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Morphological and physiological characteristics of two neotropical plants in *veredas* with different levels of drying

Características morfológicas e fisiológicas de duas plantas neotropicais em veredas com diferentes níveis de secamento

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ABSTRACT

Objective: The propose of this study was to investigate whether the species *Copaifera oblongifolia* (Fabaceae) and *Macairea radula* (Melastomataceae), occurring in *veredas* with different levels of drying, show differences in plant architecture (morphological traits) and foliar compound levels (physiological characteristics). **Methods:** Sampling was conducted in two *veredas*, which are Vereda da Almescla (preserved *vereda*) and Vereda do Peruaçu (degraded *vereda* in an advanced drying stage) located in the northern region of Minas Gerais, Brazil. Data on morphological traits (height, circumference above ground, and canopy width) and foliar compounds (concentrations of chlorophyll, nitrogen, and flavonols) were collected from 15 individuals of each species in each *vereda*. **Results:** The results showed that the structural characteristics of the studied species did not differ between the analyzed *veredas*. Similarly, foliar compounds did not exhibit significant differences in their concentrations among the plants from the two *veredas* studied. **Conclusion:** These results suggest that the water stress present in the *veredas* does not seem to influence the morphological characteristics and foliar compounds of neotropical species. This is the first study evaluating the impacts of *vereda* drying on morphological and physiological attributes of plants.

Keywords: Anthropization; Palm swamps, Plant compounds; Structural parameters; Water stress

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RESUMO

Objetivo: investigar se as espécies *Copaifera oblongifolia* (Fabaceae) e *Macairea radula* (Melastomataceae), ocorrendo em veredas com diferentes níveis de secamento, apresentam diferenças na arquitetura das plantas (características morfológicas) e nos níveis de compostos foliares (características fisiológicas). **Métodos:** A amostragem foi realizada em duas veredas, que são Vereda da Almescla (vereda preservada) e Vereda do Peruaçu (vereda degradada em estágio avançado de secagem) localizadas na região norte de Minas Gerais, Brasil. Dados sobre características morfológicas (altura, circunferência acima do solo e largura da copa) e compostos foliares (concentrações de clorofila, nitrogênio e flavonóis) foram coletados de 15 indivíduos de cada espécie em cada vereda. **Resultados:** Os resultados mostraram que as características estruturais das espécies estudadas não diferiram entre as veredas analisadas. Da mesma forma, os compostos foliares não apresentaram diferenças significativas em suas concentrações entre as plantas das duas veredas estudadas. **Conclusão:** Estes resultados sugerem que o estresse hídrico presente nas veredas não parece influenciar as características morfológicas e compostos foliares das espécies neotropicais. Este é o primeiro estudo que avalia os impactos da secagem da vereda nos atributos morfológicos e fisiológicos das plantas.

Palavras-chave: Antropização; Palmeiras, Compostos vegetais; Parâmetros estruturais; Estresse hídrico.

INTRODUCTION

Anthropogenic impacts can lead to alterations in natural ecosystems, which may even be irreversible in environments with low restoration capacity[1]. These impacts result in modifications to ecosystems, such as increased light and temperature levels, and decreased water availability, making the habitat less favorable and causing environmental stress for various species[2]. As an example of environmental stress, we can mention water stress, which occurs through a reduction in soil water availability, causing damage to plants and potentially affecting their growth and development[3]. Due to this water deficit, the plant undergoes changes in its physiology and biochemistry, developing mechanisms to withstand and adapt to this stress, such as osmotic adjustment, reduced productivity, smaller leaf production, and the presence of trichomes[4]. *Veredas* are highly sensitive to environmental changes and have a slow restoration process[5]. Due to this characteristic, when these vegetations are negatively impacted, these areas undergo a drying-out process. These actions can alter water flow through intense drainage in the *veredas*, leading to shifts in soil moisture and plant species that grow there. As these places transform from being wetlands to becoming drier, more open environments resembling to savannas[6].

