UNIVERSIDADE FEDERAL DE LAVRAS

RAFAEL COSTA CARDOSO

FLORA CAVERNÍCOLA: DIVERSIDADE E ECOLOGIA DAS COMUNIDADES VEGETAIS EM CAVERNAS

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Tese apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Ecologia Aplicada, Área de concentração em Ecologia e Conservação de Recursos em Paisagens Fragmentadas e Agrossistemas para a obtenção do título de Doutor.

Prof. Dr. Rodrigo Lopes Ferreira
Orientador

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CAVE FLORA: DIVERSITY AND ECOLOGY OF PLANT COMMUNITIES IN CAVES

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À minha avó Dinah (in memorian) por todo carinho, apoio e incentivo em vida.

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“Viva aceso, olhando e conhecendo o mundo que o rodeia, aprendendo

como um índio (...) seja um índio na sabedoria” (Darcy Ribeiro)

RESUMO

Os ecossistemas cavernícolas, com suas condições ambientais específicas, abrigam uma biodiversidade singular caracterizada por traços evolutivos e adaptativos únicos. Apesar de sua relevância, a pesquisa sobre a flora de cavernas, especialmente em ecossistemas tropicais, é escassa. Esta tese buscou investigar a ecologia da flora cavernícola e os fatores que influenciam a distribuição, riqueza e composição das comunidades vegetais ao longo do gradiente de luz nas entradas de cavernas em um ecossistema montano neotropical. O objetivo foi entender os padrões ecológicos das plantas cavernícolas, contribuindo para a redução dessa lacuna de conhecimento e promovendo estratégias de conservação. Realizamos coletas em entradas de cavernas no Parque Estadual do Ibitipoca (PEIB), em Minas Gerais, Brasil, uma área protegida com rica vegetação nativa e numerosas cavernas em rochas siliciclásticas. Utilizando um luxímetro, delimitamos zonas de luminosas até a região da penumbra escura em que foram analisadas a vegetação, solo e variáveis ambientais. Nossos resultados revelaram 134 espécies de plantas, sendo as angiospermas as mais diversas nas zonas fóticas e as briófitas nas zonas disfóticas. Observamos uma diminuição da riqueza de espécies com a profundidade e diferenças significativas na composição entre cavernas, entradas e zonas de luz. A análise do solo identificou possíveis variáveis que influenciam a estrutura das comunidades vegetais. Além disso, aprofundamos nosso entendimento sobre as comunidades de briófitas associadas às cavernas brasileiras, examinando como as características geomorfológicas das cavernas, altitude e distância entre entradas afetam a diversidade de briófitas. Em treze entradas de cavernas no PEIB, identificamos 70 espécies de briófitas, incluindo 14 novos registros para a área. As formas de vida mais comuns foram as folhosas, seguidas por formas de tapete, tufos e tramas. A pesquisa destacou que a proximidade geográfica influencia a similaridade filogenética das comunidades de briófitas, com entradas maiores suportando maior diversidade. Análises de diversidade *zeta* mostraram que espécies raras impulsionam a variação nas comunidades, evidenciando a importância das cavernas como reservatórios genéticos e refúgios potenciais para espécies raras. Estes achados ressaltam a importância ecológica e a diversidade da flora cavernícola nos Neotropicos, evidenciando a necessidade de estratégias de conservação que incluam as comunidades vegetais subterrâneas.

Palavras-chave: ambientes cársticos; flora cavernícola; ecossistemas subterrâneos; espeleobotânica; conservação.

ABSTRACT

Cave ecosystems, with their specific environmental conditions, harbor unique biodiversity characterized by distinctive evolutionary and adaptive traits. Despite their significance, research on cave flora, especially in tropical ecosystems, is scarce. This thesis investigates the ecology of cave flora, focusing on the factors influencing the distribution, richness, and composition of plant communities along the light gradient at cave entrances in a neotropical montane ecosystem. The aim is to understand the ecological patterns of cave-dwelling plants, contributing to closing this knowledge gap and promoting conservation strategies. We conducted sampling at cave entrances in Ibitipoca State Park (PEIB) in Minas Gerais, Brazil, a protected area known for its diverse vegetation and numerous quartzite caves. Using a luxmeter, we defined zones of light up to the dark penumbra region and analyzed vegetation structure, soil and environmental variables. Our results revealed 134 plant species, with angiosperms being the most diverse group in photic zones and bryophytes in dysphotic zones. Species richness decreased with depth, and significant differences in composition were observed among caves, entrances, and light zones. Soil analysis identified key variables influencing plant community structure. Additionally, we deepened our understanding of the bryoflora associated with Brazilian caves, examining how geomorphological features, altitude, and distance between entrances affect bryophyte diversity. In thirteen cave entrances within PEIB, we identified 70 species of bryophytes, including 14 new records for the area. The most common life forms were leafy forms, followed by mat, tuft, and weft forms. Our research highlighted that geographic proximity influenced phylogenetic similarity among bryophyte communities, with larger cave entrances supporting greater diversity. *Zeta* diversity analysis revealed that rare species drive turnover in communities, emphasizing the importance of caves as genetic reservoirs and potential refuges for rare species. These findings underscore the ecological significance and diversity of cave flora in the Neotropics, highlighting the need for conservation strategies that include subterranean plant communities.

Key-words: karst environments; cave flora; subterranean ecosystems; speleobotany; conservation.

INDICADORES DE IMPACTO

Os resultados da pesquisa sobre a flora cavernícola no Parque Estadual do Ibitipoca (PEIB) mostraram impactos sociais, tecnológicos, econômicos e culturais relevantes, sobretudo em potencial, dada a singularidade dos ecossistemas estudados e a escassez de estudos semelhantes em áreas tropicais. A investigação contribui para a conscientização sobre a biodiversidade subterrânea e reforça a importância da conservação dos ecossistemas cavernícolas, promovendo a educação ambiental entre as comunidades locais e os visitantes do PEIB. O trabalho está diretamente alinhado à área temática de meio ambiente da Política Nacional de Extensão, incentivando a valorização cultural e ambiental do território. Os dados gerados são essenciais para embasar políticas públicas e estratégias de manejo sustentável das cavernas, com potencial de atrair investimentos voltados ao ecoturismo responsável e à geração de renda local. Além disso, a proteção das cavernas e das comunidades vegetais associadas fortalece os serviços ecossistêmicos, beneficiando atividades como agricultura e turismo na região. Os resultados estão em consonância com diversos Objetivos de Desenvolvimento Sustentável (ODS) da Agenda 2030, incluindo o ODS 15 (Vida Terrestre), ao promover a conservação da biodiversidade, e o ODS 13 (Ação contra a Mudança Global do Clima), ao reforçar a importância das cavernas como um refúgio para a biodiversidade frente às mudanças climáticas. Por fim, os impactos da pesquisa, destacam a relevância da flora cavernícola como um campo de estudo ainda pouco explorado e com grandes possibilidades de investigação futura, contribuindo para a ampliação do conhecimento científico e para o desenvolvimento de ações sustentáveis na região.

IMPACT INDICATORS

The results of the research on cave flora in the Ibitipoca State Park (PEIB) revealed significant social, technological, economic, and cultural impacts, particularly in potential terms, given the uniqueness of the ecosystems studied and the scarcity of similar studies in tropical areas. The investigation contributes to raising awareness about subterranean biodiversity and emphasizes the importance of conserving cave ecosystems, promoting environmental education among local communities and visitors to PEIB. The work is directly aligned with the *environment* thematic area of the National Extension Policy, fostering cultural and environmental appreciation of the territory. The data generated is essential to supporting public policies and sustainable management strategies for caves, with the potential to attract investments aimed at responsible ecotourism and local income generation. Additionally, the protection of caves and their associated plant communities strengthens ecosystem services, benefiting activities such as agriculture and tourism in the region. The results are consistent with several Sustainable Development Goals (SDGs) of the 2030 Agenda, including SDG 15 (*Life on Land*), by promoting biodiversity conservation, and SDG 13 (*Climate Action*), by highlighting the importance of caves as refuges for biodiversity in the face of climate change. Finally, the research impacts underline the relevance of cave flora as a field of study that is still underexplored and holds great potential for future investigations, contributing to the expansion of scientific knowledge and the development of sustainable actions in the region.

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PRIMEIRA PARTE

INTRODUÇÃO GERAL

Gruta do Funil - Foto: Gabriel Lourenço - SEE

GERAL INTRODUCTION

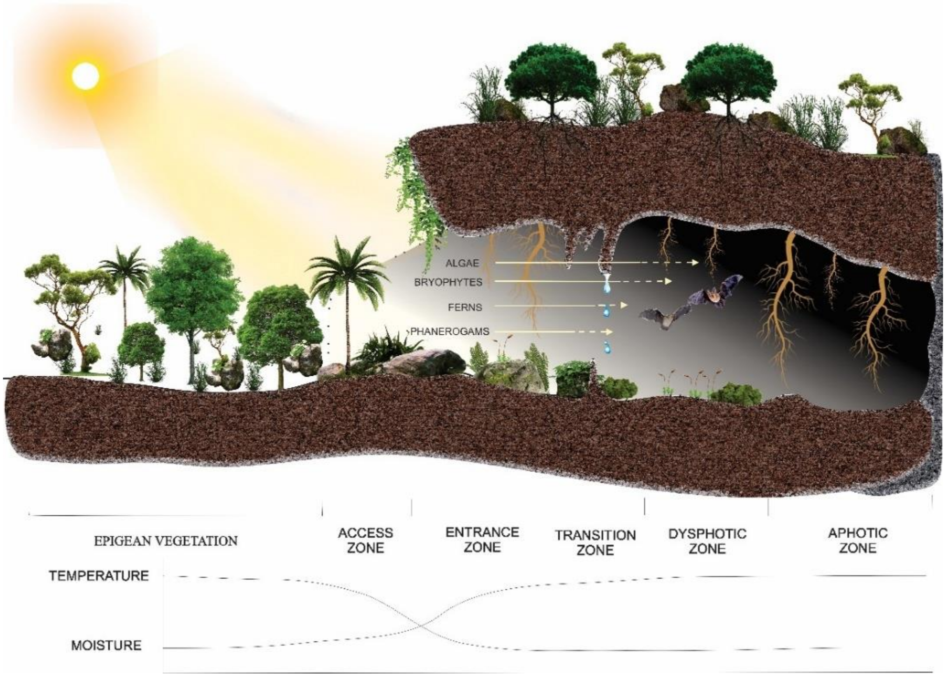
Caves are fascinating and scientifically unexplored environments, primarily due to the inaccessibility and the inherent risks associated with these environments. A diverse of physical and environmental characteristics characterize caves as unique and complex environments, serving as a powerful environmental filter that selects species capable of adapting to often extreme conditions (ROMERO, 2004; CULVER; PIPAN, 2019). Among these ecological features, low or complete absence of luminosity represents the primary selection factor for organisms inhabiting hypogean environments, limiting primary production of photosynthetic organisms in these habitats. Since the inception of speleobiology focusing on subterranean biology, fauna has been the most studied and consequently the most known group (CULVER; PIPAN, 2019). However, despite being less studied, various other organisms can adapt and colonize caves, such as microorganisms, fungi, and plants (PENTECOST; ZHAOHUI, 2001; ROMERO, 2004; FALASCO *et al.*, 2014; ENGEL, 2015; POPOVIĆ *et al.*, 2015; MONRO *et al.*, 2018; CULVER; PIPAN, 2019).

To date, studies on cave flora have primarily focused on communities associated with artificial lighting (FALASCO *et al.*, 2014). In these caves, usually tourist-oriented, areas directly affected by the light radiation emitted by lamps are colonized by a flora predominantly composed of algae, diatoms, and cyanobacteria, and and less frequently bryophytes and ferns. These communities are known as *lampenflora* and are intensively studied due to their environmental and visual impact in the subterranean environment (Figure 1). In many cases, the management and control of these communities are necessary to minimize their effects on the cave ecosystems (MULEC, 2010; FALASCO *et al.*, 2014; ESTÉVEZ *et al.*, 2019).

Although *lampenflora* has been the focus of cave flora studies so far, recent research has shown that the cave environment can harbor a very peculiar natural flora (PENTECOST; ZHAOHUI, 2001; BUZJAK *et al.*, 2010; ABDULLIN, 2011; MONRO *et al.*, 2018; PUGLISI *et al.*, 2018, 2019; REN *et al.*, 2021). Due mainly to the low incidence of light, restricted to entrances and skylights, high humidity, and lower temperatures compared to epigean environments, the species of photosynthetic organisms that grow in caves are generally hygrophytic and sciophytic species found in moist and low-light environments (DOBAT, 1998). The need for light tends to increase with the evolutionary position of plant groups, which is reflected in the composition of the flora found in the luminous gradient of cave entrances. In this sense, a gradual change in vegetation can be observed, with phanerogams, ferns, and bryophytes usually present near the entrance, while in intermediate areas, mainly ferns and bryophytes are found, and in the deepest areas, the vegetation is almost entirely composed of algae, cyanobacteria, and diatoms (DOBAT, 1998).

Based on these observations, DOBAT (1998) generically classified cave entrances into four zones according to the pattern of decreasing luminosity inside the cave, accompanied by vegetation zoning. The first region (access zone) is the one closest to the external environment, the most well-lit, and still with some soil presence. The second (entrance zone) has a decreasing layer of soil, receiving indirect sunlight and generally colonized by more hygrophilic vegetation, including ferns, bryophytes, and phanerogams, the latter already starting to exhibit etiolated growth. The third (transition zone), much less illuminated, receives weak and indirect light, and although it may still have the presence of phanerogams and ferns, bryophytes tend to be the most representative. The presence of green patches of algae and cyanobacteria is also remarkable in areas where plants have not colonized. The fourth and last (deep twilight zone) presents only surface communities of algae and/or cyanobacteria (Fig. 1).

Figure 1 - Schematic profile of the photic zones and their respective microclimatic characteristics within a cave, starting from the entrance region, representing the levels of colonization by different phototrophic organisms.



Source: Modified from DOBAT (1998).

The availability of light, water, and nutrients are directly related to the distribution of plant communities in natural environments (CARSON; SCHNITZER, 2008). Cave entrances and skylights have unique physical and climatic characteristics, such as significantly lower daily temperature and humidity range compared to epigean environments (FREITAS *et al.*, 1982). Milder temperatures and higher humidity can exert strong pressure on the species that can grow in such environments, potentially hosting plant communities that are often unique to these environments. Recent studies in China have shown that cave entrances may harbor a high diversity of vascular plant species, many of which are threatened with extinction (MONRO *et al.*, 2018). Additionally, new species endemic to these environments have been discovered, suggesting that cave entrances can serve as important refuges for plants that grow in regions where the landscape environment has more adverse conditions or has been affected by anthropogenic impacts (ZHANG; HE, 2009; HE; ZHANG, 2011; WEI *et al.*, 2011; ZHOU *et al.*, 2014; MONRO *et al.*, 2018; RODDA; MONRO, 2018; WEN, 2019; FU *et al.*, 2017, 2022).

Soil nutrient availability is crucial in determining plant communities. Species capable of tolerating different concentrations of essential elements, for example, can grow in a wider range of environments, due mainly to their plasticity in functional traits (GARNIER *et al.*, 2016; GIANASI *et al.*, 2021). On the other hand, the ability to absorb nutrients directly from rocky substrates can be an important trait for a particular species to being able to colonize cave environments (ALVES; KOLBEC, 1993).

Regarding avascular flora, knowledge about bryophytes in caves is still limited, but research has revealed that these non-vascular plants demonstrate remarkable adaptation to subterranean environments, especially in deeper regions of caves (GARNIER *et al.*, 2016). Studies in different regions, such as Sicily and China, have shown a considerable diversity of bryophytes in caves, including species with troglophilic characteristics and possibly threatened species (PUGLISI *et al.*, 2018, 2019, 2024; REN *et al.*, 2021; CONG *et al.*, 2023). Due to their pioneering characteristics and their pre-adaptation to cave-like environments, bryophytes are an essential component in projects aimed at conserving and restoring these environments, promoting and facilitating the establishment of other plant and animal communities (REN *et al.*, 2021). Moreover, they can be used as indicators of diversity and conservation of the cave environments. Studies have shown their bioindicator potential for metal pollution, such as mercury and zinc (LIU *et al.*, 2018). Thus, their adaptation to low-light environments, ability to grow in nutrient-poor substrates, and resistance to heavy metals highlight bryophytes as an important element in the ecological functioning of caves and indicators of habitat quality.

In this regard, the avascular and vascular communities of cave flora is not only important for the understanding of the diversity and adaptation of plants to extreme environments but also for conserving these ecosystems. Plant communities growing at cave entrances can play a fundamental role in maintaining subterranean biodiversity directly affecting the availability and input of resources to the hypogean environment (SOUZA-SILVA *et al*., 2011; GEEKIYANAGE et al., 2019). Recent studies show that these communities at cave entrances can favor the colonization and maintenance of invertebrates and cave bats, in addition to playing an important role in regulating the essential microclimatic conditions for the maintenance of these communities (RICHTER *et al*., 1993; GILLIESON; THURGATE, 1999; PROUS *et al*., 2005; DOMÍNGUEZ-VILLAR *et al*., 2013; RABELO *et al*., 2021; CARDOSO *et al*., 2022).

Although caves generally have peculiar physical-climatic characteristics, these conditions can be significantly different from one cave entrance to another due to their intrinsic geomorphologic formation, geographic position or landscape composition (FORD; WILLIAMS, 2007). Entrances can vary in dimensions, slope, position, and geographic orientation, as well as biogeographical aspects, including geological, pedological, and phytogeographical components, promoting distinct features to cave entrance zones and altering, for example, the quantity and depth of solar radiation reaching these environments (GUNN, 2004). The presence of dense vegetation adjacent to some cave entrance, can reduce thermal range and increase relative air humidity, while the absence of vegetation will have the opposite effect (DOMÍNGUEZ-VILLAR *et al*., 2013).

Cave entrances are often affected by human activity (CARDOSO *et al*., 2022; GAMS; GABROVEC, 1999). In Brazil and around the world, regions with cave occurrences are generally susceptible to economic activities and may undergo rapid transformations in their landscape due to deforestation for pasture cultivation or be destroyed by urbanization and mining activities, which in many cases are irreversible, which makes understanding and more detailed studies related to cave flora essential (AULER, 2016; JAFFÉ *et al*., 2018).

