UNIVERSIDADE FEDERAL DO PARANÁ

MARIA CLARA ALENCASTRO

SITE SELECTION AND SPATIAL DISTRIBUTION OF THE PHYTOTELM-BREEDING AMPHIBIAN *MELANOPHRYNISCUS VILAVELHENSIS* (ANURA, BUFONIDAE)

CURITIBA

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Dissertação apresentada ao curso de Pós-Graduação em Ecologia e Conservação, Setor de Ciências Biológicas, Universidade Federal do Paraná, como requisito parcial à obtenção do título de Mestre em Ecologia e Conservação.

Orientador: Prof. Dr. Mauricio O. Moura

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TERMO DE APROVAÇÃO

Os membros da Banca Examinadora designada pelo Colegiado do Programa de Pós-Graduação ECOLOGIA E CONSERVAÇÃO da Universidade Federal do Paraná foram convocados para realizar a arguição da dissertação de Mestrado de MARIA CLARA ALENCASTRO intitulada: SITE SELECTION AND SPATIAL DISTRIBUTION OF THE PHYTOTELM-BREEDING AMPHIBIAN MELANOPHRYNISCUS VILAVELHENSIS (ANURA, BUFONIDAE), sob orientação do Prof. Dr. MAURICIO OSVALDO MOURA, que após terem inquirido a aluna e realizada a avaliação do trabalho, são de parecer pela sua APROVAÇÃO no rito de defesa.

A outorga do título de mestra está sujeita à homologação pelo colegiado, ao atendimento de todas as indicações e correções solicitadas pela banca e ao pleno atendimento das demandas regimentais do Programa de Pós-Graduação.

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No fim, no topo ou do outro lado do mundo, nosso amor é infinito. Dedicado aos meus pais.

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Furu ike ya
kawazu tobikomu
mizu no oto

古池
蛙飛び込む
水の音

(Haicai de Bashô)

velha lagoa
o sapo salta
o som da água

(trad. Paulo Leminski)

RESUMO

Anfíbios anuros ocupam diversos tipos de habitats, dentre esses destacam-se plantas que possuem a capacidade de acumular água em sua estrutura (fitotelmos). A maneira pelo qual os indivíduos ocupam esses sítios não é aleatória, respondendo a fatores bióticos e abióticos. As escolhas dos indivíduos têm consequências em uma escala maior, da dinâmica da população, que, pode então ser estudada a partir dessa perspectiva. Para anuros, a sobrevivência aparente e a capturabilidade, podem ser influenciadas tanto pelas escolhas individuais quanto pelas condições ambientais. Deste modo o objetivo deste estudo foi determinar quais variáveis estão associadas à seleção de habitat pelo anuro fitotelmata *Melanophryniscus vilavelhensis* Steinbach-Padilha, 2009 além de descrever aspectos de história natural, informações demográficas e ajustar um modelo de dinâmica populacional. O trabalho foi conduzido em uma área de Campos Naturais associado à Floresta com Araucária no Parque Estadual Vila Velha (PEVV). A amostragem ocorreu por meio de busca ativa no período noturno em duas áreas do parque, de Novembro de 2021 a abril de 2022 e agosto de 2022 a janeiro de 2023, totalizando 15 expedições de campo. Realizou-se a mensuração (comprimento rostro cloacal - CRC) e fotoidentificação de todos os indivíduos capturados. As medidas morfométricas dos fitotelmos (altura, raio, número de folhas e densidade da mesma espécie) e as propriedades físico-químicas da água (pH, condutividade, temperatura e quantidade de detritos) foram mensuradas nas plantas com presença do anuro e em uma planta de controle (ausente). Calculou-se a probabilidade de ocorrência do anuro em função das variáveis preditoras baseado em uma seleção de modelos lineares generalizados (GLM) com erros com distribuição binomial e função link logit. Realizou-se também uma regressão linear entre os parâmetros selecionados e o tamanho dos anuros. Para a dinâmica de população foi ajustado o modelo Cormack-Jolly-Seber (CJS) para populações fechadas. Todas as análises foram realizadas pelo software R. Foram capturados 62 indivíduos (55 machos e 7 fêmeas) e entre os trinta eventos de recaptura, treze (43,33%) ocorreram na mesma planta em que o anuro foi originalmente encontrado. A sobrevivência aparente dessa população foi constante no tempo com o valor de 0.996 entre as ocasiões de coleta, e a capturabilidade variou com a média de 0.265 ± 0.354. As variáveis relacionadas à arquitetura da planta foram as mais relevantes para explicar a escolha de fitotelmo pelo anuro. *M. vilavelhensis* elege como sítio plantas agrupadas (maior densidade) e menores (em altura e raio). Além disso machos maiores habitam plantas mais altas com tanques maiores. Em conclusão, nossos resultados sugerem que o anuro fitotelmata *M.vilavelhensis* exibe uma alta fidelidade de sítio e ocupa o microhabitat baseado em restrições morfológicas (tamanho). A distribuição geográfica restrita da espécie e a dependência de fitotelmos para a reprodução ressalta a importância da conservação não só para a espécie em si, mas também para o bioma Mata Atlântica, particularmente o mosaico de Campos Naturais e Florestas com Araucária.