The *veredas* constitute one of the physiognomies within the Cerrado biome. They are characterized by herbaceous-shrub vegetation interspersed with scattered trees, typically located near watercourses, with *Mauritia flexuosa* L.f. being the primary tree species defining the *vereda* ecosystem[7]. *Vereda* soils are hydromorphic, clayey, and peat-rich, with high concentrations of organic matter[8,9], resulting in a biodiverse ecosystem. They play important ecological, hydrological, and social roles, such as maintaining water resources near *veredas*, serving as ecological corridors for the protection of flora and fauna, and providing income for small communities near these vegetations[10]. In the northern region of Minas Gerais, *vereda* areas are highly degraded due to various factors, including excessive water body drainage, anthropogenic fires, agricultural and livestock use, and deforestation[5,11]. Given the context of hydrological stress that Brazilian *veredas* are facing, it is essential to understand the impacts that this can have on the morphology and physiology of plants in these ecosystems.

Plants, when exposed to environmental conditions and stresses, tend to modify their morphological and physiological characteristics in order to adapt to these stresses[12]. Studies have shown that plants can exhibit a range of variations in their functional traits, such as changes in plant height, leaf area, biomass, leaf morphology and coloration, root morphology and length, and reduced stem growth in response to light, water, and nutritional stresses[4,13-15]. These adaptations aim to support stress levels and enable better water and nutrient absorption for improved productivity and survival in these modified environments[14]. Based on this, it is expected that the hydrological stress may affect the fitness and development of plants, which can impact their morphological attributes, such as size and circumference.

In addition to morphological attributes, leaf compounds play important roles in plants. For example, phenolic compounds and the production of antioxidants act against oxidative stress caused by exposure to UV rays, alkaloids have important photoprotective components in their properties that help protect skin damage caused by UV radiation[16-17]. Consequently, variations in the concentration of certain compounds due to biotic or abiotic stress have been reported as good indicators of stress[18,19]. One example of such a compound is flavonols, which belong to the flavonoid group of secondary metabolites and are found in plant parts such as flowers and leaves[20]. When experiencing water stress conditions, some flavonols may play a greater role in detoxifying free radicals[21]. Chlorophylls are natural pigments found in the chloroplasts of leaves, mainly in the region of the thylakoid membrane[22]. Under high irradiation and reduced water availability, chlorophyll tends to decrease, and the photoprotection process is activated through electron transfer to carotenoids to prevent damage to the photosynthetic apparatus[23]. Nitrogen is of great importance to plants as they require it in large quantities[24]. It is incorporated into carbon compounds such as amino acids, pigments, and lipids, participating in various plant processes, with growth being the primary one[25]. Chlorophylls, being molecules formed by porphyrin-derived complexes with magnesium at the central atom, bound to four other nitrogen atoms, are related to nitrogen[24]. Part of the foliar nitrogen is integrated into chlorophyll molecules, so a decrease in nitrogen also leads to a decrease in chlorophyll. Thus, variation in foliar compounds, such as flavonols and chlorophylls, is expected to occur among plants exposed to different levels of water stress.

Several studies in different vegetation types have presented significant results regarding these changes occurring in plants due to water stress. A soybean cultivation study showed that when there was a reduction in water availability, plants from sprouting to flowering stages had their height significantly affected[26]. The same effect was observed in the study by Figueirôa *et al.*[27], where young individuals of the species *Myracrodruon urundeuva* exposed to a higher water deficit treatment showed greater biomass allocation to root growth rather than aerial parts. his occurs because the plant tends to prioritize water absorption, leading to the suppression of foliar growth. Additionally, it was also observed that a prolonged period of severe drought results in a reduction in the number of leaves and a decrease in chlorophyll content[27,28]. With drought stress, plants tend to increase the production of reactive oxygen species (ROS), causing the destruction of cell membranes and degradation of chloroplasts, resulting in the inhibition of chlorophyll synthesis[28]. These studies collectively highlight the impact of water stress on plant morphology and physiology, underscoring the relevance of understanding such dynamics in ecosystems like *veredas* facing dryness challenges. However, despite recent advancements in assessing the impacts of vereda drying on plant diversity[5], no previous study has evaluated whether the hydrological stress of *veredas* affects the morphological and physiological attributes of plants.