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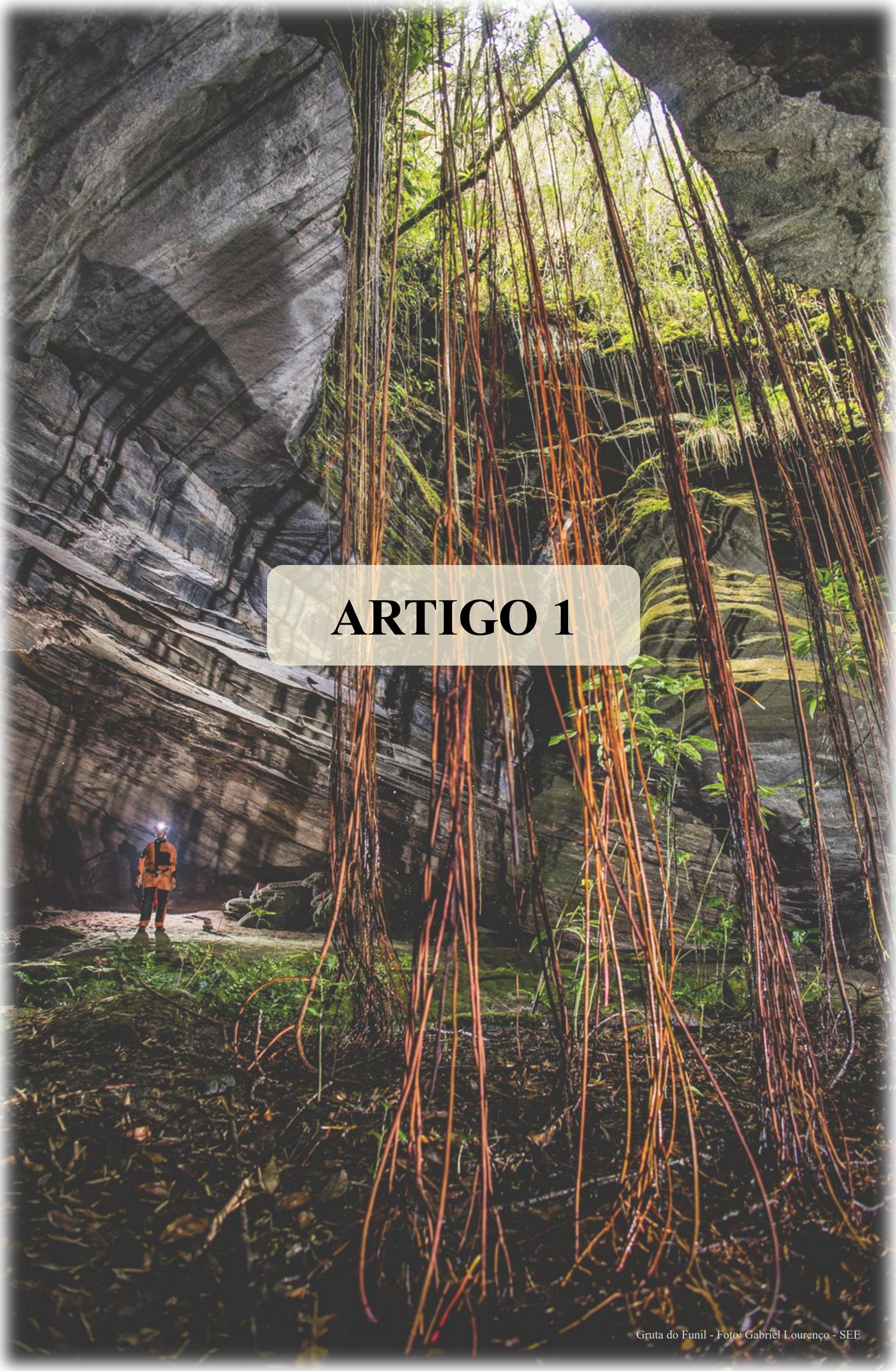
SEGUNDA PARTE - ARTIGOS

ARTIGO 1



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ARTIGO 1 - When Light Finds Darkness: Light Zonation and Soil Features Determining Flora Communities in Neotropical Non-Carbonate Caves

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ABSTRACT

Cave ecosystems, with their specific environmental conditions, harbor distinctive biodiversity characterized by unique evolutionary and adaptive traits. However, there is a notable scarcity of research on cave flora, especially in tropical ecosystems. Our study investigates the ecology of cave flora, focusing on factors influencing their distribution, richness, and composition along the ecotone light gradient of cave entrances in a neotropical Montane ecosystem. We aim to understand the ecological patterns of cave-dwelling flora, contributing to closing this knowledge gap and promoting conservation. We hypothesize that the zonation of depth and light can be delineated based on cave plant community structure and soil features. To test this, we conducted sampling within eight cave entrances in Ibitipoca State Park (PEIB), a protected area in Minas Gerais, Brazil, known for its diverse vegetation and numerous quartzite caves. We sampled vegetation up to the dark penumbra region, defining zones using a luxmeter. Soil samples were analyzed for physical and chemical properties. Our results revealed 134 plant species, with angiosperms being the most diverse group in photic zones and bryophytes in dysphotic zones. Species richness decreased with depth, and significant differences in composition were observed among caves, cave entrances, and light zones. Soil analysis showed varying proportions of granulometric and elemental composition influencing plant community structure. We identified key soil variables affecting species richness and composition. This study underscores the ecological significance and diversity of cave flora in Neotropics, emphasizing the need for conservation strategies that include subterranean plant communities.

Key-words: Cave flora, Cave ecology, Subterranean biology, Plant ecology

1. INTRODUCTION

Cave ecosystems, with their unique environmental conditions, give rise to a distinctive biodiversity characterized by unique evolutionary and adaptive traits (Culver and Pipan, 2019). However, cave life remains less understood compared to surface habitats due to habitat isolation and human accessibility challenges, though it has long captivated speleologists and researchers. While studies on cave fauna have garnered considerable attention, research on other groups, like plants, remains limited, predominantly focusing on phototrophic communities associated with the artificial illumination of tourist caves, known as *lampenflora* (Falasco et al., 2014). More intricate and comprehensive studies of natural cave flora have primarily originated from Chinese caves (Monro et al., 2018, Ren et al., 2021), leading to the discovery of new species of cave-dwelling plants (Zhang & He, 2009; He & Zhang, 2011, Zhou et al., 2014). Surprisingly, in tropical ecosystems characterized by extremely high diversity and threatened habitats, there is a notable scarcity of research on cave flora.

Cave entrance regions serve as important ecotonal zones, acting as gateways for the movement of organisms between the surface and the subterranean environment. Some of these organisms have become established and even specialized in such zones (Prous et al., 2015). This ecotonal region plays a vital role in the ecology of caves, functioning as an entry point for invertebrates, bats, and other organisms, and acting as filters (both physical and biological), which may repel, absorb, or even amplify certain species populations (Prous et al., 2015, Barros et al., 2022). The interconnectedness between the cave entrances and the hypogean environment creates a dynamic interface, influencing the supply of organic resources and shaping the ecological interactions within the subterranean ecosystem (Simon et al., 2007, Souza Silva et al., 2007).

Cave environments are characterized by unique conditions such as limited light (which becomes absent in areas far from entrances), high humidity, and stable temperatures. Despite these conditions, they can be colonized by phototrophic organisms from various taxonomic groups as far as the light can reach (Mulec, 2018). In addition to light, soil, atmospheric relative humidity, and climate stability enable the colonization of these habitats by predominantly ombrophilic and shade plant species (Dobat, 1998, Cedrés-Perdomo et al., 2024). Studies in temperate and subtropical zones have observed an incredibly diverse array of plants in caves (Monro et al., 2018), with bryophytes being the most represented group (Mulec & Kubešová, 2010, Cong et al., 2017, Puglisi et al., 2019, Cedrés-Perdomo et al., 2024). Due to their low light compensation points, bryophytes can grow under low light conditions and even reach deeper cave regions (Privitera et al., 1997, Dobat, 1998, Pentecost and Zhaohui, 2015, Puglisi et al., 2018, Cedrés-Perdomo et al., 2024).

In this study, we aim to thoroughly explore the ecology of cave flora, focusing on the factors that influence their distribution, richness, and composition along the ecotone light gradient of cave entrances. While existing research has primarily focused on temperate regions, our investigation seeks to broaden the scope by examining flora in caves within a tropical, diverse ecosystem. This approach aims to provide a more comprehensive understanding of cave flora structure in these threatened habitats. Given the critical status of these ecosystems, the need for focused investigations into cave flora becomes increasingly evident. Our research seeks to address this knowledge gap by understanding the ecological patterns of cave-dwelling flora, ultimately contributing to better foundations for decision-making in cave conservation.

We aim to identify patterns that enhance our understanding of cave flora and contribute to the broader discourse on biodiversity conservation and the ecological importance of these neglected subterranean habitats. A key aspect of our study involves identifying the predictors that affect cave flora communities along the light gradient ecotone in cave entrances. We hypothesize that the zonation of depth and light can be delineated based on both the structure of cave plant communities and soil features. We believe soil, depth zones, and light zonation are important predictors that influence plant groups associated with ecotonal subterranean environments to varying degrees. We expect that plant communities will be richer near the epigean zone, where more suitable soil conditions prevail, and poorer, composed mainly of bryophytes, in deeper areas with less suitable soil conditions. Additionally, we anticipate that caves and cave entrances will serve as important elements in selecting unique plant communities.

2. METHODS

2.1. Study Site

Ibitipoca State Park (PEIB) is a conservation unit located in southeastern Minas Gerais, in the municipality of Lima Duarte, spanning an area of 1,923.5 hectares. The park's vegetation consists of a mosaic of communities, standing out as an island of rocky fields within a region of seasonal semi-deciduous forest. PEIB is notable for having a large number of significant caves in a relatively small area (Corrêa-Neto and Batista-Filho, 1997, Wray and Sauro, 2017), including the largest and third-largest quartzitic caves in the country, Martimiano II and Bromélias caves (Rubbiolli, et al., 2019, Oliveira et al., 2024). Over the past few decades, several speleology groups have intensively researched the PEIB caves, resulting in the discovery of at least 121 registered features, totaling 63 known caves with 96 horizontal entrances and 25 vertical skylights. Of these, only 10 caves are open to tourism (Salimena-Pires, 1997, CECAV/CANIE, 2024) (Fig. 1). Despite being part of the Atlantic Forest biome, the park's geological and geomorphological features support a diverse mosaic of phytophysiognomies, ranging from high-altitude fields, rocky fields, and gallery forests to cloud dwarf forests, contributing to its rich flora (Forzza et al., 2013, Oliveira-Filho et al., 2013).

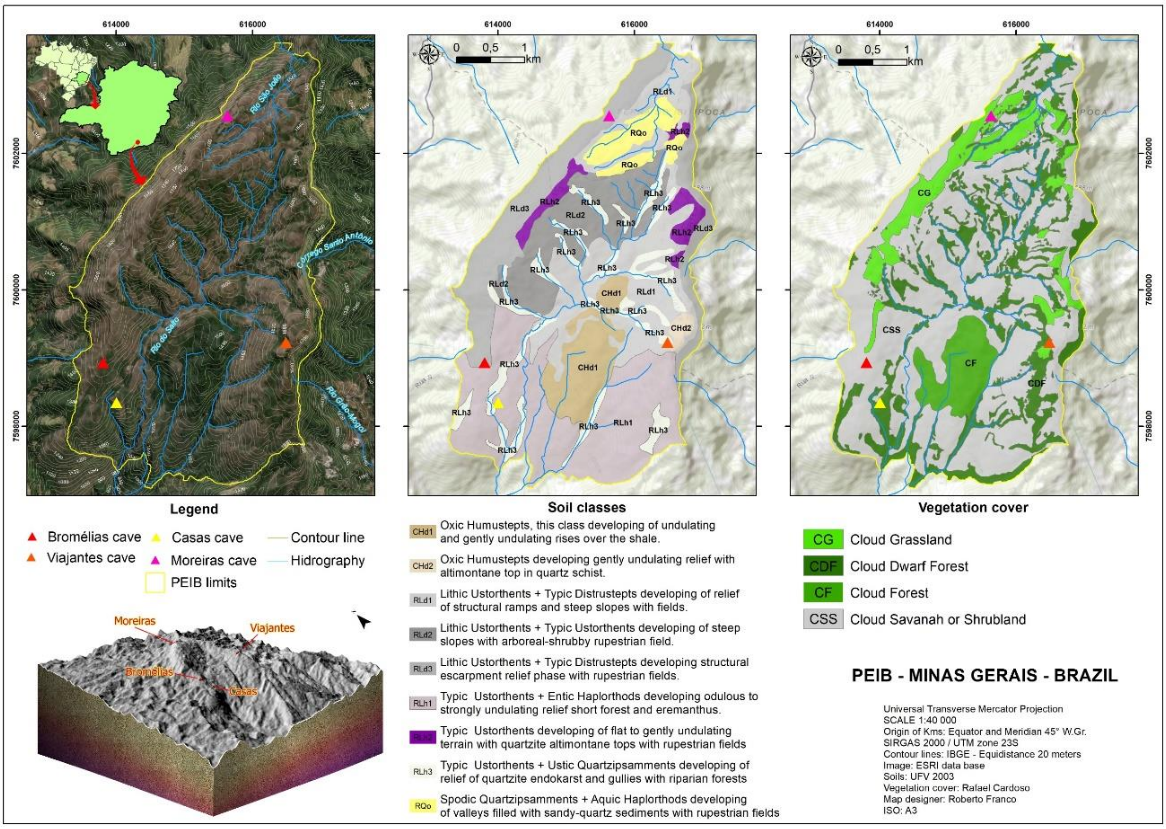

Figure 2 - Location of the study area and caves in the Ibitipoca State Park, Minas Gerais state, Brazil. The maps show the distribution of cave entrances sampled within PEIB limits in the landscape, soils classes and vegetation cover.

2.2. Field Samplings

To analyze the vegetation in the cave entrance regions, we conducted thorough sampling and identification of all individuals up to the point where light could no longer reach, known as the dark penumbra region. We used a luxmeter (Incoterm PLUX1000) to define these regions in each cave, measuring light intensity until the lux values reached zero. For this purpose, we held the luxmeter at chest height, with the sensor pointing towards the cave entrance. In this region, we established four rectangular plots, each two meters wide, equidistantly distributed to capture the luminous gradient (photic and dysphotic) and the depth zones (DZ) across each cave entrance. This approach provided data on the taxonomic composition, richness, and depth of colonization in the floor sediments of the cave entrances, as well as sampling of *topsoils* (Fig. 2).

Figure 3 – Schematic design of delimitation of cave entrances zones: depth zones 1 to 4 and light zones (photic and dysphotic) to sample cave flora and soil.

All species of cave flora, including angiosperms, ferns, and bryophytes, were collected from each plot. Specimens were prepared as herbarium vouchers for subsequent identification and deposition (Fig. 3B). Part of the material was sent to specialists in each specific group for further analysis. For *topsoil* sampling, four surface replicates (0-10 cm depth) were collected from each sampling unit and combined for laboratory analysis (Fig. 3C).

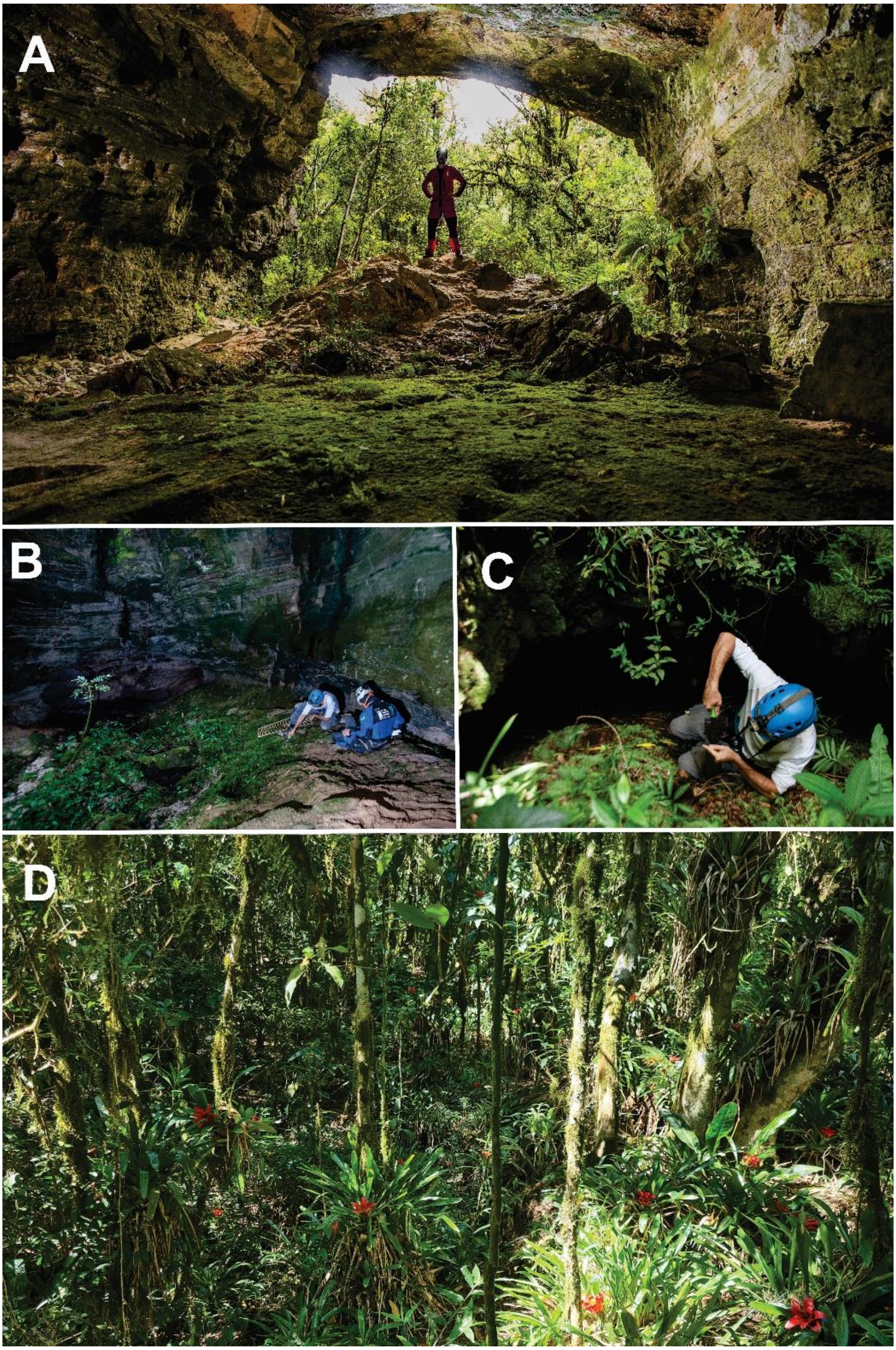

Figure 4 – Cave flora of the Ibitipoca State Park. **A -** Cave entrance view from inside; **B -** Sample of cave flora procedure inside a cave entrance; **C** – Soil sample procedure at a cave entrance; **D** – Vegetation of the surface environments.

2.3. Data analysis

2.3.1. Richness estimators

We generated size-based rarefaction and extrapolation curves for total alpha diversity for each light zone, up to double the reference sample size, for all cave entrances sampled and all groups of subterranean flora. We performed analysis using '*iNEXT.3D'* R package (Chao et al., 2021, Chao and Hu, 2023).

2.3.2. Soil analysis

Granulometric analyses were performed to assess the proportions of sand, silt, and clay. Clay content was determined using the pipette method, while sand content was measured by sieving. X-ray fluorescence analysis was conducted with a Shimadzu EDX-720 energy dispersive X-ray fluorescence spectrometer, equipped with a rhodium tube and silicon-lithium detector. Data were collected under a vacuum of 40 Pa using a 10-mm collimator. Total concentrations of carbon, nitrogen, hydrogen, and oxygen were determined using a LECO® CHNS/O elemental analyzer, TruSpec Micro model. The geochemical elemental data were also obtained using the *Shimadzu* EDX-720 energy dispersive X-ray fluorescence spectrometer.

2.3.3. Diversity Analysis

To investigate the similarities and dissimilarities in cave flora composition among the samples, we constructed Multidimensional Scaling (MDS) graphs and conducted Analysis of Similarities (ANOSIM) to test the significance of groups formed by caves, cave entrances, depth zones, and light zones. The MDS graphs provided a visual representation of the differences between groups, while ANOSIM determined if these differences were statistically significant. Analysis was performed with *vegdist* function from the "*vegan*" R package and plots were constructed using the "*ggplot2*" R package.

2.3.4. Abiotic Factors Selection

To evaluate the significance of the physical, chemical, and elemental composition of soil and sediment, we conducted a principal component analysis (PCA) using 13 variables: Si, Al, Fe, K, Ti, S, P, Ca, Zr, H, C, N, O, clay, and sand. PCA provides an intuitive interpretation from a pedological perspective, with each component representing key factors and processes characterizing the soil and sediments at cave entrances (Horák-Terra et al., 2014). We used PCA components instead of raw data because the soil parameters were not orthogonal, invalidating their use in classical statistical analysis (Manly, 2005). The data were scaled to standardize the disparate values before analysis (Reimann et al., 2008). PCA was conducted using the *'prcomp'* function with scaled variables from the "factorextra" R package, and the significance of PCA axes and variables was assessed using the "PCAtest" R package. This package evaluates the overall significance of a PCA, each PC axis, and the contributions of each observed variable to the significant axes based on permutation-based statistical tests.