Palavras-chave: Fidelidade de sítio; Fitotelmo; Seleção de habitat; Dinâmica de população; História natural.

ABSTRACT

Anuran amphibians occupy various types of habitats, including plants that have the ability to accumulate water in their structures, known as phytotelms. The way individuals occupy these sites is not random and may be related to biotic and abiotic factors. By extrapolating individual choices to a larger scale, the contribution of these and other factors to population dynamics can be investigated. For anurans, apparent survival and detectability can be influenced by both individual choices and environmental conditions. Therefore, the objective of this study was to determine which variables are associated with habitat selection by the anuran species *Melanophryniscus vilavelhensis* Steinbach-Padilha, 2009 besides describing natural history data, demographic information, and adjusting a population dynamics model. The study was conducted in an area of natural grasslands associated with Araucaria Forest in Vila Velha State Park (VVSP). Active searches were carried out during the nighttime in two areas of the park from November 2021 to April 2022 and August 2022 to January 2023, totaling 15 field expeditions. Measurement was carried out (snout-vent length - SVL) and photo-identification of all captured individuals was performed. The morphometric measurements of the phytotelmata (height, radius, number of leaves, and density of conspecifics) and the physico-chemical properties of the water (pH, conductivity, temperature, and amount of debris) in the plants with the presence of the anuran and in a control plant (absent) were assessed. The probability of occurrence of the anuran was calculated based on predictor variables using a selection of generalized linear models (GLMs) with binomial distribution errors and a logit link function. A linear regression was also conducted between the selected parameters and the size of the anurans. For population dynamics, the Cormack-Jolly-Seber (CJS) model for closed populations was used. All analyses were performed using the R software. A total of 62 individuals were captured (55 males and 7 females), and among the thirty recapture events, thirteen (43.33%) occurred on the same plant where the anuran was originally found. The apparent survival of this population remained constant over time, with a value of 0.996, while capturability varied across occasions with a mean of 0.265 ± 0.354. Variables related to plant architecture were the most relevant in explaining the phytotelm selection by the anuran. *M. vilavelhensis* preferentially selected clustered plants (higher density) and smaller ones (in terms of height and radius). Additionally, larger males were found inhabiting taller plants with larger tanks. In conclusion, our findings suggest that the phytotelm-breeding anuran *M. vilavelhensis* exhibits high site fidelity and occupies the microhabitat based on morphological constraints (size). The species' restricted geographic distribution and dependence on phytotelms for reproduction underscore the importance of conservation, not only for the species itself but also for the Atlantic Forest biome, particularly the mosaic of Natural Grasslands and Araucaria Forests.

Keywords: Site fidelity; Phytotelm; Habitat selection; Population dynamics; Natural history

SUMÁRIO

1 INTRODUCTION

The habitat of a species can be defined as a spatially limited area with a subset of physicochemical and biotic conditions (Morris, 2003). The within-habitat species spatial dynamics is a nonrandom behavioral process (Fretwell & Calver, 1969) that reflects how individuals interact with the environment (Morris, 2003; De Oliveira & Navas, 2004; Matthiopoulos *et al.*, 2015). The habitat selection strategy of a species reflects then choices that could maximize fitness, through survival and reproductive success (Jaenike & Holt, 1991; Morris, 2003; Von May *et al.*, 2009; Cunha & Napoli, 2016) . Therefore, habitat selection theories encompass density-dependent ecological and evolutionary mechanisms resulting from interspecific and intraspecific biotic factors such competition and predation, as well as abiotic factors such as the physicochemical conditions of the environment (Morris, 2003; Rudolf & Rödel, 2005; Von May *et al.*, 2009; Domingos *et al.*, 2015; Mageski *et al.*, 2016).