Studies analyzing the functional traits of plants in contrasting habitats are of great importance because they provide insights into how species survive in their typical habitats and how these species may adapt in the face of drastic changes in future climate scenarios[29]. There are few studies in the Neotropical region investigating the potential impacts of *vereda* degradation on morphological and physiological modifications in plants, such as variations in the concentration of plant compounds and plant structure. Thus, the objective the present study was to investigate whether there are differences in the structural characteristics and foliar compound concentrations of plants of *veredas* at different stages of drying, located in the northern region of Minas Gerais, Brazil. We tested the hypothesis that drying of the *veredas* makes the environment stressful, thereby promoting changes in plant physiology resulting in alterations in the concentration of foliar compounds and plant functional traits. Thus, it is expected that preserved *vereda* areas (i.e., with good water conditions) exhibit structurally more complex plants with higher concentrations of chlorophyll, nitrogen, and flavonols, whereas *veredas* in advanced stages of drying present fewer complex individuals with lower levels of foliar compounds.

METHODS

Study area

The study was conducted in two *vereda* areas located within of sampling sites of the Long-Term Ecological Research Program - Veredas project (PELD-VERE). The areas are located in the municipalities of Januária, Bonito de Minas, and Cônego Marinho, Minas Gerais, Brazil. The region is located in the Brazilian semi-arid zone, one of the areas with the most significant water deficit problems in the country. The climate in the region of these areas is considered a tropical dry climate, classified as *Aw* according to the Köppen classification, with well-defined rainy and dry seasons[30]. The average annual precipitation ranges from 242 to 461 mm, with rainfall concentrated from September to March, and the average temperature varies from 19°C to 26°C (FIG. 1). The studied region is situated within the Brazilian Cerrado biome, where various vegetation types coexist, and *veredas* typically occur as oases amidst vast areas of cerrado *sensu stricto*[31]. The areas are located at an altitude ranging from 800 to 900 meters above sea level.