2.3.5. Abiotic Drivers of Plant Communities on Cave Entrances

To assess the effects of environmental factors (soil physicochemical features) and depth zones on species richness in the cave entrances, we constructed Generalized Linear Mixed Models (GLMMs) using significant PCA axes and depth zones as predictors, with cave entrances as a random factor. We applied Poisson distributions for the response variable in the GLMMs. Using the *'dredge'* function from the 'MUMIN' package, we tested all possible combinations of the variables included in the full model, including the null model. The final model was constructed using the mean contributions of all models with delta < 2. To test the effects of depth on cave soil physicochemical features (PCA axes), we performed ANOVA followed by pairwise Tukey tests.

To test the effects of abiotic factors (soil chemistry) on the composition of the plant community along the ecotonal gradient of cave entrances, we performed a permutational multivariate analysis of variance (PERMANOVA) using the significant PCA groups. The statistical significance of this analysis was tested using 9999 permutations of the residuals in a reduced model and Type III (partial) sums of squares (Anderson, 2001). We used the *"AICcPermanova"* R package, which generates all possible linear models for a given set of predictor variables, using the distance matrix as a response variable. This function allows the user to specify the maximum number of variables in a model, useful when there are many predictors. For model selection, we used the *fit_models* function, which fits PERMANOVA models for all combinations of variables, arranges the models by Akaike Information Criterion (*AICc*) score, and calculates the maximum variance inflation factor (*max_vif*) for each model. Final models were constructed using the mean contributions of all significant models with deltaAICc < 2.

3. RESULTS

3.1. Cave flora diversity and distribution

A total of 134 plant species were observed in the sampled cave entrances, including 81 angiosperm species from at least 30 families, 13 fern species from at least 7 families, and 39 avascular species. The avascular species included 19 liverwort species from 9 families and 20 moss species from 13 families (Appendix). Angiosperms were the richest group in depth zones DZ1, DZ2, and DZ3, while avascular flora were richer in DZ4 (Fig. 4a). Total species richness significantly decreased along the depth zone gradient, with the model explaining about 60% of the total variance (GLMM; $z = 12.4$, $p < 0.001$, $R^2 = 0.6$) (Fig. 4b).

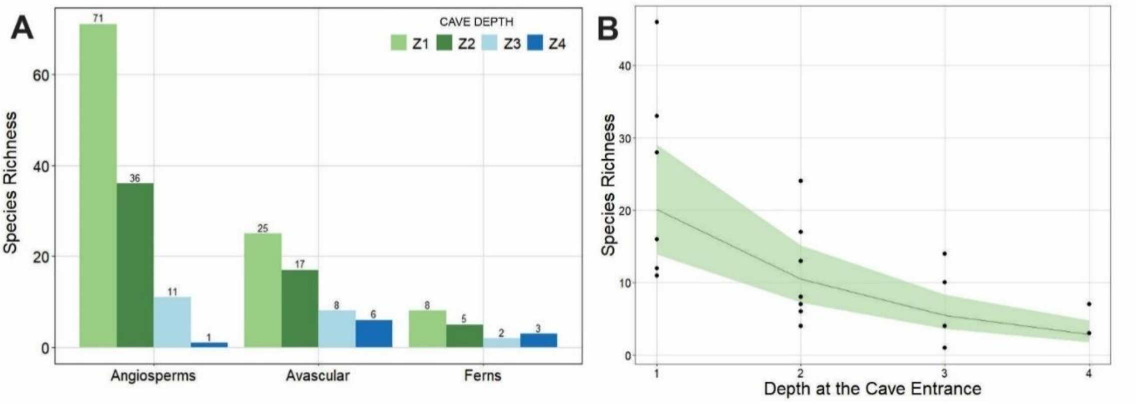

Figure 5- A- Total species richness of each plant group in the four depth zones delimited on cave entrances of PEIB; **B-** GLMM model of the effects of depth zones delimited in cave entrances of PEIB on total species richness of cave flora.

In DZ1, 105 species were recorded, with the families Myrtaceae (9 spp.), Melastomataceae (7 spp.), and Solanaceae (5 spp.) being the richest among the angiosperms, Aspleniaceae (2 spp.) as the richest fern family, and Dicranaceae and Plagiochilaceae as the richest avascular families. In DZ2, 58 species were recorded, with the families Myrtaceae (4 spp.), Melastomataceae (4 spp.), and Araceae (3 spp.) being the richest among the angiosperms, Dryopteridaceae (3 spp.) as the richest fern family, and Dicranaceae and Plagiochilaceae as the richest avascular families. In DZ3, 30 species were recorded, with Solanaceae (2 spp.) as the richest angiosperm family, Dryopteridaceae (1 spp.) and Aspleniaceae (1 spp.) as the only fern families, and Plagiochilaceae (3 spp.), Fissidentaceae (3 spp.), and Pallaviciniaceae (3 spp.) as the richest avascular families. In DZ4, 10 species were recorded, with Piperaceae (1 spp.) as the only angiosperm family, Dryopteridaceae (1 spp.), Aspleniaceae (1 spp.), and Blechnaceae (1 spp.) as the only fern families, and Pallaviciniaceae (2 spp.) as the richest avascular family.

Leucobryum crispum (Bryophyta:Leucobyaceae), *Capsicum pereirae* (Tracheophyta: Solanaceae) and *Cupania vernalis* (Tracheophyta: Sapindaceae) were the most common species in DZ1, *Leucobryum crispum* (Bryophyta: Leucobyaceae) and *Arachniodes denticulata*

(Tracheophyta: Dryopteridaceae) were the most common species in DZ2. In DZ3 and DZ4 no species had more than one occurrence.

Size-based rarefaction and extrapolation curves for the total cave flora community did not show a tendency to reach an asymptote. However, the extrapolated richness suggested a trend towards stabilization. The expected richness for the total community exceeded 150 species, with approximately 100 species for angiosperms, 13 species for ferns, and 44 species for bryophytes (Fig. 5 A, C, E, G).

For the total alpha diversity curves (Fig. 5B), DZ1 was the richest zone. However, the observed richness was far below the extrapolated richness for twice the sample size. In DZ2 and DZ3, the extrapolated richness showed a small overlap of the 0.95 confidence intervals, indicating no significant difference in alpha diversity between these two regions if the sampling were doubled. DZ4 presented the lowest richness, but its curve's asymptote was close to stabilizing.

For angiosperms (Fig. 5D), alpha diversity was significantly different across all light zones for both observed and extrapolated richness, with DZ3 and DZ4 showing curves tending toward stabilization. For ferns (Fig. 5F), both observed and extrapolated richness in DZ2 and DZ3 did not show significant differences, and the extrapolated richness of DZ4 did not differ significantly from that of DZ3. DZ3 had the lowest alpha diversity, with its extrapolated richness close to the observed richness.

For the non-vascular component of cave flora (Fig. 5H), the first three depth zones did not show differences in their observed and extrapolated alpha diversity, with the actual diversity not yet reached by the extrapolated value. DZ4 was the least rich, with its curve tending to stabilize.

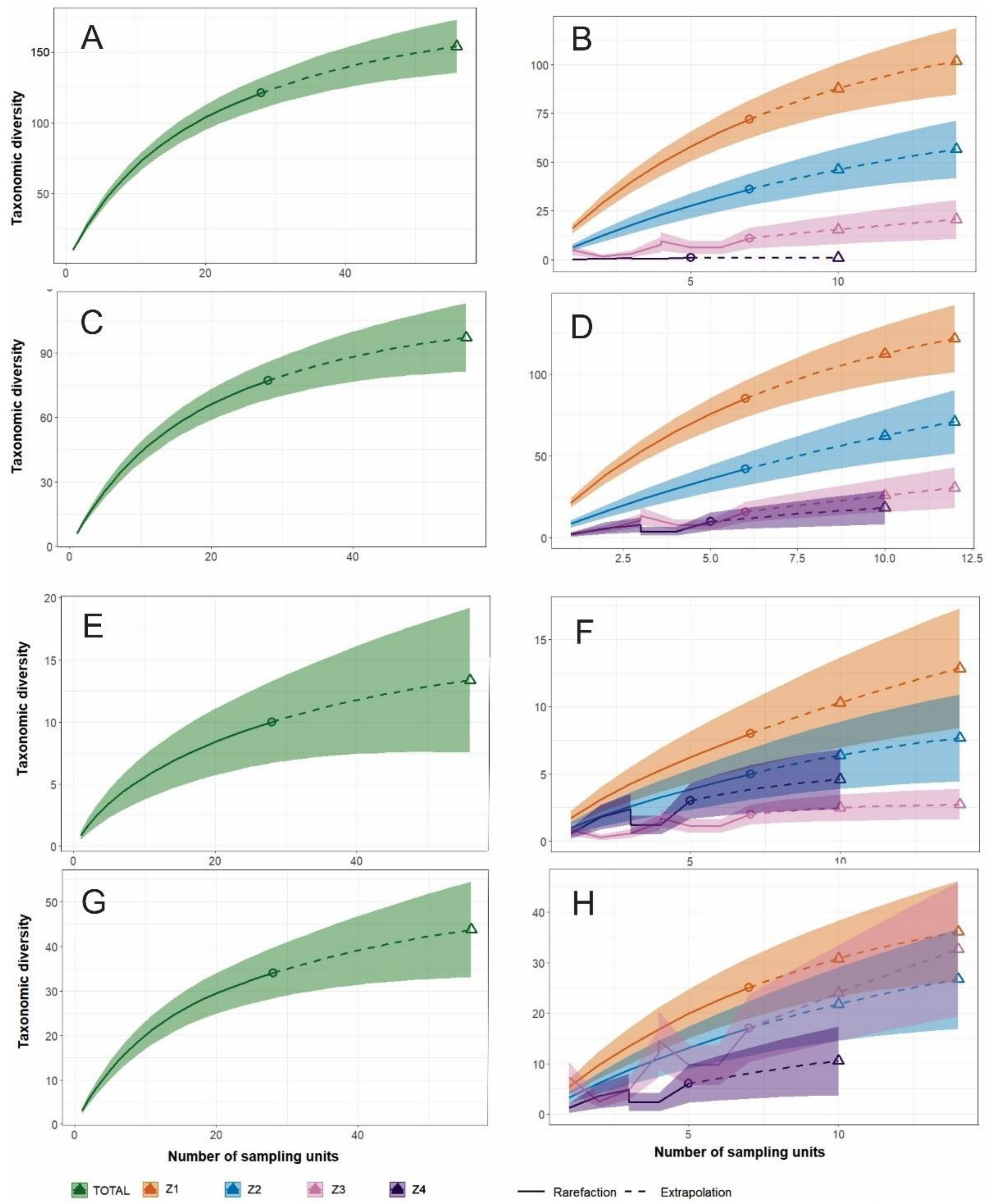

Figure 6 - Sample-size-based rarefaction (solid curves) and extrapolation (dashed curves) with gamma diversity and alpha diversity. A – Total community; B – Total communities in each depth zone; C – Total angiosperms community; D - Total angiosperms community in each depth zone; E – Total ferns community; F - Total ferns community in each depth zone; G – Total bryophytes community; H - Total bryophytes community in each depth zone.

Examining the MDS plots, we observed a clear differentiation in the plant species composition among the four caves sampled and between photic and dysphotic zones (Fig. 6a; Fig. 6b). ANOSIM analyses indicated significant differences in cave flora composition among caves (R= 0.6433; *p*= 0.001), cave entrances (0.6508; *p*= 0.001) and light zones (0.1378; *p*=

0.005). However, no significant differences were found between depth zones (R=0.3065; *p*= 0.087) (Table 3).

The species *Plagiochila exigua*, *Symphyogina leptothelia, Justicia sebastianopolitana*, *Geonoma schottiana*, *Leucobryum crispum*, *Frulania beyrichiana*, *Myrciaria tenella* and *Miconia doriana* were significative to MDS1 axis. *P. exigua* and S. *leptothelia* were more related to the dysphotic zones while *J. sebastianopolitana*, *G. schottiana*, *L. crispum*, *F. beyrichiana*, *M. tenella* and *M. doriana* were more related to the photic zones.

When analyzing plant groups separately, we observed clear differentiation in species composition among the four caves and the eight entrances sampled for both angiosperms and bryophytes (Fig. 6C; Fig. 6D). ANOSIM analyses revealed significant differences in angiosperm composition among caves (R= 0.5125; p= 0.001) and cave entrances (R= 0.7305; p= 0.001), but not between depth zones (R= 0.0215; p= 0.395) and light zones (R= 0.682; p= 0.282). Similarly, for bryophytes, significant differences were found among caves (R= 0.6433; p= 0.001) and cave entrances (R= 0.6508; p= 0.001), but not between depth zones (R= 0.1378; p= 0.005) and light zones (R= 0.3065; p= 0.087) (Table 1). For ferns, the number of observed species was insufficient to perform the nMDS analysis.

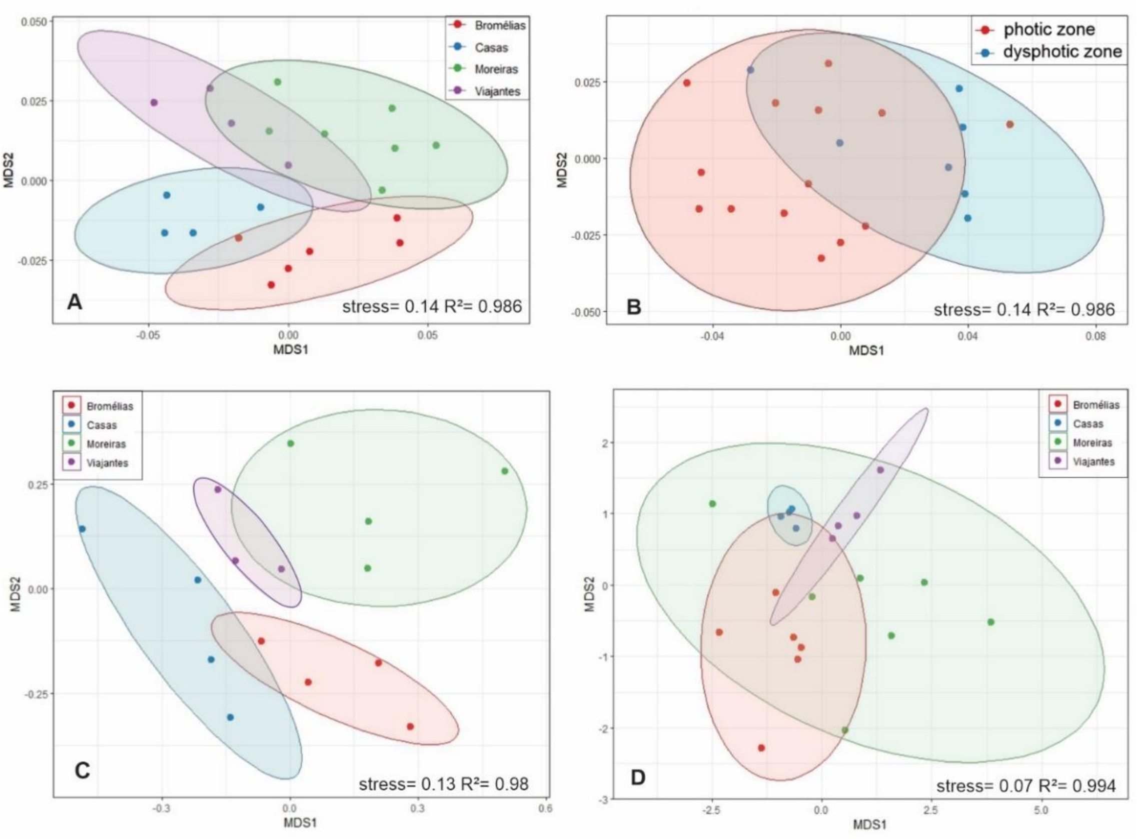

Figure 7 - MDS plots showing the dissimilarities of cave flora composition among caves and light zonation. A - Comparisons of the total community within different caves; B - Comparisons of the total community among photic and dysphotic zones; C - Comparison of the angiosperm's community within different caves; D - Comparison of the bryophyte's community within different caves.

Table 1- ANOSIM analysis of the dissimilarities on the cave flora communities of PEIB under different approaches for total community, angiosperm's community and bryophyte's community.

	Groups	ANOSIM statistic R	Significance *p*-value
Total Community	Among Caves	0.6433	0.001*
	Among Cave Entrances	0.6508	0.001*
	Among Depth Zones	0.3065	0.087
	Among Light Zones	0.1378	0.005*
Angiosperms	Among Caves	0.5125	0.001*
	Among Cave Entrances	0.7325	0.001*
	Among Depth Zones	0.0215	0.352
	Among Light Zones	0.0682	0.264
Bryoph	Among Caves	0.7366	0.001*

Groups	ANOSIM statistic R	Significance *p*-value
Among Cave Entrances	0.6991	0.001*
Among Depth Zones	-0.0828	0.834
Among Light Zones	0.0072	0.413

3.2. Organic and inorganic elemental composition

The sediments at cave entrances predominantly consist of sand (99.7% ± 9%) with minimal clay content (0.28% ± 0.1%). While the proportion of sand increases in deeper zones, the clay content tends to be higher in more external zones (Appendix).

The elemental composition of the cave entrances is dominated by Si (mean 51.7% ± 9.1%), Al (~24.3% ± 7.3%), C (~7.3% ± 5.8%), O (~4.5% ± 1%), K (~4.1% ± 1.4%), Fe (~3.9% ± 1.6%), H (~1.2% ± 0.4%), Ti (~0.8% ± 0.33%), S (~0.54% ± 0.45%), N (~0.53% ± 0.32%), P (~0.45% ± 1.26%), Ca (~18% ± 0.38%), and Zr (~0.05% ± 0.03%). The C/N ratios ranged from 1.28 to 25.2, with a mean of 12 ± 4.5 (Appendix). Although several elements did not show a distinct trend across depth zones, most plant nutrients (C, H, N, Fe, S, Ca) exhibited a noticeable decrease further from the cave entrance.

The first 3 PCA axes are significant and account for 71.5% of the total variation. Principal Component Analysis axis 1 (PC1) explains up to 42% of the data variability with C, N, H, S, Al, clay, sand and C:N ratio significant correlated with PC1 (Fig. 7A). PCA axis 2 (PC2) accounted for 16.5 % of the variation with Si, Fe, Al, K and S significant correlated with PC2 (Fig. 7A). PCA axis 3 is also significant and accounts for 13.5% of the total variation with Zr, Ti, O, clay and sand significant correlated with PC3. In general, just PC1 values tend to change among depth zones, with higher to lower values from the more external to the deeper zones (Fig. 7B, 7C, 7D).

