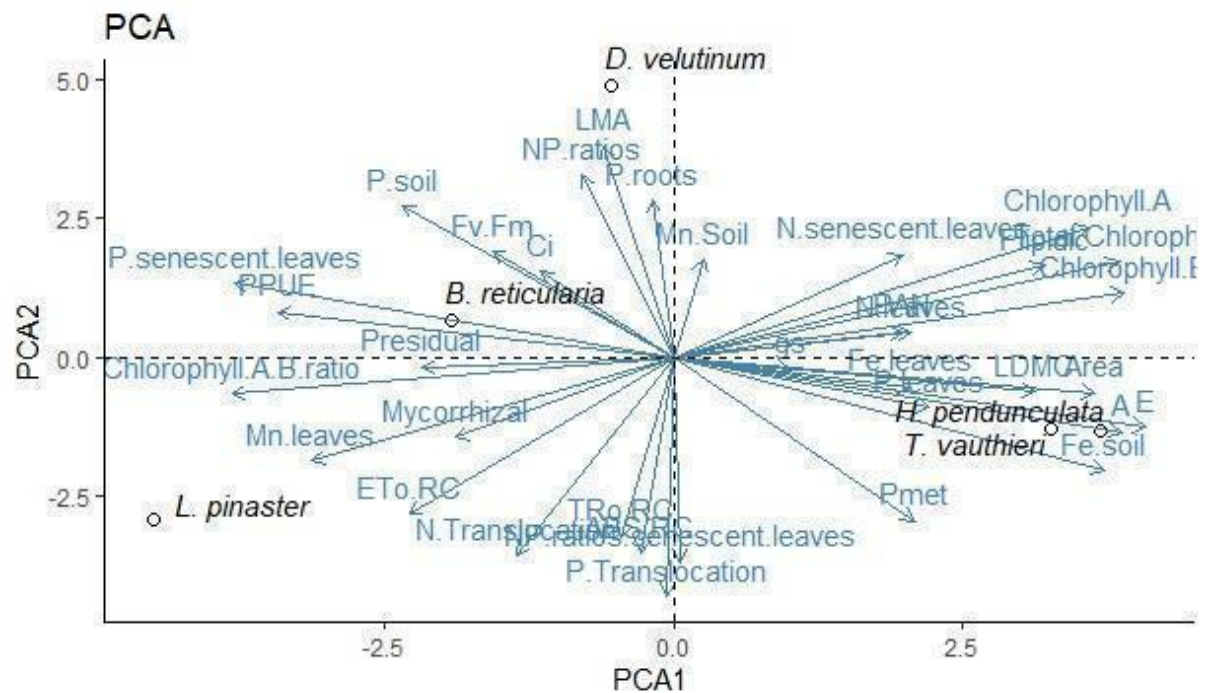


Figure S3: Principal component analysis (PCA) analysis using all the measurements performed during the study period in *Hololepis pedunculata*, *Trixis vauthieri*, *Dasyphyllum velutinum*, *Baccharis reticularia* and *Lychnophora pinaster*. N, nitrogen; P, phosphorus; Fe, iron; Mn, manganese; LMA, leaf mass per area; LDMC, leaf dry mass content; Area, foliar area; A, net photosynthesis; *gs*, stomatal conductance; *Ci*, internal carbon; *E*, transpiration; *Fv/Fm*, maximum potential quantum efficiency of Photosystem II; PPUE, Photosynthetic phosphorus-use efficiency; ABS/RC, electrons absorbed by active reactive center; TRo/RC, electrons captured by active reactive center; ETo/RC, electrons transported by active reactive center; Pmet, Metabolic Phosphorus Fractions; Nucleic Acid P, Nucleic Acid Phosphorus Fraction; Residual P, Residual Phosphorus Fraction; Lipidic P, Lipidic Phosphorus Fraction; P retranslocation, Percentage of Translocated Phosphorus; N retranslocation, Percentage of Translocate Nitrogen; Mycorrhiza, percentage of mycorrhizal colonization; P senescent leaves, phosphorus concentration on senescent leaves; N senescent leaves, nitrogen phosphorus concentration on senescent leaves; P soil, phosphorus concentration on the soil; Mn soil, manganese concentration on the soil; Fe soil; iron concentration on the soil; P leaves, phosphorus concentration on young leaves; N leaves, nitrogen concentration on young leaves; Mn leaves, manganese concentration on young leaves.

4. GENERAL CONCLUDING REMARKS

The Brazilian *canga* is part of the OCBIL's environments and, yet, we noticed a lack of specific information about these five chosen species in their natural habitat. We did find information on distribution and ecological aspects; however, there is a great gap of data regarding its functional and physiological responses.

These environments suffer from great anthropological pressures, including (but not limited to) biodiversity, illegal extraction and industrial activities such as mining. On top of that, climatic changes. In such threatened environments, the risk of losing biodiversity is great. Therefore, it is necessary that we conduct research on these sites to better understand its biodiversity before it is lost. This present work intended to better characterize and understand five native species of a nutrient impoverished landscape. In this area, there's also a great biodiversity and endemism rate of species in ferruginous *campo rupestre*. There's also a great functional traits' diversity and the Asteraceae family is one of the most representative in the *canga* landscape.

Although forestry environments have plenty of data published, when we look at grasslands such as the *canga*, there is more unknown about the physiological strategies in response to the poor nutrient availability than the opposite. This not only makes the data more intriguing but we also need to pay closer attention to how the experiments unveil. We also have a fine chance to confront literary indications regarding other species of the same (and other) botanic families and come to conclusions for that particular species in that particular environment.

Beyond the obvious need to further basic research in this area for purely descriptive reasons, there is an underlying need to do so for not only scientific reasons but also economic ones, including how can we use endemic species to mitigate impacts in their own environment and similar ones, preserving better the original landscape prior to the impact.