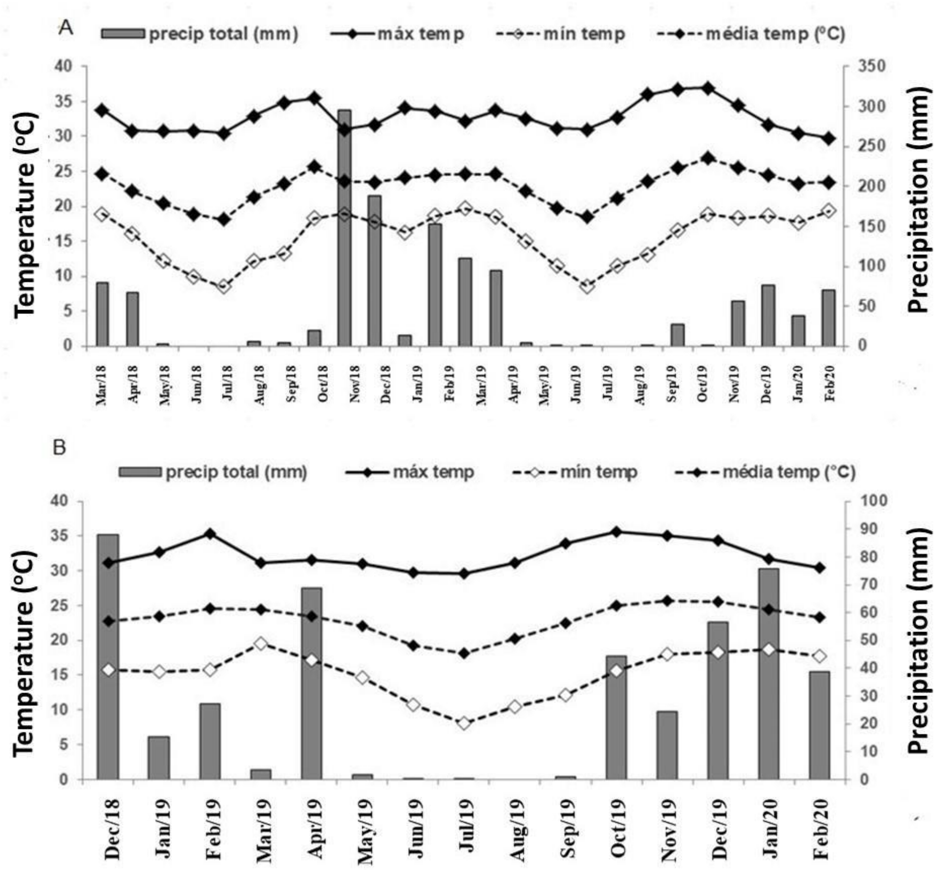

Figure 1. Average temperature and total precipitation data for the two study areas cover the years 2018-2021. A) Meteorological data from Vereda do Peruaçu; B) Meteorological data from Vereda da Almescla. Data sourced from Nunes *et al.* (2022).

Study was performed in two *vereda* areas in the northern region of Minas Gerais (FIG. 2A). The first area, known as Vereda do Peruaçu (14°59'58.2"S 44°42'40.2"W), is located within the Veredas do Peruaçu State Park, spanning the municipalities of Januária and Cônego Marinho. In this *vereda*, there was a significant drop in the water table and advanced soil drying, resulting in a degraded appearance (FIG. 2B). This degradation is evidenced by the high mortality of *Mauritia flexuosa* palms (buritis) and the invasion of cerrado *sensu stricto* plants within the *vereda*. These changes are attributed to factors such as climate change and anthropogenic activities, including disturbed soils, pastures, and wildfires[5]. Due to these characteristics, the Vereda do Peruaçu exhibits a high level of environmental stress. The second area, known as Vereda da Almescla (15°21'30.8"S 44°54'39.7"W), is located within the Rio Pandeiros Environmental Protection Area, in the municipality of Bonito de Minas[32]. The study site features a relatively well-preserved *vereda* environment with a low level of drying and the presence of above-ground water in some areas, categorizing this *vereda* as having a low environmental stress level (FIG. 2C). Each *vereda* studied encompasses an area of approximately 6 kilometers.