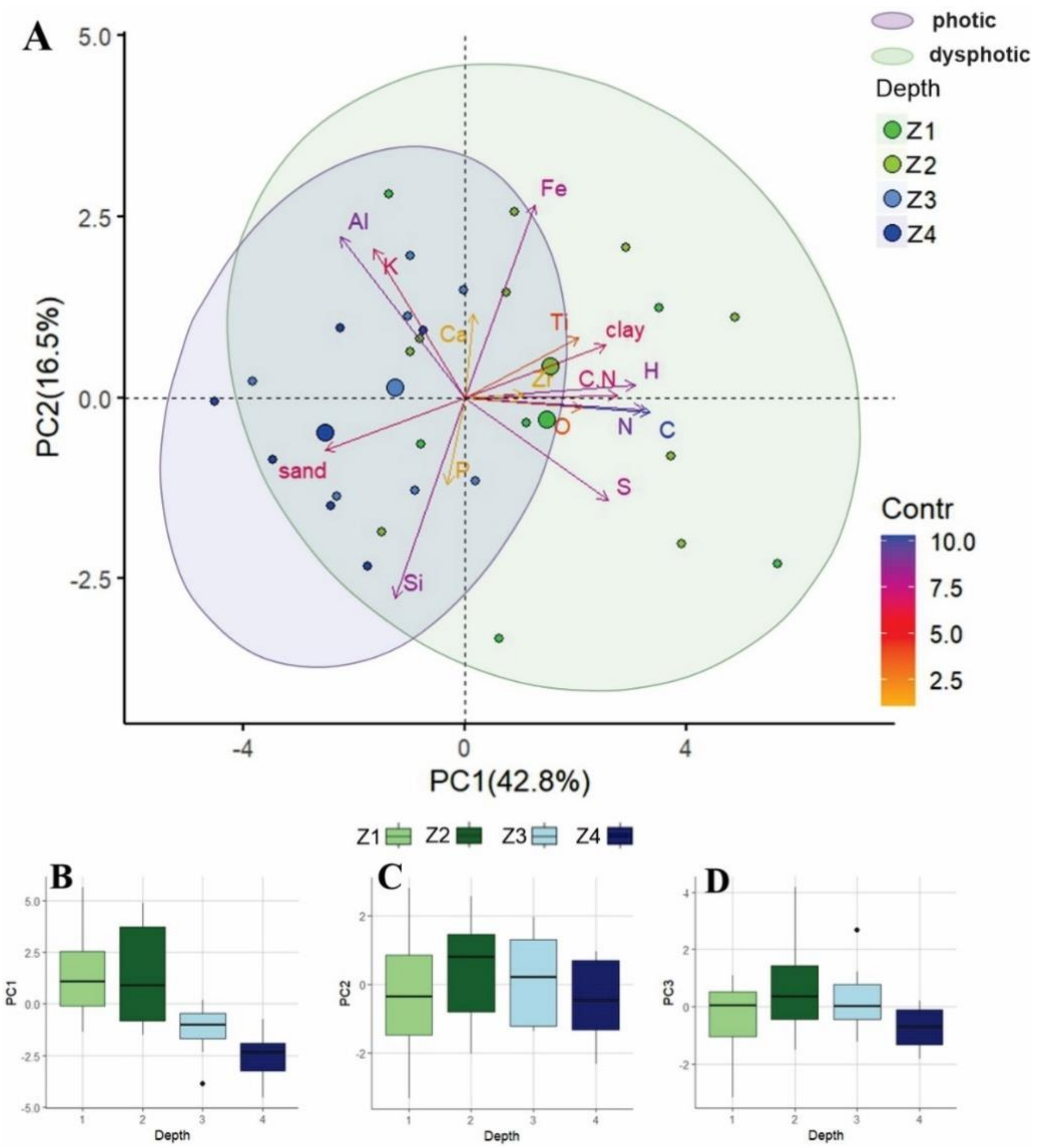

Figure 8 - Principal component analysis (PCA) of the soil variables sampled in cave entrances environments of the PEIB. **A** – PCA plot of two dimensions. the strength of the contribution of environmental variables is indicated by the color gradient of the arrows, as shown to right of the PCA under heading 'Contrib'. Points are colored according to each depth zones sampled and elipses evidence the light zonation groups (photic and dysphotic). **B, C and D** and shows mean variation of significant PCA axes values (PC1, PC2 and PC3 respectively) (y axes) among the four depth zones (x axes).

The GLMM model, which incorporated total cave flora species richness and the first three significant PCA axes (PC1, PC2, and PC3), explained approximately 45% of the total variance. This model revealed significant and positive effects of PC1 (characterized by higher values of C, N, H, S, C:N, and clay, and lower values of sand and Al) on species richness.

Conversely, PC2 had significant and negative effects on richness (characterized by higher values of Al, Fe, and K, and lower values of Si and S). PC3 did not show significant effects on cave flora species richness (GLMER; $z=7.85$, $p < 0.001$, $R^2=0.48$) (Fig. 8A). For angiosperms, the best model accounted for 38% of the variance and showed significant and positive effects of PC1 and significant and negative effects of PC2, with no effect from PC3 (Fig. 8B). For ferns and bryophytes, the best models indicated no significant effects of any PCA axis on species richness (Fig. 8C, 8D).

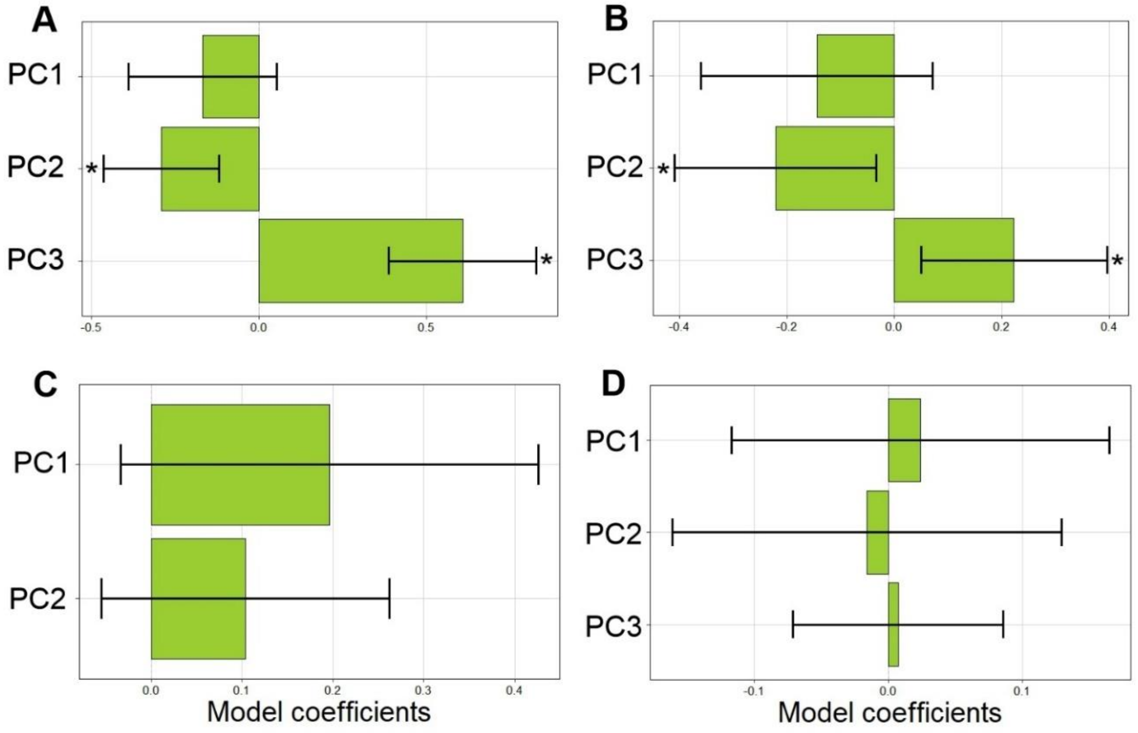

Figure 9 – Graphs of best GLMM models for the effects of soil features (PCA axes) on cave flora species richness of PEIB (* shows significant effect of the variable in the model). **A-** Effects of PCA axes on total richness; **B -** Effects of PCA axes on angiosperms richness; **C -** Effects of PCA axes on ferns richness; D - Effects of PCA axes on bryophytes richness.

We then analyzed the effects of PCA axes on cave flora community with multivariate analysis of variance (PERMANOVA) and selected the mean contribution of each variable in all models with Delta AICc ≤ 2. PC1 and PC3 had significant effects on the total community composition, with PC1 accounting for 7.8% of the variability in the community composition, PC3 explaining 7.3% of the variability. PC2, which accounted for 5.1% of the variability, did not significantly affect the community composition. A substantial proportion of the variability (78.3%) in the total community composition was not explained by our final model (Fig 9A). For angiosperms the best model shows the effects of just PC3 on community composition explaining 14% of the total variation (Fig 9B). For ferns the models were not significative and none of the PCA axes predict the specie composition (Fig 9C). For bryophytes the three axes significative predicted species composition and explain about 32% of the total variation (Fig 9D).

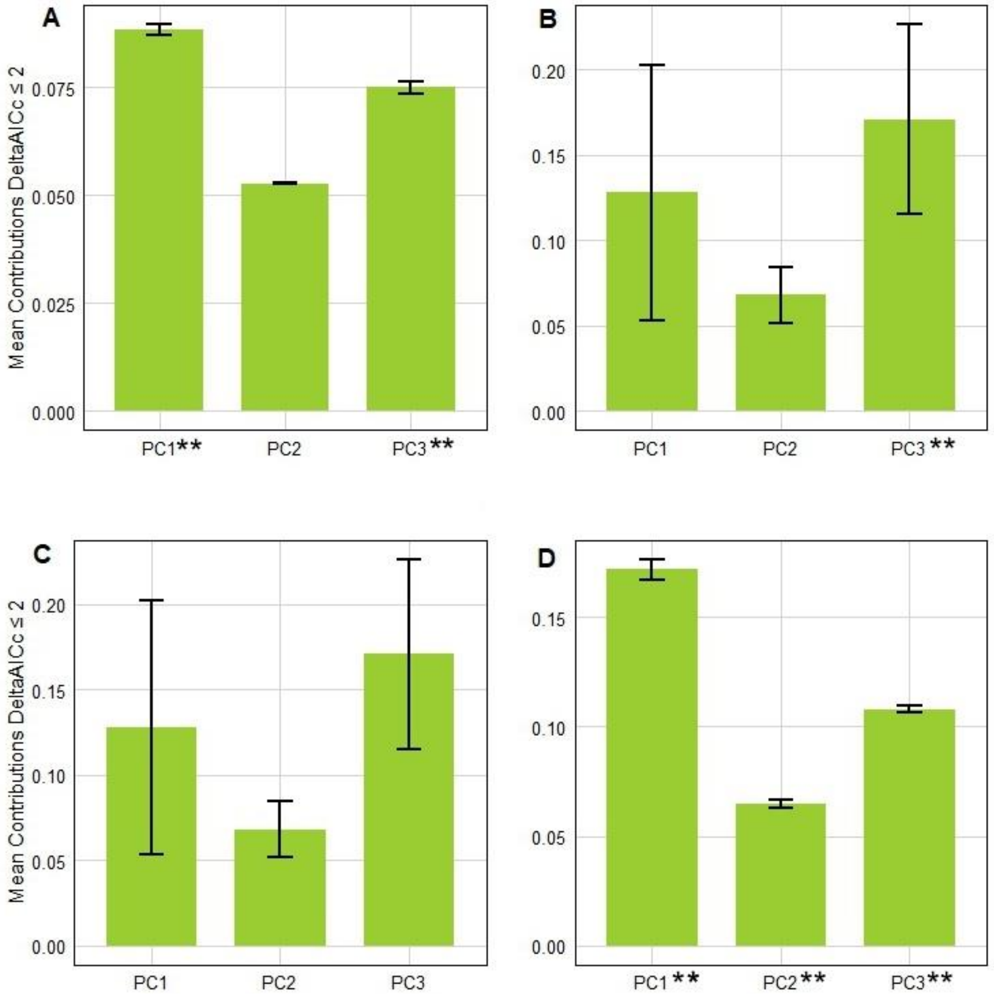

Figure 10 - Graphs of best PERMANOVA models for the effects of soil features (PCA axes) on cave flora species community composition of PEIB (** shows significant effect of the variable in the model). **A-** Effects of PCA axes on total community composition; **B -** Effects of PCA axes on angiosperms community composition; **C -** Effects of PCA axes on ferns community composition; D - Effects of PCA axes on bryophytes community composition.

4. DISCUSSION

Despite the importance of cave vegetation and soils for understanding cave ecology, especially in ecotonal zones, our knowledge of these components remains limited, posing a fundamental question in subterranean biology (Monro et al., 2018, Mammola et al., 2019, 2020). Our study represents the first comprehensive investigation into the ecology of cave flora in Neotropics and in non-carbonate caves. We not only identified a diverse range of species associated with these habitats but also gained insights into the distribution patterns of plant communities and the physicochemistry of the substrates on which they grow. Additionally, we identified depth zones that influence community structure within the ecotonal gradient of cave entrances.

The plant diversity observed in the sampled caves was substantial, with 134 species from 50 families. Extrapolation curves suggest that, with double the sampling effort, species richness could exceed 160. For all individual groups, the expected richness was also higher than the observed numbers. For comparison, Monro et al. (2018) documented over 418 vascular plant species from 83 families in 61 caves in China, with extrapolation curves predicting up to 846 species regionally. In the PEIB cave entrances, the average species richness was up to 16 species per entrance. Similarly, Monro et al. (2018) recorded fewer than 15 species per cave, though their study did not include avascular plants. At the entrances of the caves studied here, the most common angiosperm species were forest understory and rock-dwelling species previously identified in botanical surveys of the PEIB (Moreira et al., 2018). Flowering plants were the most diverse group in the epigean environments of cave entrances, while bryophytes were more prevalent in the deeper penumbral zones.

Some studies indicate that angiosperms constitute the largest proportion of cave flora (Pentecost and Zhaohui, 2001, Monro et al., 2018). Due to their adaptation to low light conditions in the understory of external vegetation, these species are well-suited for colonizing cave entrances, representing 93% of the species observed in some areas (Monro et al., 2018). However, other research highlights bryophytes as a significant and sometimes dominant component of cave flora (Mulec, Kubešová, 2010, Cong et al., 2017, 2024, Puglisi et al., 2018, 2024, Ren et al., 2021; Cedrés-Perdomo et al., 2024). Bryophytes, as primitive pioneering plants, thrive in harsh environments and are crucial for soil formation, soil and water conservation, plant succession, and environmental enhancement in karst caves (Kurschner and Frey, 2013, Cao et al., 2020). They also serve as indicators of habitat quality and the health of vascular plant communities in cave entrance habitats (Liu et al., 2018, Ren et al., 2021).

It is noteworthy that there are at least 120 known cave entrances in Ibitipoca State Park, and our study identified 39 bryophyte species across just eight entrances. This accounts for 14% of the total known bryophyte species in PEIB (284 spp.) (Yano et al., 2014), with our extrapolation suggesting that up to 44 species could be discovered with double the sampling effort. Cave entrances may thus play a crucial role in conserving bryophyte flora (Cong et al., 2024, Cedrés-Perdomo et al., 2024, Puglisi et al., 2024). For comparison, caves in the Balearic Islands and Azores are predicted to harbor approximately 25% and 35% of the bryophyte species known from these islands, respectively (Rosselló and Pericàs, 2011, Gabriel et al., 2004). This underscores the potential of using the diversity and rarity of bryophyte species in cave entrances as indicators of their conservation value (Puglisi et al., 2024).

Cave entrance zones exhibit a gradual decrease in light intensity from the epigean to the hypogean regions. This reduction in light leads to decreased plant diversity in deeper zones, where bryophytes become dominant in the penumbral environments, as anticipated. Specifically, the bryophytes *P. exigua* and *S. leptothelia* were significantly more common in dysphotic zones. These liverworts are typically found in the understory of ombrophile forests, thriving in humid, shaded substrates (REFLORA, 2024). Several studies have highlighted that changes in plant group diversity within karst caves are closely related to light intensity (Monro et al., 2018, Ren et al., 2021, Cong et al., 2024, Cedrés-Perdomo et al., 2024). However, other abiotic factors, such as microclimatic conditions and soil features, also play crucial roles (Monro et al., 2018, Ren et al., 2021).

Species associated with more illuminated zones, such as *J. sebastianopolitana*, *G. schottiana*, *L. crispum*, *F. beyrichiana*, *M. tenella*, and *M. doriana*, are predominantly angiosperms from the montane forest communities of PEIB (Oliveira-Filho et al., 2013, Carvalho et al., 2000, Moreira et al., 2018, Moreira et al., 2021). Notably, *L. crispum* and *F. beyrichiana* are moss and liverwort, respectively, often growing as epiphytes on the stems and branches of vascular plants (Yano and Luizi-Ponzo, 2014). Different species exhibit convergent adaptations that enable them to thrive in cave entrance environments at varying levels, which may include adaptations in life-forms, phylogenetic relationships, and leaf nutrient concentration ratios (Kurschner and Frey, 2013, Puglisi et al., 2018, Bai et al., 2020). Further exploration of these physiological traits could improve our understanding of how plants withstand the challenges of subterranean environments and high levels of shading.

Although our rarefaction curves clearly show differences in total richness and angiosperm richness between depth zones, these distinctions were less pronounced for fern and bryophyte communities. Discrepancies in dissimilarity analyses reveal that, overall, cave units, cave entrances, and light zones significantly accounted for the separation of the total flora community. However, when analyzed individually, only cave units and entrances were significant for specific groups. This variation may be linked to environmental adaptations, such as soil factors (Bai et al., 2020), where vascular plants respond more distinctly, whereas most avascular plants are rock-dwelling or, to a lesser extent, epiphytic.

Some studies suggest that cave entrances serve as climatic refuges and biodiversity reservoirs, preserving species with disjunct distributions due to their habitat preferences and the unique conditions of cave environments (Monro et al., 2018, Cedrés-Perdomo et al., 2024, Cong et al., 2024, Puglisi et al., 2024). Caves offer protection against external climatic fluctuations and habitat alterations (Herrero-Borgoñón, 2002, Monro et al., 2018, Cong et al., 2024). Ecological disturbances affecting surface plant communities may not impact subterranean habitats, where plants find physical refuge and suitable microclimates. This protective effect makes caves vital reservoirs of plant diversity, preserving species that may no longer thrive in the surrounding landscape (Herrero-Borgoñón, 2003, Monro et al., 2018).

Certain plant species are adapted to thrive in very low light levels, enabling them to grow deep within caves under specific conditions. For instance, mosses have been documented in the extremely low-light dysphotic zones of caves (Puglisi et al., 2024, Cong et al., 2024, Cedrés-Perdomo et al., 2024). Research into the minimum light requirements for plant growth in subterranean environments has shown that, in some Spanish caves, ferns, liverworts, and mosses can survive with light levels as low as 10 lux for some moss species (Ginés and Ginés, 1992, Fiol, 1995). These low-light conditions act as stringent ecological filters, often limiting the survival and reproduction of many organisms (Monro et al., 2018). This is primarily because photosynthesis is heavily affected by shade, favoring plants that can adjust leaf nutrient concentrations, such as N, P, Mg, and Ca, to optimize photosynthetic activity under varying light conditions (Hawkesford et al., 2012). Consequently, species with specialized ecological niches can be found in subterranean habitats (Puglisi et al., 2024, Cong et al., 2024, Cedrés-Perdomo et al., 2024), often including rare species with restricted distributions (Zhang et al., 2009, Zhou et al., 2014, Deng et al., 2024).

Soil and sediments from the cave entrances in PEIB were predominantly composed of sand and organic matter (Appendix). A similar granulometric composition was observed by Brandão et al. (2022) in sediment deposits from a quartzitic cave located 75 km north of PEIB, where sand made up 91.7 to 99.1% of the sediment. The sediment there had comparable levels of Si, Al, and K, reflecting the mineralogy dominated by quartz and muscovite. In contrast, the PEIB cave lithology is characterized by quartzites containing approximately 95% quartz and 5% muscovite plus chlorite (Pinto, 1991). The sediment composition in caves may differ from the surrounding rocks and soils due to their role as sediment traps (Ford and Williams, 2007), and the sediment geochemistry can indicate varying contributions from epigeous material and the surrounding rock (Brandão et al., 2022).