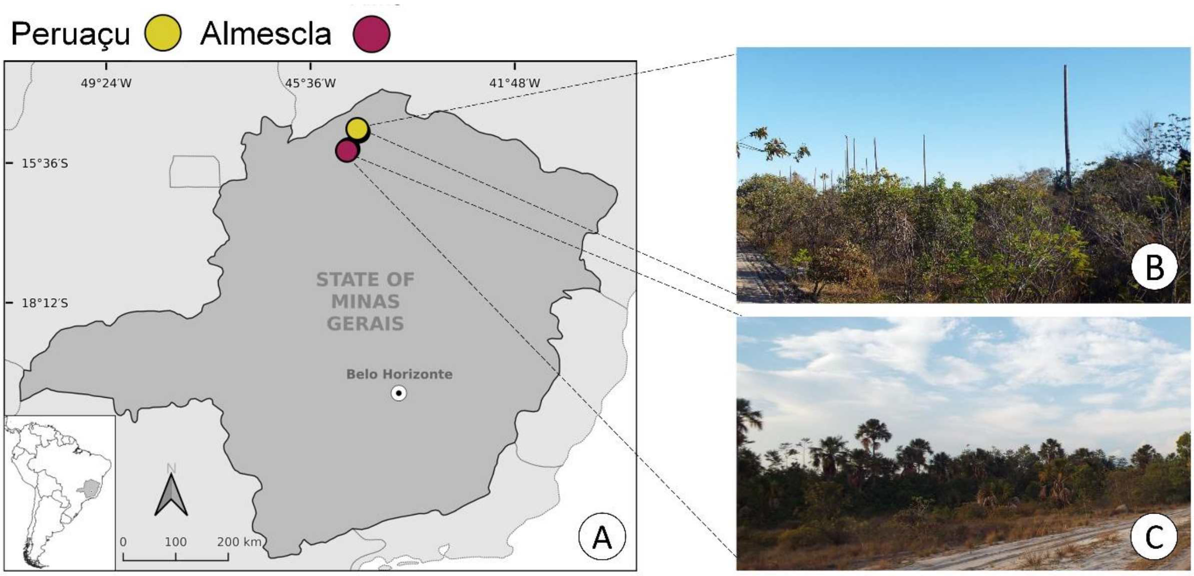

Figure 2. Characterization of the two *veredas* studied in the northern region of Minas Gerais, Brazil. A) Location of Vereda da Almescla and Vereda do Peruaçu; B) general aspect of Vereda do Peruaçu in an advanced stage of drying (degraded *vereda*) characterized by the advance of cerrado *sensu stricto* vegetation into the *vereda* and the high mortality of buriti palms due to advanced drying; and C) general aspect of Vereda da Almescla (preserved *vereda*), which features typical *vereda* vegetation with surrounding grassland and healthy buriti palms.

Studied plant species

For the study, two abundant plant species commonly found in both study areas were selected. The first species is *Copaifera oblongifolia* Mart. Ex Hayne, belonging to the Fabaceae family, which is a shrub commonly known as “pau d’ólinho” and is found in the North, Northeast, Central-West, and Southeast regions of Brazil, as well as in the Amazon and Cerrado biomes[30]. In the study areas, *C. oblongifolia* occurs in the vicinity of the *veredas* in cerrado *sensu stricto* vegetation zones. The species is characterized by branched individuals with a height of 2 to 3 meters, featuring sessile flowers, fruits, and seeds with orange aril coloration[33].

The other species is *Macairea radula* (Bonpl.) DC., belonging to the Melastomataceae family. This shrub is distributed in the North, Northeast, Central-West, and Southeast regions of Brazil, as well as in the Amazon, Caatinga, Cerrado, and Atlantic Forest biomes. The species is typically associated with moist soils and occurs on the edges and within the *veredas*. In the study areas, *M. radula* individuals were found on the *vereda*'s edge in a region with more humid soil. This species is characterized by branched individuals with a height of approximately 2 meters, featuring purple, lilac, or pink-colored flowers, fruits, and seed[34]. Thus, the two studied species occur in different vegetation zones (on the *vereda*'s edge and in the adjacent cerrado).

Sampling

In each *vereda*, a total of 15 individuals of *C. oblongifolia* and 15 individuals of *M. radula* were sampled in the morning and afternoon, resulting in a total of 30 individuals for each species. For each individual, using a measuring tape and a tape measure, the following morphological traits were measured: height (meters), circumference at breast height (CBH) (centimeters), and canopy width (meters). For the sampling of foliar compounds in the plants, readings were conducted using the Dualex device, which is an optical sensor developed by Force-A for measuring the content of flavonols[21] and chlorophylls[22] (μg/cm²) in the leaf epidermis. Additionally, the same was performed to access the Nitrogen Balance Index (NBI), which assesses leaf nitrogen conditions through the calculation of the ratio between chlorophyll and flavonols related to nitrogen/carbon allocation. All measurements were taken within the same time window (May 2021). The measurements were taken directly on the plant leaves in the field. Therefore, the architectural and compound data were collected in the field without the need to collect fragments from the individuals.

Data analyses

To compare the morphological traits (height, circumference at breast height, and canopy width) and foliar compounds (flavonols, chlorophylls, and nitrogen) of each plant species between the Vereda da Almescla (preserved *vereda*) and Vereda do Peruaçu (degraded *vereda*), we used Generalized Linear Models (GLM's) followed by ANOVA. The choice of GLM allows for the comparison of multiple morphological and foliar variables between areas with different conservation statuses, accommodating different data types and controlling for confounding variables. In all models, the *vereda* (Verada da Almescla vs. Vereda do Peruaçu) was included as a response variable. We checked the error distribution of the data and used a Poisson distribution for the morphological trait variables and a Gaussian distribution for the measurements of foliar compounds. Additionally, Spearman correlation tests were conducted to assess the correlation between foliar compounds for each plant species. For all analyses the significance level considered in the tests was $\alpha < 0.05$. All analyses were performed using R software version 4.1.1[35].

RESULTS

The height of *C. oblongifolia* individuals ranged from 1.33 to 5.50 meters (mean: 2.37 ± 0.91), while *M. radula* individuals ranged from 1.35 to 3.70 meters (mean: 2.08 ± 0.54). Circumference at breast height varied from 1.39 to 19.74 centimeters (10.12 ± 5.28) in *C. oblongifolia* individuals and from 3.20 to 23.44 centimeters (12.04 ± 4.32) in *M. radula* individuals. Canopy width ranged from 1.10 to 5.86 meters (3.34 ± 3.53) in *C. oblongifolia* and from 1.10 to 4.79 meters (1.91 ± 0.80) in *M. radula*. Comparing the analyzed structural variables (plant height, CAP, and canopy width), no significant differences were observed among individuals of each species in the two *veredas* (Tab. 1).

Table 1. Results of the models (GLM's) to compare the morphological characteristics of individuals of the species *Copaifera oblongifolia* and *Macairea radula* between Vereda da Almescla (preserved *vereda*) and Vereda do Peruaçu (degraded *vereda*), North of Minas, Brazil. Sum Sq: Sum of squares; Mean Sq: Mean square; F: F value; CBH: Circumference at breast height.

Plant species	Response variables	Vereda da Almescla (preserved *vereda*)	Vereda do Peruaçu (degraded *vereda*)	Sum *Sq*	Mean Sq	F	*P* value
Copaifera oblongifolia	Height (m)	2.21 ± 0.77	2.54 ± 1.03	0.83	0.830	1.006	0.324
	CBH (cm)	11.50 ± 5.35	8.74 ± 5.00	57.40	57.400	2.137	0.155
	Canopy Width (m)	3.96 ± 4.79	2.72 ± 1.41	11.60	11.610	0.932	0.343
Macairea radula	Height (m)	2.08 ± 0.66	2.18 ± 0.56	0.068	0.068	0.182	0.673
	CBH (cm)	12.30 ± 4.72	11.78 ± 4.03	2.01	1.975	0.103	0.751
	Canopy Width (m)	2.03 ± 0.90	1.80 ± 0.68	0.388	0.388	0.605	0.443

Among the sampled foliar compounds, only chlorophyll and nitrogen were positively correlated for both *C. oblongifolia* individuals ($R_{Spearman} = 0.90$; $p < 0.05$) and *M. radula* ($R_{Spearman} = 0.87$; $p < 0.05$). Regarding the analyzed leaf compounds, the environmental condition of the *veredas* did not influence the concentration of chlorophylls, flavonols, and foliar nitrogen in individuals of *C. oblongifolia* (Tab. 2). Similarly, no differences were observed in the concentrations of foliar compounds of the plant *M. radula* between the preserved and degraded *veredas*.

Table 2. Results of the models (GLM's) to test the concentration of leaf compounds in the species *Copaifera oblongifolia* and *Macairea radula* between Vereda da Almescla (preserved *vereda*) and Vereda do Peruaçu (degraded *vereda*), North of Minas, Brazil. Sum. Sq.: Sum of squares; Mean Sq: Mean square; F: F value.

Plant species	Response variables	Vereda da Almescla (preserved *vereda*)	Vereda do Peruaçu (degraded *vereda*)	Sum Sq	Mean Sq	F	*P* value
Copaifera oblongifolia	Chlorophylls (µg/cm²)	39.60 ± 4.60	40.63 ± 4.98	7.9	7.874	0.343	0.563
	Flavonoids (µg/cm²)	1.44 ± 0.07	1.42 ± 0.06	0.0	0.002	0.641	0.430
	Nitrogen (µg/cm²)	27.91 ± 3.92	28.88 ± 4.15	7.1	7.083	0.434	0.515
Macairea radula	Chlorophylls (µg/cm²)	38.95 ± 5.69	35.45 ± 4.18	86.5	86.540	3.471	0.073
	Flavonoids (µg/cm²)	1.46 ± 0.17	1.37 ± 0.15	0.2	0.191	1.178	0.287
	Nitrogen (µg/cm²)	29.13 ± 6.74	26.64 ± 4.66	19.4	19.360	0.548	0.465