The entrances of PEIB are predominantly surrounded by cloud-dwarf forests, which develop soils capable of supporting larger vegetation in high-altitude rupestrian complexes. These forests are characterized by dystrophic and acidic soils with high aluminum (Al^{3+}) saturation and significant organic carbon content (Dias et al., 2003). Such conditions prevent the decomposition of organic matter, resulting in high organic carbon levels and reduced microbial activity due to aluminum toxicity and low temperatures (Benites et al., 1999). The reduced content of organic elements in deeper cave zones is likely related to the decreasing input of organic matter as one move away from the cave entrance (Simon et al., 2007). Consequently, sediments in the deeper cave zones are generally poorer in nutrients, particularly organic ones, compared to those near the entrances.

Soil composition was significantly related to plant community structure, particularly for angiosperms, accounting for 45% of the total variance in species richness and 20.2% of the variance in plant community composition. However, the distinct differences in soil composition between depth zones, especially in PC1 scores (Figure 7), with a notable decrease in carbon and associated elements (N, H, S) in deeper zones, make it unclear how much these factors overlap. While soil conditions significantly influenced angiosperm community richness and structure in our study, Ren et al. (2021) found that bryophyte composition in Chinese caves is also affected by soil properties. Although bryophytes are primarily rock-dwelling and epiphytic, and a direct response to edaphic factors might not be expected, they have demonstrated potential as bioindicators of soil pollution from metals like mercury and zinc, and thus as indicators of habitat quality (Liu et al., 2018). The observed decrease in carbon content, from 10% in DZ1 to less than 3% in DZ4, may reflect the ability of cave-dwelling flora to alter their environment, rather than being solely an edaphic control on community structure. In these sandy soils and sediments, cation exchange capacity and nutrient availability are primarily due to organic matter, which is lower in deeper regions, thus creating selective filters for the survival of plant species that must also contend with the other challenging conditions typical of subterranean environments (Dias et al., 2003).

5. CONCLUSIONS

Our study highlights the ecological significance and diversity of cave flora in Neotropics and non-carbonate caves. It revealed a rich variety of species, including flowering plants, ferns, and bryophytes, and showcased distinct plant community patterns and substrate characteristics within cave entrance ecotones.

Although clear differentiation was observed in total richness and angiosperm richness across depth zones, patterns for ferns and bryophytes were less pronounced. This variability may be due to specific adaptations to the subterranean environment, including soil factors and light intensity gradients. Our similarity analysis showed significant separation based on cave units, entrances, and light zones, with certain plant species in low-light, deep cave zones demonstrating remarkable adaptations to extreme conditions.

In conclusion, the diversity and distribution of cave flora in the PEIB underscore the importance of incorporating subterranean plant communities into conservation strategies for neotropical caves. Future research should encompass broader areas with varying lithologies to deepen our understanding of the physiological adaptations of these plants and explore the roles of soil properties, leaf nutrients, geomorphological features, and microclimatic conditions in shaping cave flora.

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APPENDIX I

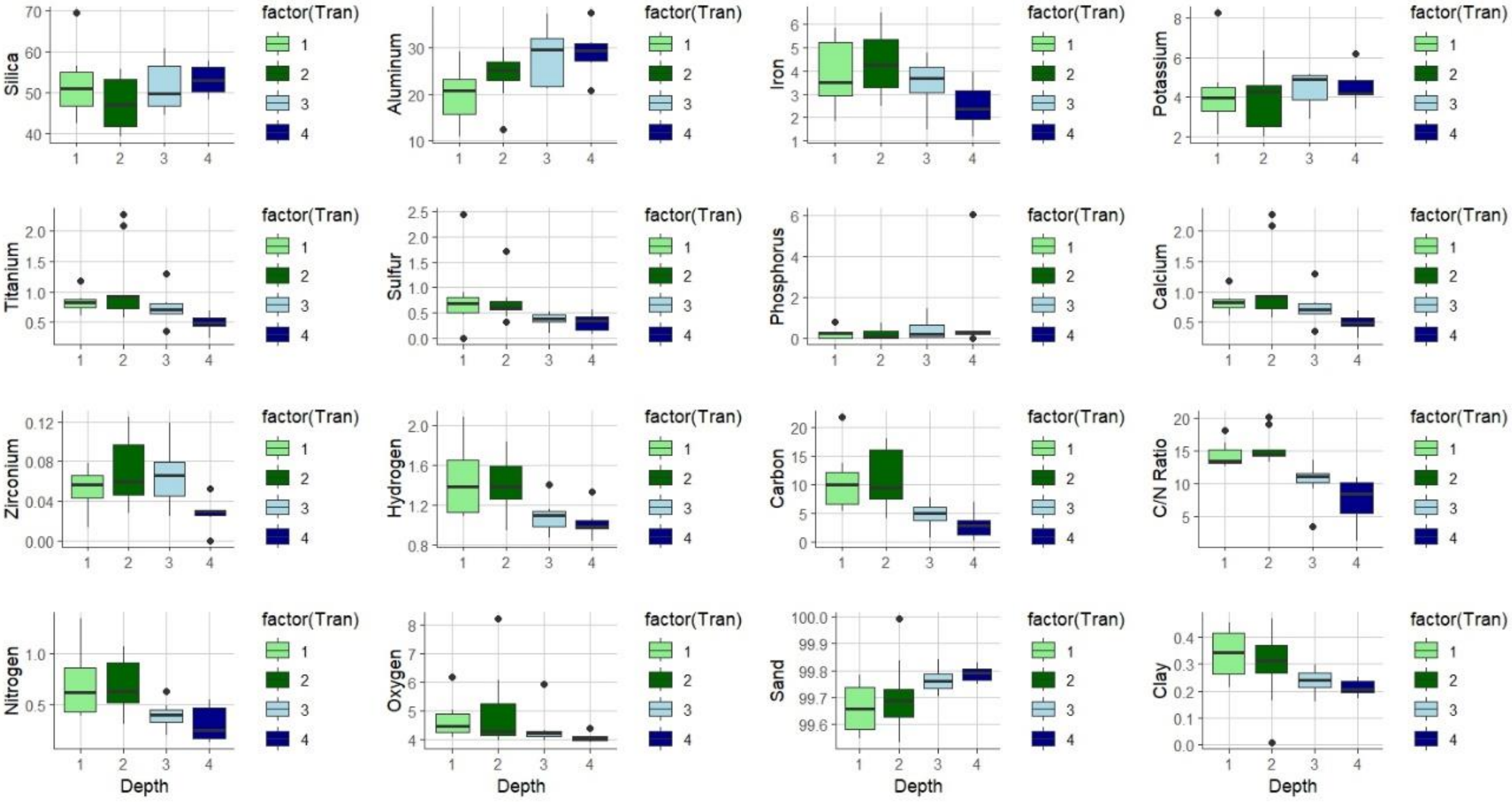

Figure 1 - Elementary contents in cave entrances transects sampled at Ibitipoca State Park.

APPENDIX II

Table 1- Species of cave flora sampled at entrances of caves at Ibitipoca State Park.

Family	Species
Angiosperms	
Acanthaceae	*Justicia sebastianopolitanae* Profice
	Mendoncia sp.
Annonaceae	*Annona emarginata* (Schltdl.) H.Rainer
Araceae	*Anthurium* sp.
	Asterostigma luschnathianum Schott
	Philodendron propinquum Schott
Arecaceae	*Geonoma schottiana* Mart.
Asteraceae	*Mikania lindbergii* Baker
	Mikania ternata (Vell.) B.L.Rob.
Begoniaceae	*Begonia angularis* Raddi
	Begonia angulata Vell.
	Begonia apparicioi Brade
Bignoniaceae	Bignoniaceae
	Handroanthus bureavii (Sandwith) S.Grose
Bromeliaceae	*Bilbergia* sp.
	Tillandsia sp.
Commelinaceae	*Commelina obliqua* Vahl
	Commelina rufipes Seub.
Curcubitaceae	Curcubitaceae
Cyperaceae	*Carex seticulmis* Boeckeler
	Scleria leptostachya Kunth
	Scleria sp.
Eriocaulaceae	*Paepalanthus viridulus* Ruhland
Erythroxylaceae	*Erythroxylon* sp.
Euphorbiaceae	*Alchornea triplinervia* (Spreng.) Mull.Arg.
Fabaceae	*Dalbergia villosa* (Benth.) Benth.
	Senna macranthera (DC. ex Collad.) H.S.Irwin & Barneby
	Senna sp.
Gesneriaceae	*Sinningia tuberosa* (Mart.) H.E.Moore

Family	**Species**
NI	NI 1
	NI 2
	NI 3
	NI 4
Lauraceae	*Ocotea corymbosa* (Meisn.) Mez
Melastomataceae	*Bertolonia* sp.
	Leandra glazioviana Cogn.
	Leandra riedeliana (O.Berg ex Triana) Cogn.
	Miconia cf. doriana Cogn.
	Miconia paniculata (DC.) Naudin
	Miconia mellina DC.
	Miconia sellowiana Naudin
	Miconia willdenowii Naudin
	Miconia dolichostachya (Naudin) R.Goldenb.
	Pleroma sp.
Meliaceae	*Cabralea canjerana* (Vell.) Mart.
Monimiaceae	*Mollinedia elegans* Tul.
Myrtaceae	*Blepharocalyx salicifolius* (Kunth) O.Berg
	Eugenia longipedunculata Nied.
	Marlierea sp.
	Myrcia lenheirensis Kiaersk.
	Myrcia loranthifolia (DC.) G.P.Burton & E.Lucas
	Myrcia splendens (Sw.) DC.
	Myrciaria tenella (DC.) O.Berg
	Mytaceae sp1
	Psidium sp1
Ochnaceae	*Ouratea semisserrata* (Mart. & Nees) Engl.
	Sauvagesia cf. vellozii (Vell. ex A.St.-Hil.) Sastre
Piperaceae	*Peperomia cf pseudoestrellensis* C.DC.
	Piper cubataonum C.DC.
	Piper richardiifolium Kunth

Family	Species
	Piper solmsianum C.DC.
Poaceae	*Aulonemia radiata* (Rupr.) McClure & L.B.Sm.
	Pseudechinolaena polystachya (Kunth) Stapf
Primulaceae	*Myrsine* sp.
	Myrsine umbellata Mart.
Rubiaceae	*Coccocypselum erythrocephalum* Cham. & Schltdl.
	Coccocypselum lanceolatum (Ruiz & Pav.) Pers.
	Palicourea tetraphylla Cham. & Schltdl.
	Psychotria cf. leiocarpa Cham. & Schltdl.
	Psychotria forsteronioides Müll.Arg.
	Psychotria stachyoides (Benth)
	Psychotria suterella Müll.Arg.
	Rubiaceae 1
	Rubiaceae 2
Sapindaceae	*Cupania vernalis* Cambess.
	Paulinia sp.
Smilacaceae	*Smilax* sp.
Solanaceae	*Capsicum pereirae* Barboza & Bianch.
	Solanaceae 1
	Solanaceae 2
	Solanaceae 3
	Solanum sp.
	Solanum swartzianum Roem. & Schult.
Symplocaceae	*Symplocos celastrinea* Mart.
Marchantiophyta	
Aneuraceae	*Riccardia digitiloba* (Spruce ex Steph.) Pagán
	Riccardia hymenophytoides (Spruce) Meenks
Cephaloziaceae	*Cephalozia crossii* Spruce
	Odontoschisma brasiliense Steph.
Frullaniaceae	*Frullania beyrichiana* (Lehm. & Lindenb.) Lehm. & Lindenb.
Lejeuneaceae	*Lejeunea flava* (Sw.) Nees

Family	**Species**
	Lejeunea laetevirens Nees & Mont.
Lepidoziaceae	*Bazzania hookeri* (Lindenb.) Trevis.
	Lepidozia cupressina (Sw.) Lindenb.
Lophocoleaceae	*Heteroscyphus marginatus* (Steph.) Fulford
Pallaviciniaceae	*Symphyogyna brasiliensis* (Nees) Nees & Mont.
	Symphyogyna leptothelia Taylor
	Symphyogyna podophylla (Thunb.) Mont. & Nees
Plagiochilaceae	*Plagiochila cf. aerea* Taylor
	Plagiochila exigua (Taylor) Taylor
	Plagiochila gymnocalycina (Lehm. & Lindenb.) Lindenb.
	Plagiochila rutilans Lindenb.
	Plagiochila simplex (Sw.) Lindenb.
Radulaceae	*Cladoradula boryana* (F. Weber) M.A.M. Renner, Gradst., Ilk.-Borg. & F.R. Oliveira-da-Silva
	Radula pallens (Sw.) Nees & Mont.
Bryophytes	
Bartramiaceae	*Philonotis cernua* (Wilson) Griffin & W.R.Buck
Brachytheciaceae	*Rhynchostegium serrulatum* (Hedw.) A.Jaeger
Calymperaceae	*Syrrhopodon prolifer* Schwägr.
Catagoniaceae	*Catagonium brevicaudatum* (Müll. Hal.) ex Broth.
	Catagonium nitens (Brid.) Cardot
Fissidentaceae	*Fissidens elegans* Brid.
	Fissidens scariosus Mitt.
	Fissidens submarginatus Bruch
Leucobryaceae	*Campylopus arctocarpus* (Hornsch.) Mitt.
	Campylopus filifolius (Hornsch.) Mitt.
	Campylopus lamellinervis (Müll. Hal.) Mitt.
	Campylopus savannarum (Müll. Hal.) Mitt.
	Leucobryum albicans (Schwägr.) Lindb.
	Leucobryum crispum Müll. Hal.
Leucomiaceae	*Rhynchostegiopsis brasiliensis* Broth.

Family	**Species**
Phyllogoniaceae	*Phyllogonium viride* Brid.
Pilotrichaceae	*Hypnella pilifera* (Hook. & Wilson) A.Jaeger
	Trachyxiphium guadalupense (Brid.) W.R.Buck
Polytrichaceae	*Pogonatum* sp
Pylaisiadelphaceae	*Isopterygium tenerum* (Sw.) Mitt.
Rhizogoniaceae	*Pyrrhobryum spiniforme* (Hedw.) Mitt.
Sematophyllaceae	*Jirivanaea cuspidifera* (Mitt.) U.B. Deshmukh & Rathor
	Sematophyllum beyrichii (Hornsch.) Broth.
	Sematophyllum subsimplex (Hedw.) Mitt.
Sphagnaceae	*Sphagnum perichaetiale* Hampe
	Sphagnum subsecundum Nees
Ferns	
Aspleniaceae	*Asplenium pseudonitidum* Raddi
	Asplenium radicans L.
	Asplenium sp.
Blechnaceae	*Blechnum polypodioides* Raddi
Cyatheaceae	*Cyathea* sp.
Dryopteridaceae	*Arachniodes denticulata* (Sw.) Ching
	Elaphoglossum beaurepairei (Fée) Brade
	Elaphoglossum vagans (Mett. ex Kuhn) Hieron.
Hymenophyllaceae	*Hymenophyllum hirsutum* (L.) Sw.
	Hymenophyllum sturmii Bosch
Indeterminate	NI
	Fern sp1
	Fern sp2
	Fern sp3
	Fern sp4
Pteridaceae	*Pteris splendens* Kaulf.

ARTIGO 2

Gruta do Alonso - Foto: Gabriel Lourenço - SEE

ARTIGO 2 - How deep can we go? Uncovering environmental drivers of Bryophyte communities in Neotropical cave entrances

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ABSTRACT

Caves can host a unique and specialized biodiversity, being a fundamental part of the ecosystems where they are found. However, the study of cave-dwelling plant communities, particularly in tropical regions, remains underexplored. While most research has focused on cave fauna, plant communities have received less attention despite their distinctiveness and ecological importance. Light, humidity, and temperature are important factors influencing plant growth in caves, where conditions differ significantly from those in surface habitats. Among the various plant groups associated with cave entrances, bryophytes often dominate. Bryophytes are highly adaptable to low-light and high-humidity conditions, making them essential components of cave-entrances communities. They play key roles in functions such as water regulation, influence on pedogenesis and as well as promoting the establishment of other plant groups. In this study, we aimed to deepen our understanding of the bryoflora associated with Brazilian caves, hypothesizing that cave geomorphological features, altitude, and the distance between entrances influence bryophyte diversity. We expected to find species with specific functional traits within cave entrances, potentially leading to new records of bryophyte species for the studied area. Our research was conducted in thirteen cave entrances within Ibitipoca State Park (PEIB) in southeastern Minas Gerais, Brazil, a region renowned for its extensive quartzite cave systems and rich bryophyte diversity, including over 300 species. We analyzed the influence of environmental features on bryophyte taxonomic distance and zeta diversity among cave entrance communities using segmented piecewise regressions and the MS-GDM approach. Our study identified 70 bryophyte species, including 37 mosses, 32 liverworts, and one hornwort, with 14 new records for PEIB. The most common life-forms were leafy forms, followed by mat, tuft, and weft forms. The preferred substrates for cave bryophytes were rupicolous (rock), terricolous (soil), and corticicolous (bark). Our research revealed that geographic proximity influenced the phylogenetic similarity among bryophyte communities, with larger cave entrances supporting greater diversity. Zeta diversity analysis showed that rare species drive species turnover, and that entrance dimensions, slope, and colonization depth significantly predict bryophyte diversity. These findings highlight the importance of caves as genetic reservoirs and potential refuges for rare species, underscoring the need for targeted conservation efforts.

Keywords: Cave flora, subterranean ecology, neotropical bryophytes, speleobotany

1. INTRODUCTION

Caves, as unique geomorphological formations, are far more than just subterranean voids, they are integral components of larger ecosystems with inestimable value. Despite their significance, studies and knowledge about the biology of subterranean environments remain limited compared to the extensive research focused on terrestrial surface habitats (Mammola et al., 2020). Most scientific studies on subterranean environments have traditionally focused on the specialized fauna that inhabit these ecosystems (Romero & Fenolio, 2009; Mammola, 2019). However, the flora that thrives in these dimly lit environments, where conditions differ markedly from other terrestrial habitats, is equally remarkable (Cedrés-Perdomo et al., 2024). However, the cave flora has historically received less attention within speleological research, although interest has been increasing in recent years, particularly in temperate and subtropical regions (Monro et al., 2018; Puglisi et al., 2018; Puglisi et al., 2019, 2024; Bai et al., 2020; Ren et al., 2021; Cong et al., 2023; Cedrés-Perdomo et al., 2024).

The distribution and intensity of light at cave entrances directly influence the composition, arrangement, and distribution of the flora, making light the primary limiting factor for plant life in these environments (Dobat, 1998; Abdulin, 2011; Mazina et al., 2023). However, soil, humidity and temperature are also key environmental filters for plant growth in subterranean habitats. The balance between these factors acts as a driving force in the growth, development, and reproduction of plants (Fiol, 1995; Monro et al., 2018; Puglisi et al., 2018, 2019; Bai et al., 2020). Additionally, geomorphological features of caves, combined with their placement in diverse landscapes, create complex environments, resulting in habitats with unique characteristics from one cave entrance to another (Ford & Williams, 2007), being able to drive the communities' structure of cave plants (Monro et al., 2018; Cong et al., 2023; Puglisi et al., 2024; Cedrés-Perdomo et al., 2024). The influence of geomorphological factors on plant communities within neotropical subterranean ecosystems remains underexplored and was initially explored by Alves & Kolbek (1993) with preference habitats of penumbral plant communities in small quartzite caves and rock shelters in Brazil and more recently by Badia et al., (2021).

Despite inhabiting environments with distinct and unique environmental characteristics, in general, cave flora, in general, thrives in a critical ecotonal zone, linking a variety of external ecosystems with the oligotrophic conditions that prevail in subterranean environments. While the ecotonal role of cave entrances has been well-documented for faunal communities (Prous et al., 2015; Mammola & Isaia, 2018), this concept likely extends to plant communities as well.