DISCUSSION

The results indicate that the parameters of plant structure were not influenced by the level of *vereda* drying. The same pattern appears to occur in the concentration of foliar compounds, as no significant variation was found between plants in the preserved *vereda* and the *vereda* in an advanced stage of drying for both analyzed species. Previous studies have suggested structural differences among *vereda* plants under different levels of environmental disturbance[36,37]. For instance, Guimarães *et al.*[37] demonstrated that plants located in the preserved slope of the *vereda* had a higher average height compared to plants in the anthropized slope. The main explanation provided by the authors is the greater cattle grazing in anthropized areas, leading to vegetation removal. In the present study, no structural differences related to height, circumference at breast height, and canopy width were observed among plants in *veredas* under different water conditions. One possible explanation for this is that the analyzed plants exhibit significant intraspecific variation, with individuals showing substantial structural variation within the population.

In the present study, the concentration of foliar compounds did not differ between *C. oblongifolia* and *M. radula* plants. It is expected that, in response to water stress, the concentration of chlorophyll tends to decrease because this pigment decreases to allow carotenoids to assist in protecting the photosynthetic apparatus[23]. Since there is a high correlation between foliar nitrogen and photosynthetic pigments, as nitrogen is incorporated into the chlorophyll molecule[38], a decrease in nitrogen concentration should also occur. However, this pattern was not found in our study, as there was no difference in the amount of nitrogen and chlorophyll in individuals of the species between the *veredas*.

We believe that this result may be due to the it may be due to off-season rainfall in the degraded *vereda*. With this event, there was an increase in water availability, allowing plants to absorb water and nutrients, and consequently, not affecting physiological processes. Thus, with this environmental factor occurring, the levels of nitrogen and chlorophyll in individuals from the modified *vereda* may be similar to those in individuals from the preserved *vereda*. Therefore, the factor of rainfall in the modified *vereda* environment reduces water stress, thus causing no damage to photosynthetic processes.

Previous studies suggest that plants tend to decrease the levels of flavonols in response to water stress because the presence of this stress leads to a reduction in the enzymes of the shikimic acid and phenylpropanoid pathways, which in turn affects the production of certain flavonols[22]. However, this pattern was not supported in the present study, as there was also no difference in flavonol levels between the *veredas*. Since there was no change in chlorophyll concentration in either *vereda*, which would result in alterations in photosynthetic processes, plants do not need to accumulate flavonols to act in the photoprotection of photosynthetic processes.

The results of this study provide critical insights into the conservation of *veredas*, indicating that plant structure parameters and foliar compound concentrations were not influenced by the level of *vereda* drying. While previous studies have shown structural differences in plants under varying environmental disturbances, no significant differences were found between plants from preserved and degraded *veredas*. These findings suggest that short-term water availability fluctuations, such as recent rainfall, can significantly influence physiological responses to stress, highlighting the importance of considering temporal environmental factors in plant stress response studies and conservation strategies for *veredas*.

CONCLUSION

In this study, the structural characteristics (plant height, circumference at breast height, and canopy width) and foliar compounds (chlorophyll, nitrogen, and flavonols) of the species *C. oblongifolia* and *M. radula* did not differ between the *veredas* with different levels of drying. These results suggest that the water stress present in the areas was alleviated by the rainfall that occurred in the month prior to the collection, affecting our measurements in both species. Therefore, we suggest conducting studies at the site during other times of the year or new studies with different analytical approaches (e.g., histochemical and laboratory photochemical analyses), in other locations (e.g., other *veredas* with different levels of drying), or even with different plant species to determine if this pattern is repeated in other systems.

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