These entrances often host diverse plant assemblages that include species from multiple vegetation types (Monro et al., 2018; Puglisi et al., 2019, 2024; Cong et al., 2023; Cedrés-Perdomo et al., 2024).

The relationship between subterranean ecosystems and the surrounding landscape is a crucial aspect of subterranean ecology (Oslon, 2017; Geekiyanage et al., 2019; Cardoso et al., 2022; Oliveira & Ferreira, 2024). Cave entrances, embedded within diverse landscapes, can be in distinct phytogeographic contexts, with varying altitudes and climate conditions, leading to different plant communities associated with each cave's surroundings (Monro et al., 2018; Puglisi et al., 2019; Cong et al., 2023). Additionally, local factors unique to each cave, such as entrance size, shape, orientation, slope, lithology, and soil composition, can significantly influence the ecological conditions plants encounter (eg. Alves & Kolbek, 1993; Monro et al., 2018; Badia et al., 2021; Bai et al., 2020; Ren et al., 2021). Among these factors, entrance size is particularly important, as it directly affects light penetration, air circulation, and nutrient availability, all of which have a significant impact on plant community and functional composition (Alves & Kolbek, 1993; Silva-Mota et al., 2018; Gianasi et al., 2020; Badia et al., 2021).

The ecological conditions within subterranean environments create habitats for plants that are markedly different from those on the surface, due to their unique environmental factors (Carmo et al., 2016; Badia et al., 2021). As a result, plants in these environments face distinct challenges, and those pre-adapted to such conditions, like sciophytes and ombrophiles, are more likely to thrive (Monro et al., 2018; Puglisi et al., 2018; Puglisi et al., 2019; Cong et al., 2023). Plant groups with varying light requirements tend to follow the gradient of decreasing light as one moves deeper into the cave, eventually reaching the aphotic zone (Dobat, 1998; Abdulin, 2011; Mulec, 2020). As one progresses further into the subterranean environment, flowering and seed plants (spermatophytes) are the first to disappear, followed by ferns (pteridophytes), liverworts, and mosses (bryophytes), with algae and cyanobacteria persisting the longest (Dobat, 1998; Mulec, 2020). Research in temperate and subtropical regions has shown that cave entrances can harbor a high diversity of plant communities, with bryophytes often being the most dominant group (Gabriel et al., 2004; Mulec & Kubešová, 2010; Puglisi et al., 2019; Cong et al., 2023; Cedrés-Perdomo et al., 2024).

Bryophytes are highly adaptable and thrive in a wide range of extreme environments, from humid tropical forests to semi-arid regions, with some species even inhabiting freshwater ecosystems (Vanderpoorten & Goffinet, 2009; Cong et al., 2017; Landuyt & Calster, 2022). These plants colonize various substrates, including soil (terricolous), rocks (saxicolous), decomposing logs (epixylic), living tree trunks and branches (corticicolous), leaves (epiphytic), and artificial surfaces (casmophytic) (Landuyt & Calster, 2022). Moreover, bryophytes play a critical role in ecosystem function, contributing significantly to biomass, influencing water dynamics through absorption and release, acting as bioindicators, facilitating vegetational succession, providing substrates for other plants and seeds germination, and offering habitats for faunal communities (Rodrigues et al., 2016; Liu et al., 2018; Ren et al., 2021).

In cave ecosystems, bryophytes, known for their exceptional adaptability to low-light and high-humidity environments, are typically the predominant plant group (Puglisi et al., 2018, 2019; Cong et al., 2023). Their influence extends beyond their own survival; recent studies in subtropical regions suggest that bryophyte diversity may serve as an indicator of overall plant community diversity at cave entrances (Ren et al., 2021). This indicates that bryophytes might act as ecological architects, shaping the diversity of other organisms and potentially playing a key role in cave entrance restoration.

Bryophytes are particularly sensitive to adverse climatic conditions, and many require humid environments to complete their life cycles (Landuyt & Calster, 2022). Consequently, cave environments could serve as refuges for bryophytes that may be relics of the regions where the cave is located, with several species classified as rare or threatened (Gabriel et al., 2004). Given the current climate scenario, global warming could lead to declines not only in already rare and threatened plant species but also in those that require cooler and more humid conditions (Geekiyanage et al., 2019; Mammola et al., 2019a, 2019b). This underscores the importance of caves in flora conservation, where the species diversity and rarity of bryophytes at cave entrances could also serve as indicators of conservation value (Gabriel et al., 2004).

Given the importance of understanding cave flora communities and their environmental drivers for conservation in tropical regions, this study delves into the ecology of bryophytes at cave entrances in a diverse region of Brazil. We hypothesize that variations in geomorphological features, altitude, colonization depth, and entrance distance will shape bryophyte diversity, with rare species being more affected by distance and common species responding to structural features. We expect that the distance between entrances will have more pronounced effects on rare species, likely due to their limited dispersion rates, while geomorphological features will affect common species more significantly. We also anticipate that taxonomic distinctness and species turnover among communities will be influenced by geomorphological features and depth of colonization, beyond just distance. Through this research, we aim to generate valuable insights to guide conservation efforts focused on protecting the unique and sensitive ecosystems within cave environments.

2. METHODS

2.1. Study Site

Ibitipoca State Park (PEIB) is a protected area located in the southeastern region of Minas Gerais, Brazil, within the municipality of Lima Duarte. Spanning 1,923.5 hectares (Fig. 1), PEIB is renowned for its significant quartzite caves within a relatively small area (Corrêa-Neto, 1997; Willems et al., 2008; Wray & Sauro, 2017). Notably, it includes the largest and the third-largest quartzitic caves in Brazil: Martimiano II and Bromélias caves (Rubbiolli et al., 2019; Oliveira et al., 2024).

The region has a Cwb climate classification (Köppen), characterized by a humid mesothermal climate with mild, rainy summers and cold, dry winters. Average annual precipitation is 2,200 mm, with temperatures ranging from 12°C to 15°C in the coldest season and 18°C to 22°C in the warmest. The area's rugged topography features altitudes between approximately 1,050 m and 1,784 m (Rodela & Tarifa, 2002) and is associated with acidic quartzite soils with high aluminum saturation (Dias et al., 2003). The distribution of streams, rivers, and waterfalls supports diverse habitats, including patches of cloud forest, savannah, and grassland throughout PEIB (Moreira et al., 2021).

The PEIB is known for its significant diversity of bryophytes nationwide. Over 300 bryophyte species have been recorded in Ibitipoca State Park (Amorim et al., 2011; Luizi-Ponzo et al., 2013; Yano & Luizi-Ponzo, 2014; Machado et al., 2016). Notably, the park is home to 209 species of liverworts (Marchantiophyta), which constitutes a significant proportion of the total liverwort species reported for Brazil (Machado et al., 2016).

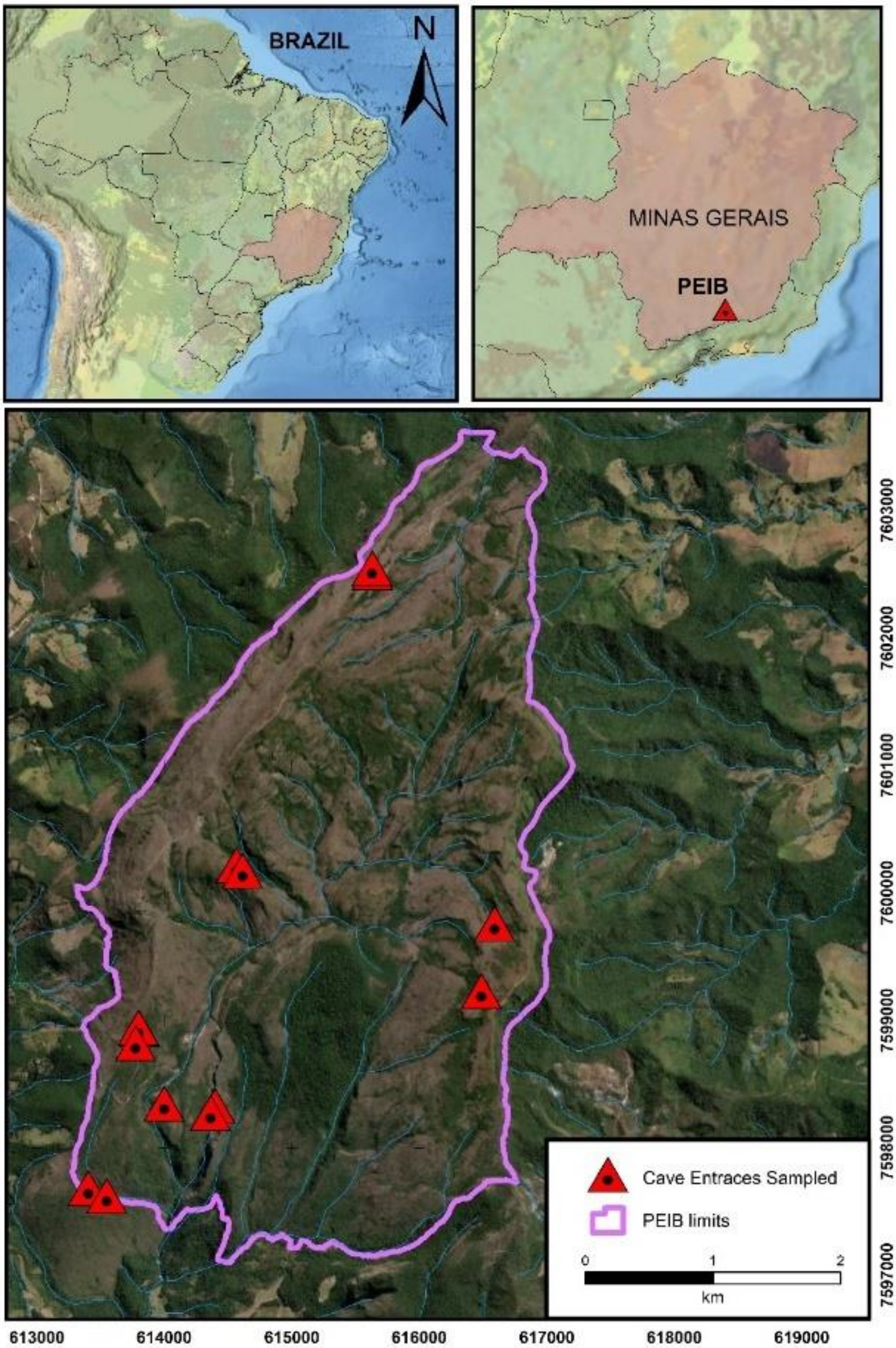

Figure 11 – Ibitipoca State Parque limits and the distribution of cave entrances sampled Ibitipoca State Parque, located in Minas Gerais state, Brazil.

2.2. Field Samplings

Thirteen cave entrances within Ibitipoca State Park were selected for this study (Fig. 1). Sampling took place between March and June 2022. At each entrance, we surveyed at least one specimen of every avascular component (bryophytes) species present, which were then placed into kraft specimen bags with annotated collection data. We defined cave flora as all specimens occurring from the speleometric start of the cave (plan closure). From this, data on bryophyte species composition was obtained for each sampled cave entrance.

Geographic variables (X and Y coordinates) and altitude were obtained using a handheld Garmin GPSMAP 64s. For the morphological characterization of the cave entrances, their dimensions (height and width), altitude, and slope of the entrance zone were measured using a handheld compass and a laser rangefinder. In our analysis, the dimensions of each cave entrance are considered as a ratio between height and width.

The bryophyte samples placed into kraft specimen bags were dried at room temperature. Specialists at the Bryophytes Laboratory of the Federal University of Juiz de Fora (UFJF) identified the plant samples using specialized literature. Identification was performed under an Olympus SZ040 or Zeiss Stemi-200C stereoscopic microscope, using tweezers, scalpels, and blades for sample preparation. Taxonomically significant structures were extracted and mounted between a slide and coverslip for observation under an Olympus BX41 or Zeiss Primo Star light microscope. The classification followed Goffinet et al., (2009), Frey & Stech, (2009) and Crandall-Stotler et al. (2009). All voucher specimens were deposited in the Herbarium Professor Leopoldo Krieger of the Federal University of Juiz de Fora (CESJ).

2.3. Data Analysis

To access the life-forms and substrate preferences intrinsic to each bryophyte species, we used the information available in the Flora do Brasil database (REFLORA, 2024). To check previous records of each species in PEIB, we consulted publications on PEIB bryoflora by Yano & Ponzo (2014) and Machado et al. (2016) and the scientific collections database *SpeciesLink* (SpeciesLink, 2024).

2.4. Phylogenetic Distance Computation

We used the mean pairwise distance (MPD) metric to calculate the average phylogenetic distance between species from different communities using the comdist function from the “picante” R package. We assessed the MPD for 70 bryophytes sampled at the cave entrances we studied. All species were identified with taxonomic accuracy at species level and just one to the genus level.

To access the phylogenetic distance, we converted the hierarchical taxonomic identities into a cluster format that measured the distances between taxonomic levels. These distances were then normalized by the number of levels, following the approach suggested by Clarke & Warwick (1999). We performed a Principal Coordinate Analysis (PCoA) using the *capscale* function from the "vegan" R package, applying the Euclidean distance metric. The first two axes of the PCoA (MDS1 and MDS2) were then used as proxies for phylogenetic distance in our models.

2.5. Effects of environmental features on phylogenetic distance

We used the mean pairwise distance (MPD) values between pairs of communities to perform piecewise regression analysis on the environmental variables measured using generalized linear models (Gaussian distributed errors with logistic link function for response variables) and transforming the predictor variables into pairwise differences using Euclidean distance matrices. These analyses allow us to identify trend changes (breakpoints) in relation to the response variables. We compared AICc values between different regression types (linear or segmented) to choose the best model, following the approaches of Ochoa-Quintero et al. (2015) and Magnago et al. (2015). The results were expressed by considering the variation range (Δ) of the segmented model compared to the linear one (Oliveira & Ferreira, 2024). We performed the piecewise regression using the "segmented" R package and the code provided by Oliveira & Ferreira (2024).

2.6. Patterns of compositional turnover

Zeta diversity (ζ) is a metric that quantifies the number of species shared across multiple assemblages, offering a comprehensive method for assessing biological diversity by combining incidence-based diversity measures, patterns, and relationships (Hui and McGeoch 2014; McGeoch et al. 2019). The zeta order indicates the number of assemblages analyzed: ζ1 represents alpha diversity, ζ2 corresponds to pairwise beta diversity, ζ3 denotes species shared among three sites, and so on. We applied the zeta diversity method to evaluate bryophyte species across multiple sites (Latombe et al., 2017). This method explores spatial variations in species assemblages, assuming that common species are found across many sites, while rare species are present in fewer sites. Consequently, zeta diversity tends to decline as the number of sites or orders increases. A steep decline in lower zeta orders suggests turnover driven by rare species, whereas a gradual decline indicates turnover due to common species. The retention rate measures the persistence of species as zeta order increases. By comparing zeta declines and retention rates across regions, we can better understand the roles of common and rare species in turnover (Latombe et al., 2018).

2.7. Zeta diversity computations

To explore general diversity patterns, we calculated species shared across two to six sites (zeta values ζ2 to ζ6) and average richness per site (alpha diversity, ζ1). In addition to raw zeta values (absolute species counts), we used normalized zeta values to address significant differences in site richness (McGeoch et al., 2019). We employed Simpson-equivalent zeta (ζij/Sj) and Sørensen-equivalent zeta (ζik/Søk) to assess nestedness' impact on turnover, as the Sørensen version incorporates nestedness while the Simpson version does not (Baselga 2010). Zeta diversity declines were analyzed using the 'Zeta.decline' function from the 'zetadiv' package in R (Latombe et al., 2018).

2.8. Environmental drivers of compositional turnover

To evaluate how spatial distance and environmental differences explain species turnover, we performed generalized dissimilarity models (GDM) with zeta diversity through MS-GDM, that provides a robust framework for explaining zeta diversity across varying numbers of sites. MS-GDM uses regression models based on environmental and spatial differences to account for how these factors influence species turnover. It employs non-linear splines to assess the impact of environmental variables and distances on pairwise turnover, extending this analysis to different zeta diversity orders, allowing explore how the influence of distance and environmental variables varies for species of different rarity levels and whether common species follow different rules compared to rare species.

We followed the approach outlined by Latombe et al. (2017, 2018) using the ‘zetadiv’ R package. Our analyses encompassed zeta orders 2 to 6, considering five environmental variables and the distances between sites.

3. RESULTS

3.1. Bryophyte diversity

In total, from the 13 entrances of eight caves sampled in the Ibitipoca State Park, 70 bryophyte species were recorded belonging to 27 families, including 37 Bryophyta (mosses), 32 Marchantiophyta (liverworts), and one species of Anthocerotophyta (hornworts).

Syrrhopodon prolifer Schwägr. (Calymperaceae) and *Leucobryum crispum* Müll. Hal. (Leucobryaceae), both mosses, were the most frequently encountered species in the sampled cave entrances, appearing in 10 entrances each. They were followed by *Symphyogyna podophylla* (Lindenb. & Gottsche) Grolle (Pallaviciniaceae), *Lejeunea flava* (Sw.) Nees (Lejeuneaceae), *Hypnella pilifera* (Hook. & Wilson) A. Jaeger (Pilotrichaceae), and *Campylopus lamellinervis* (Müll. Hal.) Mitt. (Leucobryaceae). The family Leucobryaceae was the most diverse, occurring in 12 entrances, followed by Calymperaceae, Pallaviciniaceae, Pilotrichaceae, and Lejeuneaceae (Fig. 2).

Figure 12 – Cave bryophytes sampling and cave entrances studied in PEIB.

The cave entrances sampled in our study revealed 14 species previously unrecorded in PEIB. Of these, seven were liverworts from five families: *Riccardia hymenophytoides* (Spruce) Meenks (Aneuraceae), *Cephalozia crossii* Spruce, *Odontoschisma brasiliense* Steph. (Cephaloziaceae), *Heteroscyphus marginatus* (Steph.) Fulford, *Lophocolea heterophylla* (Schrad.) Dumort. (Lophocoleaceae*), Symphyogyna leptothelia* Taylor (Pallaviciniaceae), and *Plagiochila aerea* Taylor (Plagiochilaceae). The remaining seven were mosses from six families: *Rhynchostegium serrulatum* (Hedw.) A. Jaeger (Brachytheciaceae), *Syrrhopodon gardneri* (Hook.) Schwägr. (Calymperaceae), *Fissidens serratus* Müll. Hal., *F. submarginatus* Bruch (Fissidentaceae), *Campylopus thwaitesii* (Mitt.) A. Jaeger (Leucobryaceae), *Lepidopilidium nitens* (Hornsch.) Broth. (Pilotrichaceae), and *Sphagnum magellanicum* Brid. (Sphagnaceae).

Additionally, eleven of the thirteen species recorded by Yano & Ponzo, (2014) reported to cave environments were not found in our study, specifically: *Brachymenium consimile* (Mitt.) Jaeger (Bryophyta: Bryaceae), *B. wrightii* (Sull.) Broth. var. *mnioides* (Besch.) (Bryophyta: Bryaceae), *Rhodobryum beyrichianum* (Hornsch.) Müll. Hal. (Bryophyta: Bryaceae), *Holomitrium olfersianum* Hornsch. (Bryophyta: Dicraniaceae), *Chryso-hypnum elegantulum* (Hook.) Hampe (Bryophyta: Hypnaceae), *Lepidopilum brevipes* Mitt., *L. scabrisetum* (Schwägr.) Steere (Bryophyta: Pilotrichaceae), *Leptoscyphus amphibolius* (Nees) Grolle (Marchantiophyta: Geocalycaceae), *Saccogynidium caldense* (Ångstr.) Grolle (Marchantiophyta: Geocalycaceae), *Syzygiella trigonifolia* (Steph.) Herzog, Hedwigia (Marchantiophyta: Jamesoniellaceae).

Regarding the life-forms, the most frequent in cave bryophytes were the leafy forms, followed by mat, tuft and welft forms (Fig. 3A). Rupiculous, terricolous and corticicolous were the most frequent preferred substrates of cave bryophytes (Fig. 3 B).

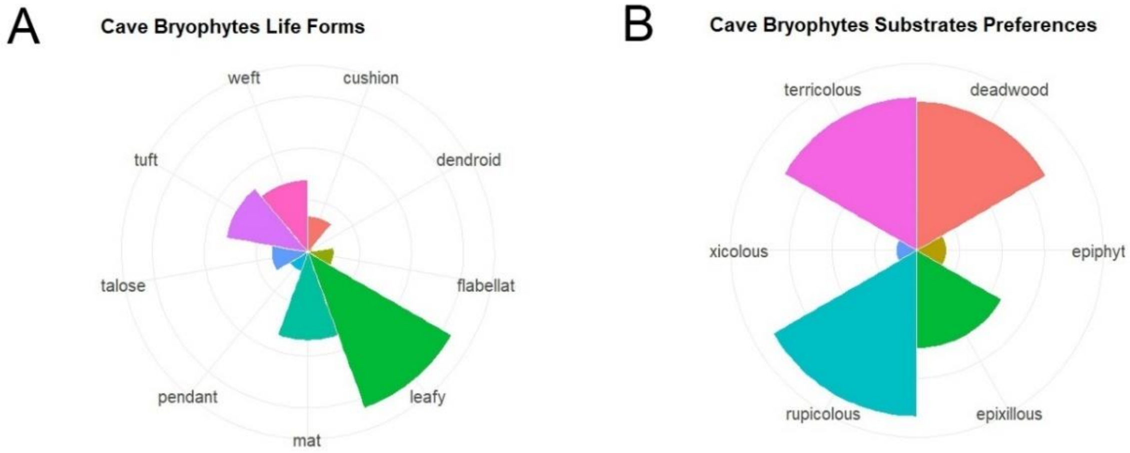

Figure 13 – A) Life Forms and B) and substrate preferences of the species collected at the entrances of the caves studied.

3.2. Effects of environmental features on phylogenetic distance

Geographical distance was a significant predictor of phylogenetic distance. Caves located closer to each other had bryophyte communities with greater phylogenetic similarity (Fig. 4a). Additionally, for cave entrances with a height-to-width ratio differences of less than 1, the bryophyte communities were found to be more phylogenetically similar. This indicates that as the differences of the ratio between height to width of the entrances decreased, the phylogenetic similarity between the bryophyte communities increased (Fig. 4b).

Models incorporating phylogenetic distance in relation to altitude, colonization depth, and slope did not provide significant explanations for the bryophyte communities sampled in the caves.

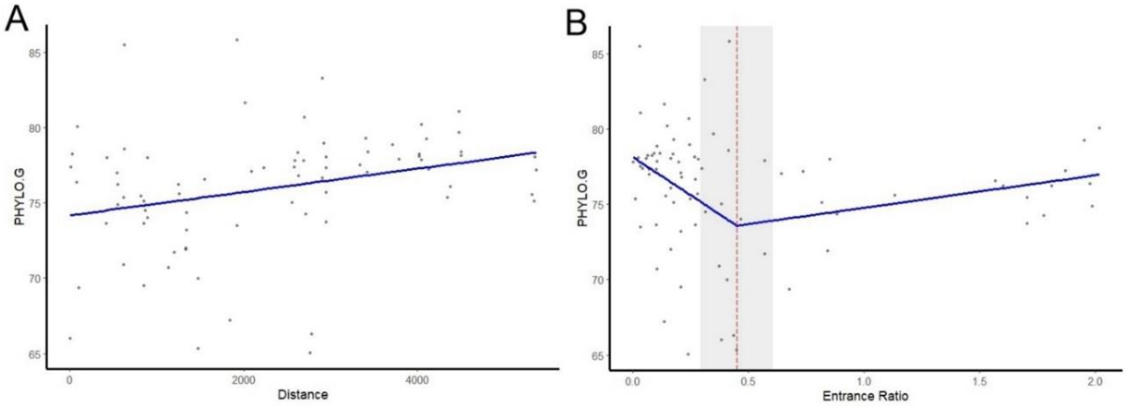

Figure 14 - Relationship between the PD among cave bryophyte communities and environmental variables. The plot A) shows the best model selected for distance and shows a linear effect of distance predicting PD. In B) The graph display threshold for entrance ratio which was a significant predictor of PD (the dashed vertical line (in blue) indicates the significant breakpoint ($p < 0.050$), the grey area represents the standard error). The ΔAICc denotes the variation of such model AIC in relation to the linear one.

3.3. Patterns of compositional turnover

Zeta diversity values rapidly declined to zero, while retention rates increased sharply. The patterns of decline were similar for both Sørensen and Simpson analyses (Fig. 5A, C), although the decline was slightly steeper for Sørensen at lower zeta orders, indicating minimal effects of nestedness in the system. Species retention rates began to decrease at higher zeta orders, suggesting that, for these higher orders, even the most common species are less likely to be retained as more sites are added. This trend highlights the influence of rare or site-specific species on species turnover (Fig. 5B, D).

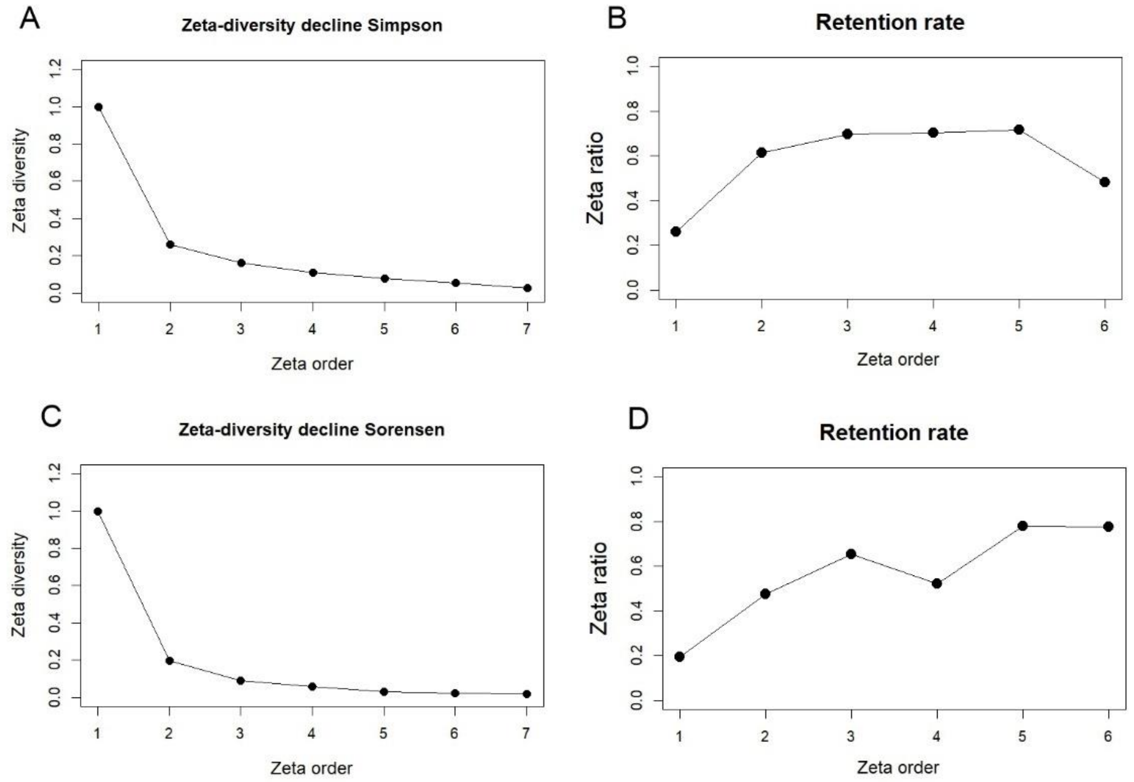

Figure 15 - Zeta diversity decline (A, C) and retention rate (C, D) for the bryophytes communities of cave entrances sampled. A) and B) shows the Simpson equivalent curves and C) and D) sows the Sørensen equivalent ones.

3.4. Environmental drivers of compositional turnover

The shapes of the *I-splines* curves reveal the range of each variable where its effect on species turnover is most significant, with steeper slopes indicating greater importance. The spatial distance between sites was a key variable explaining turnover for rare species (ζ2,3) but not for more common species (ζ4-6). For ζ2, the entrance ratio, slope, and depth of colonization also influenced turnover, while slope was the primary factor for ζ3. For common species (ζ4-6), turnover was primarily driven by slope and depth of colonization (Fig. 6).

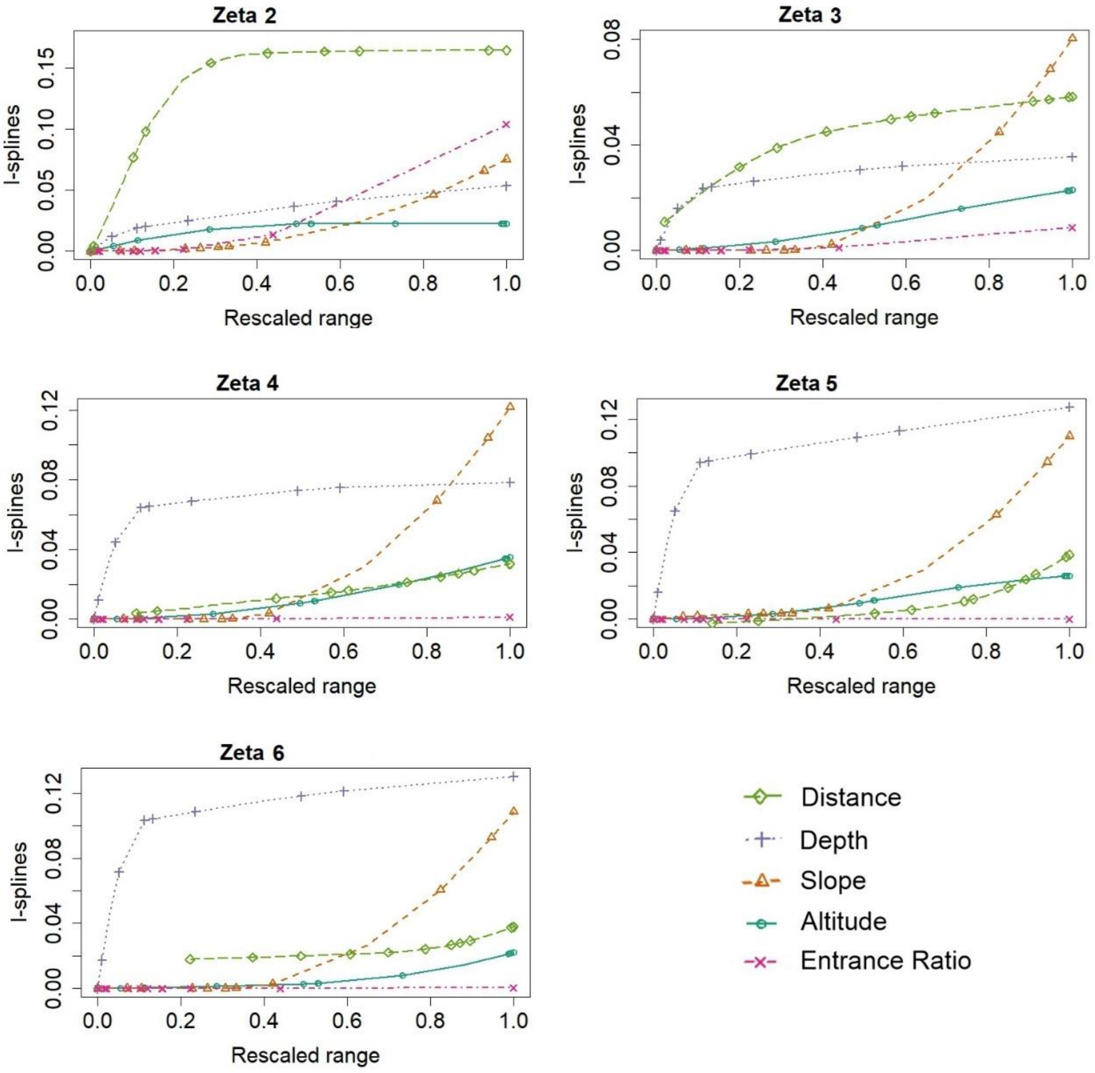

Figure 16 - MS-GDM analyses for zeta orders 2-6 using environmental features and straight-line distances between sites based on their geographic coordinates of the cave entrances bryoflora communities sampled.

The variance explained for the MS-GDM models were 42.9 % for zeta order 2, 59.7 % for zeta order 3, 62.5 % for zeta order 4, 56.1 % for zeta order 5 and 41.7 % for zeta order 6 (Fig. 7).

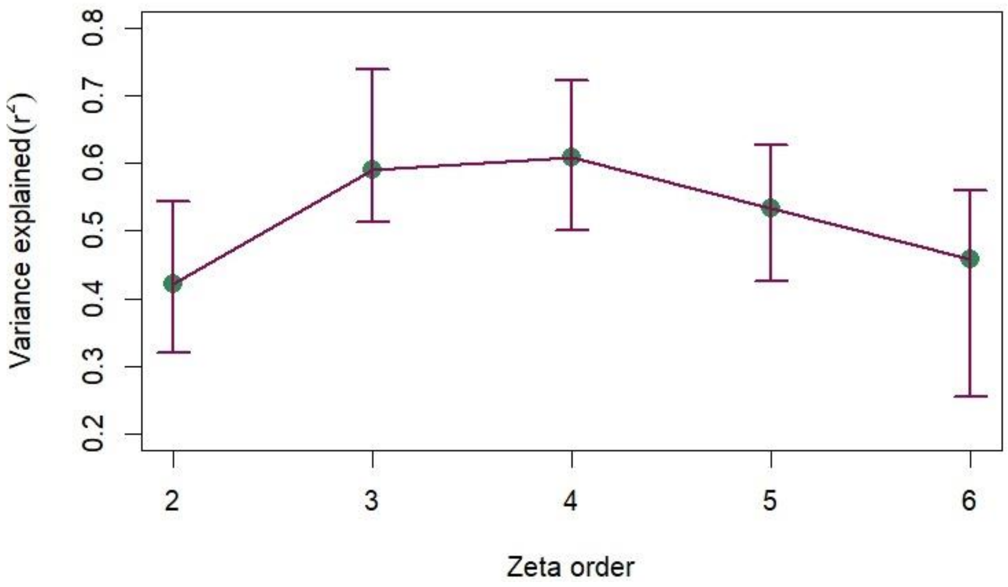

Figure 17 – Total variance explained by each zeta order MS-GDM model of bryophytes communities sampled at cave entrances.

4. DISCUSSION

Our study of 13 cave entrances in Ibitipoca State Park revealed a rich bryophyte diversity, with 70 species across 27 families. And 14 species previously unrecorded in the park. Geographical distance among caves and entrances and geomorphology, particularly the differences in height-to-width ratio, significantly influenced phylogenetic similarity in bryophyte communities. While factors such as altitude, slope, and colonization depth predicted in community diversity, spatial distance was crucial in driving turnover, especially for rare species.

4.1. Bryophyte diversity

Tropical regions are typically known for their rich biological diversity, with their forests ranking among the most threatened biomes globally (Myers et al., 2000). Caves in these regions also face significant anthropogenic threats (Ferreira et al., 2022). Despite ongoing conservation efforts aimed at protecting cave environments, particularly in the field of subterranean biology, there remains a notable gap in studies focusing on cave-dwelling flora within tropical environments.

Neotropical regions are home to numerous karst systems, with thousands of known caves. In Brazil, caves are widely distributed across all biomes, particularly in the two biodiversity hotspots, the Cerrado and Atlantic Forest (Myers, 2000). These caves are found in various rock types, including limestone, sandstone, and quartzite. The state of Minas Gerais, Brazil, contains several significant karst areas, especially in sandstone and quartzite terrains, showcasing a complex array of subterranean and surface karstic formations (Willems at al., 2008; Wray & Sauro, 2017). Ibitipoca State Park, located within a highly diverse Atlantic Forest formation, is notable for containing some of the country's most important caves in siliciclastic rock (Rubbiolli et al., 2019; Oliveira et al., 2024).

Furthermore, Ibitipoca State Park is particularly renowned for its exceptionally high bryophyte diversity within a relatively small area, compared to other regions in Brazil, and for its significant concentration of large caves (at least 60). In our study, which focused on thirteen cave entrances, we identified 70 bryophyte species. Previous temporal sampling by researchers documented at least 309 bryophyte species throughout the entire park (Yano & Ponzo, 2014; Machado, 2016). According to Yano and Ponzo (2014), only 13 of these species had been collected from cave entrances, most of which were in caves not included in our study. Eleven of the thirteen species recorded by these authors were not found in our research, highlighting the potential to expand the checklist of cave bryophytes in PEIB and to document new records.

Despite the extensive period of bryophyte studies and surveys at PEIB, our study identified 14 species in the caves that had not been previously recorded. Research has shown that caves can serve as important genetic reservoirs for flora, acting as refuges and habitats for endemic species, and this holds true for bryophytes as well (Gabriel et al., 2004; Cong et al., 2023). For instance, Gabriel et al. (2004) found that caves in the Azores harbor at least 35% of the known bryoflora for those areas.

Previous surveys at Ibitipoca State Park have identified liverworts as the most diverse group, followed by mosses (Yano & Ponzo, 2014). However, in our study of PEIB caves, mosses were found to be the more diverse group, though the difference was relatively small. Our results suggest that bryophyte substrate preferences in cave habitats align with patterns observed in montane forests within PEIB and are closely related to the types of substrates available in a given area (Yano & Ponzo, 2014). Thalloid liverworts typically colonize soil (terrestrial substrates), while leafy liverworts occupy a range of substrates, including rocks (saxicolous), decomposing logs (epixylic), living tree trunks and branches (corticicolous), and leaves (epiphytic) (Yano & Ponzo, 2014; Souza et al., 2020). In contrast, mosses are generally more habitat-generalists and can thrive under very low light conditions (Shaw & Goffinet, 2000; Herrero-Borgoñón, 2024). In cave entrances, substrates such as living tree trunks and branches, decomposing organic matter, and leaves (typically used by epiphytes) are scarce due to the oligotrophic nature of these environments and are more likely to be found near epigean habitats.

This scarcity may explain the smaller difference in diversity between liverworts and mosses compared to the external environment.

Regarding the life-forms observed in our study, it is evident that the subterranean environmental conditions act as significant filters, selecting the communities that can thrive there. In addition to leafy forms, turf, weft, and mat forms were the most frequent bryophyte life-forms found. These life-forms are functional traits that indicate how bryophytes respond to key environmental factors (Vittoz et al., 2010). For example, a relatively dense colony of shoots, such as those in cushion or turf forms, offers advantages over weft or fan colonies by storing water in the capillary spaces created between the gametophytes. However, dense life-forms receive less light due to self-shading, which aligns with our findings in caves where low-light conditions prevail (Wang et al., 2016). Based on this, bryophytes with mat and weft forms are expected to prefer shady and humid environments, while cushion and turf forms are more adapted to sun-exposed, arid conditions (Bates, 1998).

4.2. Phylogenetic distance

Regarding phylogenetic distance (PD), the distance between cave entrances and the entrance height-to-width ratio were key factors influencing the differences observed among the bryophyte groups sampled. As anticipated, the distance between entrances had a moderate impact. Most bryophytes thrive in relatively humid environments, as water is essential for their development, metabolism, and the motility of the male gamete (antherozoid) for fertilization (Lemos-Michel, 2001). Consequently, their dispersal is often moisture-dependent, and distance can serve as a filtering mechanism. Furthermore, given that PEIB is a mountainous region, greater distances between entrances create more substantial geographical barriers to species dispersal.

The dimensions of cave entrances had two significant relationships with phylogenetic distance. When the height-to-width ratio values of the entrances was less than one, that is, the height was less than the width, the effects on phylogenetic distance were negative. Conversely, entrances with greater height relative to width showed positive effects. This is expected, as taller entrances allow for more light penetration, fostering the growth of more diverse plant groups and increasing phylogenetic distance among them.

While we did not test the orientation of cave entrances in this study (due to limited variation in this factor among the entrances we examined) we anticipate it could be a significant factor related to incident light radiation and thus cannot be ruled out. The interaction between entrance dimensions and geographical orientation could potentially be important drivers of bryological community composition. Similar to small shelters, cracks, and crevices in rocky vegetation environments in Brazil, subterranean environments promote soil accumulation and water retention, creating less arid conditions by reducing overheating and drought and had significantly effects on the distribution of functional plant groups (Alves & Kolbek 1993; Conceição & Pirani 2005; Porembski 2007; Carmo et al., 2016, Badia et al., 2021).

4.3. Turnover Patterns

Zeta decline analyses reveal a rapid decrease from the second to higher zeta orders, underscoring the presence of a substantially greater number of rare species compared to more common ones within the cave bryophyte communities in PEIB. The decline patterns were similar in both Sørensen and Simpson analyses, with Sørensen showing a slightly steeper decline at lower zeta orders, suggesting minimal to no effect of nestedness within these communities (Latombe, 2017). Typically, such an exponential decline indicates an equal likelihood of species retention as the number of sites increases, highlighting the significant role of stochastic processes in species distribution (Fonte et al., 2021).

The pattern observed in the species retention rate curves, where retention rates initially increase but then decline at higher zeta orders, suggests that even the most common species become less likely to be retained as more sites are included. This indicates that compositional turnover is mainly driven by variations in rare species rather than common ones. The results reveal a small number of common species shared across multiple sites, alongside a large number of rare species with limited distribution. Caves often serve as habitats for rare species due to their unique environments, which differ significantly from one entrance to another, and because they act as geographic barriers, especially for smaller species (Culver & Pipan, 2019, Oliveira & Ferreira, 2024). This pattern is also evident in plants, with studies showing that caves can function as refuges for plant species, including endemics, as well as a high number of rare, relict, and threatened species (Gabriel et al., 2004; Monro et al., 2018; Cong et al., 2023; Puglisi et al., 2024; Cedrés-Perdomo et al., 2024; Deng et al., 2024).

4.4. Drivers of Turnover

The MS-GDM models accounted for a significant portion of the variation in species turnover. Spatial distance between entrances was a key variable in explaining turnover for rare species (ζ2-3) but had less impact on more common species (ζ4-6). Similarly, entrance dimensions were crucial for rarer species (ζ2), indicating that phylogenetic distance is lower when more species are shared. As zeta orders increased, slope and colonization depth became more significant factors. Steeper negative slopes at the entrances, as observed in the studied caves, may correlate with higher nutrient and moisture inputs from the surface environment, which provide substrates conducive to greater species colonization. This could account for the effects observed on more common species. Many of these plants thrive in relatively humid conditions, as water is vital for their development, metabolism, and the motility of male gametes (antherozoids) necessary for fertilization (Lemos-Michel, 2001).

5. CONCLUSIONS

In conclusion, our study underscores that, similar to findings in non-tropical caves, some Brazilian caves serve as important habitats for bryophyte communities, supporting highly diverse and unique assemblages. Our research in Ibitipoca State Park identified 70 bryophyte species, including 14 new records, and highlighted that cave geomorphological features (such as entrance dimensions and slope) as well as geographic distance, are significant factors in predicting bryophyte diversity and phylogenetic similarity among subterranean habitats. Furthermore, rare species appear to play a key role in driving species turnover. This research provides valuable insights into the ecology of cave flora in tropical regions and supports ongoing conservation efforts aimed at protecting cave habitats and their associated flora.

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APPENDIX I

Table 1 - Bryophytes species surveyed within cave entrances of the Ibitipoca State Park.

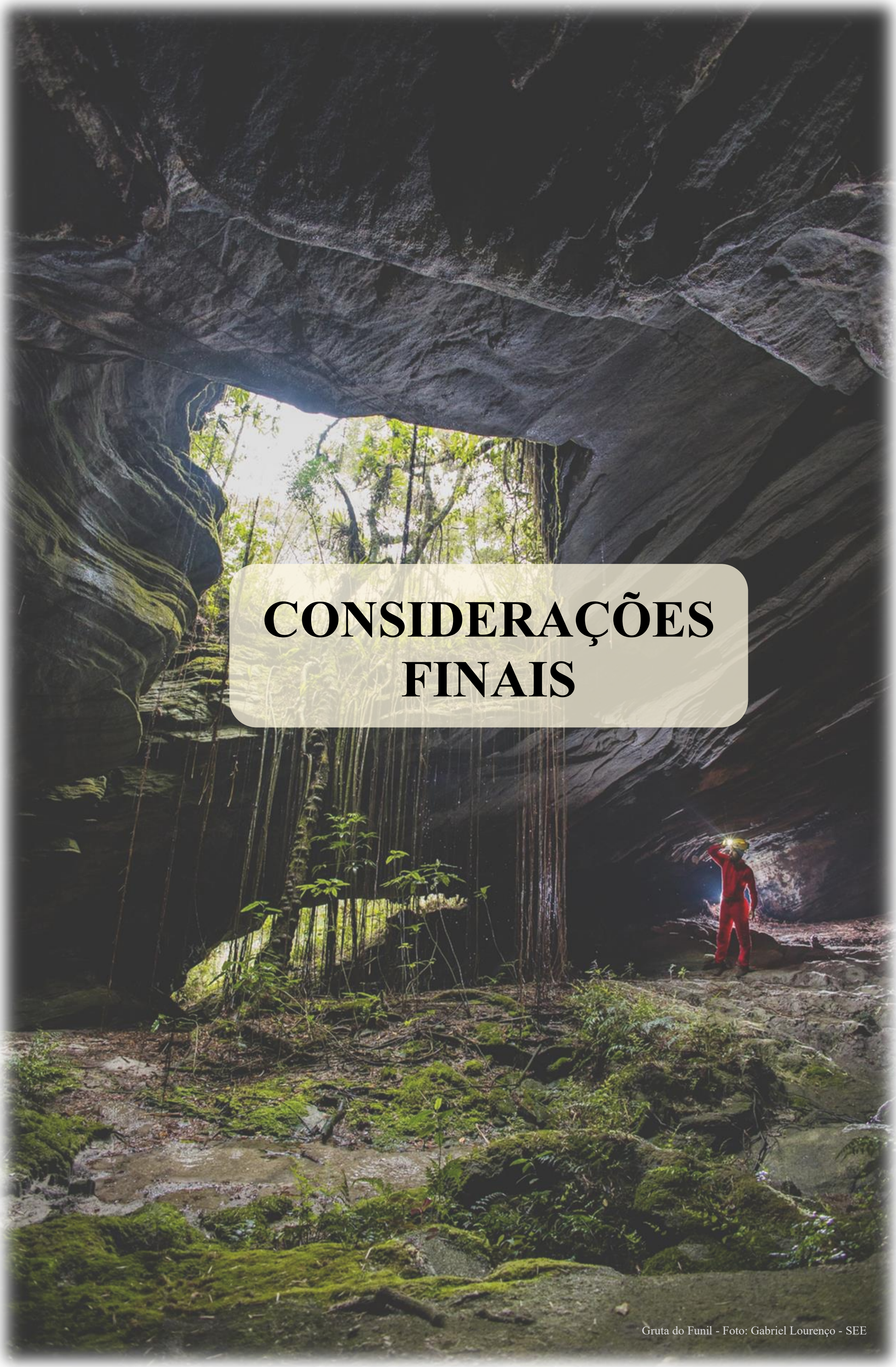
Family/Species	Cave	Voucher
Anthocerotophyta		
Dendrocerotaceae		
Nothoceros vincentianus (Lehm. & Lindenb.) J. C. Villarreal	PP	R.C. Cardoso, 249
Bryophyta		
Bartramiaceae		
Philonotis cernua (Wilson) D.G. Griffin & W.R. Buck	Vi, Ab	R.C. Cardoso, 240
Brachytheciaceae		
Rhynchostegium serrulatum (Hedw.) A. Jaeger	Mo	R.C. Cardoso, 313
Calymperaceae		
Syrrhopodon gardneri (Hook.) Schwägr.	Ab	R.C. Cardoso, 256
Syrrhopodon prolifer Schwägr.	Bo, Ab, Mn, Br, Mo, Vi, PP	R.C. Cardoso, 244
Syrrhopodon gaudichaudii Mont.	PP, Bo, Ab	R.C. Cardoso, 249
Catagoniaceae		
Catagonium brevicaudatum Müll. Hal.	Mo	R.C. Cardoso, 280
Catagonium nitens (Brid.) Cardot	Mo	R.C. Cardoso, 320
Fissidentaceae		
Fissidens elegans Brid.	Bo, Mn, Mo	R.C. Cardoso, 244
Fissidens scariosus Mitt.	Mo, Ca	R.C. Cardoso, 291
Fissidens serratus Müll.Hal.	Mn	R.C. Cardoso, 263
Fissidens submarginatus Bruch	Mo	R.C. Cardoso, 291
Hypopterygiaceae		
Hypopterygium tamarisci (Sw.) Brid. ex Müll. Hal.	PP, Ab	R.C. Cardoso, 246
Leucobryaceae		
Campylopus arctocarpus (Hornsch.) Mitt.	Mo	R.C. Cardoso, 298
Campylopus filifolius (Hornsch.) Spruce	Br	R.C. Cardoso, 293
Campylopus heterostachys (Hampe) A. Jaeger	PP	R.C. Cardoso, 247
Campylopus lamellinervis (Müll. Hal.) Mitt.	PP, Bo, Br	R.C. Cardoso, 246
Campylopus savannarum (Müll. Hal.) Mitt.	Vi	R.C. Cardoso, 297
*Campylopus thwaitesii (*Mitt.) A. Jaeger	Ab	R.C. Cardoso, 255
Leucobryum albicans (Schwägr.) Lindb.	Br	R.C. Cardoso, 311
Leucobryum crispum Müll. Hal.	PP, Ab, Mn, Br, Mo, Vi, Ca	R.C. Cardoso, 248
Ochrobryum gardneri (Müll. Hal.) Mitt.	Bo	R.C. Cardoso, 242
Phyllogoniaceae		
Phyllogonium viride Brid.	Mn, Br	R.C. Cardoso, 264
Pilotrichaceae		
Hypnella pilifera (Hook. & Wilson) A. Jaeger	Br, Vi	R.C. Cardoso, 307
Lepidopilidium nitens (Hornsch.) Broth.	Mo	R.C. Cardoso, 312
Thamniopsis undata (Hedw.) W.R. Buck	PP	R.C. Cardoso, 251
Trachyxiphium guadalupense (Brid.) W.R. Buck	Br	R.C. Cardoso, 269
Polytrichaceae		
Pogonatum sp	Mo	R.C. Cardoso, 306
Pylaisiadelphaceae		
Isopterygium tenerum (Sw.) Mitt.	Ab, Ca, Mo	R.C. Cardoso, 255

Family/Species	Cave	Voucher
Rhizogoniaceae		
Pyrrhobryum spiniforme (Hedw.) Mitt.	Ab, Vi	R.C. Cardoso, 255
Sematophyllaceae		
Jirivanaea cuspidifera (Mitt.) U.B. Deshmukh & Rathor	Mn, Br	R.C. Cardoso, 272
Sematophyllum beyrichii (Hornsch.) Broth.)	Vi, Mn	R.C. Cardoso, 289
Sematophyllum subsimplex (Hedw.) Mitt.	Br	R.C. Cardoso, 299
Sphagnaceae		
Sphagnum perichaetiale Hampe	Mo	R.C. Cardoso, 316
Sphagnum subsecundum Nees	PP, Vi	R.C. Cardoso, 252
Sphagnum magellanicum Brid.	PP	R.C. Cardoso, 248
Marchantiophyta		
Aneuraceae		
Riccardia digitiloba (Spruce ex Stephani) Pagán	Mo, PP	R.C. Cardoso, 292
Riccardia hymenophytoides (Spruce) Meenks	Br, Br	R.C. Cardoso, 269
Cephaloziaceae		
Cephalozia crossii Spruce	Vi	R.C. Cardoso, 290
Odontoschisma brasiliense Steph.	Mo, Vi	R.C. Cardoso, 279
Odontoschisma denudatum (Mart.) Dumort.)	Bo	R.C. Cardoso, 238
Frullaniaceae		
Frullania beyrichiana (Lehm. & Lindenb.) Lehm. & Lindenb.	Mn, Ca	R.C. Cardoso, 261
Geocalycaceae		
Calypogeia laxa Gottsche & Lindenb.	Mn	R.C. Cardoso, 271
Lejeuneaceae		
Lejeunea flava (Sw.) Nees	Mn, Br, Mo	R.C. Cardoso, 276
Lejeunea laetevirens Nees & Montagne	Ca	R.C. Cardoso, 282
Lejeunea phyllobola Nees & Mont.	Mn	R.C. Cardoso, 266
Lepidoziaceae		
Bazzania hookeri (Lindenb.) Trevis	Vi	R.C. Cardoso, 287
Bazzania nitida (F. Weber) Grolle	Bo	R.C. Cardoso, 238
Kurzia brasiliensis (Steph.) Grolle)	Bo	R.C. Cardoso, 237
Kurzia capillaris (Sw.) Grolle).	Bo	R.C. Cardoso, 238
Lepidozia cupressina (Sw.) Lindenb.	Vi	R.C. Cardoso, 288
Telaranea nematodes (Gottsche ex Austin) M. Howe	Bo, Mn	R.C. Cardoso, 244
Leucomiaceae		
Rhynchostegiopsis brasiliensis Broth.	Br	R.C. Cardoso, 323
Lophocoleaceae		
Heteroscyphus marginatus (Steph.) Fulford	Br	R.C. Cardoso, 281
Leptoscyphus amphibolius (Nees) Grolle	Mn	R.C. Cardoso, 274
Lophocolea heterophylla (Schrad.) Dumort.	Mn	R.C. Cardoso, 262
Lophocolea bidentata (L.) Dumort.	Bo	R.C. Cardoso, 244
Pallaviciniaceae		
Symphyogyna aspera Steph. ex F.A. McCormick	Ab	R.C. Cardoso, 260
Symphyogyna brasiliensis Nees	Mn, Mo	R.C. Cardoso, 270
Symphyogyna leptothelia Taylor	Mo	R.C. Cardoso, 283
Symphyogyna podophyla (Lindenb. & Gottsche) Grolle	PP, Vi, Mo, Br, Bo	R.C. Cardoso, 249
Plagiochilaceae		

Family/Species	Cave	Voucher
Plagiochila exigua (Taylor) Taylor	Mo	R.C. Cardoso, 300
Plagiochila cf aerea Taylor	Br	R.C. Cardoso, 299
Plagiochila gymnocalycina (Lehm. & Lindenb.) Lindenb.	Mo	R.C. Cardoso, 316
Plagiochila rutilans Lindenb.	Vi, Br	R.C. Cardoso, 321
Plagiochila simplex (Sw.) Lindenb.	Ca, Mo	R.C. Cardoso, 318
Radulaceae		
Cladoradula boryana (F. Weber) M.A.M. Renner, Gradst., Ilk.-Borg. & F.R. Oliveira-da-Silva	Mo	R.C. Cardoso, 314
Radula pallens (Sw.) Nees & Mont.	Vi, Mo	R.C. Cardoso, 253

TERCEIRA PARTE - CONSIDERAÇÕES FINAIS

CONSIDERAÇÕES FINAIS



Gruta do Funil - Foto: Gabriel Lourenço - SEE

CONSIDERAÇÕES FINAIS

A investigação conduzida nesta tese demonstrou, de maneira pioneira, a notável diversidade, complexidade ecológica e características adaptativas da flora de cavernas em sistemas neotropicais, especificamente no Parque Estadual do Ibitipoca. Os resultados destacam como as zonas de entrada das cavernas, moldadas por características geomorfológicas e ambientais únicas, funcionam como ecótonos essenciais que sustentam assembléias distintas de plantas, incluindo espécies raras e comunidades de briófitas diversificadas filogeneticamente. Este estudo contribui para nossa compreensão da flora subterrânea, enfatizando o papel desses habitats na preservação da biodiversidade de plantas e seu potencial de atuação como refúgios climáticos, especialmente em ecossistemas vulneráveis e ameaçados, como a Mata Atlântica.

A diferenciação na composição de espécies ao longo das zonas de profundidade e das entradas destaca a importância dos fatores abióticos, como gradientes de luz, propriedades do solo e geomorfologia das entradas, como propulsores da estrutura das comunidades de plantas subterrâneas. Além disso, os efeitos significativos da distância geográfica e das dimensões das entradas na similaridade filogenética reforçam o papel dos componentes espaciais na configuração dessas comunidades, particularmente para as briófitas. Esses insights têm implicações mais amplas para a conservação, pois sustentam a noção de que as cavernas abrigam assembléias ecológicas únicas, frequentemente compostas por espécies raras e especializadas com distribuições restritas.

Em conclusão, a inclusão da flora subterrânea nas políticas de conservação, especialmente em hotspots de biodiversidade, é extremamente importante, e é urgente que mais estudos sejam realizados para criar bases mais robustas sobre a flora de cavernas brasileiras. Proteger os ambientes cavernícolas não apenas conserva flora única, mas também apoia funções mais amplas do ecossistema. Estudos futuros são encorajados a explorar sistemas de cavernas adicionais com litologias diversas, visando aprofundar nosso entendimento das adaptações ecológicas e evolutivas das plantas de cavernas. Esses esforços serão cruciais para o avanço das iniciativas de conservação e mitigação dos impactos das mudanças ambientais nesses habitats únicos e frequentemente negligenciados.

FINAL REMARKS

The investigation conducted in this thesis has demonstrated, in a pioneering way, the remarkable diversity, ecological complexity, and adaptive traits of cave flora in Neotropical cave systems, specifically within Ibitipoca State Park. The findings highlight how cave entrance zones, shaped by unique geomorphological and environmental features, function as essential ecotones that support distinct plant assemblages, including rare species and phylogenetically diverse bryophyte communities. This study contributes to our understanding of subterranean flora, emphasizing the role these habitats in preserving plant biodiversity and possible acting as climatic refugees, particularly within vulnerable and endangered ecosystems like the Atlantic Forest.

The differentiation in species composition across depth zones and entrances underscores the importance of abiotic factors, such as light gradients, soil properties, and entrance geomorphology, as drivers of subterranean plant community structure. Furthermore, the significant effects of geographical distance and entrance dimensions on phylogenetic similarity reinforce the role of the spatial components in shaping these communities, particularly for bryophytes. These insights have broader implications for conservation, as they support the notion that caves harbor unique ecological assemblages, often comprising rare and specialized species with restricted distributions.

In closing, necessity of including subterranean flora in conservation policies, particularly within biodiversity hotspots it is extremely important, and it is urgent that more studies be carried out to create more robust bases on the Brazilian cave flora. Protecting cave environments not only conserves unique flora but also supports broader ecosystem functions. Future studies are encouraged to explore additional cave systems with diverse lithologies, aiming to deepen our understanding of the ecological and evolutionary adaptations of cave dwelling plants. Such efforts will be crucial for advancing conservation initiatives and mitigating the impacts of environmental changes on these unique and often overlooked habitats.