Among vertebrates, amphibians occupy various types of habitats and exhibit a wide range of reproductive modes (Peloso *et al.*, 2012; Haddad *et al.*, 2013; Nunes-de-Almeida, Haddad & Toledo, 2021). Within the variety of microhabitats, phytotelms are water-filled cavities within plants or plant parts. This creates specialized tanks that foster the formation of communities within micro-habitats with unique trophic dynamics (Kitching, 2000). The communities are assembled because the rainwater accumulated between leaves and/or central tanks of plants sustains a diversity of invertebrates and vertebrates (Protázio *et al.*, 2013; Mageski *et al.*, 2016; Sanches *et al.*, 2019; Jorge *et al.*, 2020). Some species of amphibians establish occasional relationships with phytotelms, using them only as shelter during dry periods or for foraging. However, others have an obligatory association and are found throughout the year occupying phytotelms, depending on this environment for reproduction (Peixoto, 1975).

Communities are subject to high levels of spatial and temporal variation in resources, particularly temperature and precipitation which impacts community structure (Céréghino *et al.*, 2018). So, anurans inhabiting phytotelms are subjected to situations such as desiccation and unpredictability in food availability. The development of tadpoles is closely associated with water levels and quality (Castro & Pinto, 2000; Cunha & Napoli, 2016). Thus, these microhabitats can be more restrictive and exhibit a greater effect of microclimatic seasonality, playing a role as a limiting resource for reproduction compared to larger water bodies (De Oliveira & Navas, 2004).

In this case, selective habitat occupation may be related to physical factors of plant structure (height, radius, water storage capacity, number of leaves, individual density) and physicochemical factors related to water quality (pH, conductivity, dissolved oxygen, saturated oxygen, water temperature, debris quantity) (De Oliveira & Navas, 2004; Domingos *et al.*, 2015; Cunha & Napoli, 2016; Mageski *et al.*, 2016). There is evidence that plants occupied by anurans may be closer to the ground(De Oliveira & Navas, 2004; Mageski *et al.*, 2016)have larger tanks (De Oliveira & Navas, 2004), greater water retention capacity (Von May *et al.*, 2009; Domingos *et al.*, 2015), and a higher plant density (Cunha & Napoli, 2016). The water in these habitats may have a more alkaline pH (De Oliveira & Navas, 2004; Domingos *et al.*, 2015), low conductivity (Mageski *et al.*, 2016), and fewer debris (Cunha & Napoli, 2016). The choice of habitat based on these, and other characteristics can affect tadpole survival and most likely the reproductive success of adults (De Oliveira & Navas, 2004).

When individual-scale decisions are extrapolated to a larger scale, the contribution of these and other factors to population dynamics can be investigated. For anurans, population parameters such as apparent survival and capturability can be influenced by both individual choices and environmental conditions (Duellman & Trueb, 1994). It is known that amphibian dynamics, which are based on reproduction and survival, are directly influenced by factors such as humidity, temperature, and precipitation (Duellman & Trueb, 1994; Beebee & Griffiths, 2005; Hiert & Moura, 2010). Thus, how an individual selects its reproductive site has effects scaled to the population level.

At least 99 anuran species worldwide exclusively depend on bromeliads as phytotelm to complete their life cycle (Sabagh, Ferreira & Rocha, 2017). Most studies have focused on species from the families Hylidae and Dendrobatidae (De Oliveira & Navas, 2004; Protázio *et al.*, 2013; Domingos *et al.*, 2015; Cunha & Napoli, 2016; Mageski *et al.*, 2016), indicating a data gap for species from other families. Within the Bufonidae family, only seven from fifty-three genera exhibit a phytotelm-breeding habit, with four occurring in tropical regions (Langone *et al.*, 2008; Frost, 2023). For the genus *Melanophryniscus*, the focus of this study, 5 out of 31 valid species have been described as phytotelm-breeding (Bornschein et al., 2015; Langone et al., 2008; Steinbach-Padilha, 2008). All phytotelm-breeding *Melanophryniscus* spp. are distributed in Southern Brazil, restricted to the states of Paraná and Santa Catarina.

For this study, we selected *Melanophryniscus vilavelhensis*, which is found exclusively in Vila Velha State Park (VVSP) in the municipality of Ponta Grossa, Paraná State, in a mosaic of natural grasslands and Araucaria forests. The species is associated with two phytotelm genera: *Eryngium* sp. (Apiaceae) and *Eriocaulum* sp. (Eriocaulaceae) (Steinbach-Padilha, 2008). The lack of behavioral and demographic data for this species results in an undefined conservation status. The dependence on phytotelms for reproduction highlights the importance of conservation not only for the species itself but also for the Atlantic Forest biome, particularly the mosaic of Araucaria forests and natural grasslands in which the species is found (Steinbach-Padilha, 2008).

The main objective of this study is, therefore, to determinate: 1) if *M. vilavelhensis* selects plants or not; 2) which plant traits *M. vilavelhensis* uses to select a site for shelter and reproduction. Also, we aim to measure 3) the population dynamics parameters of apparent survival and capturability and enhance 4) the natural history data.

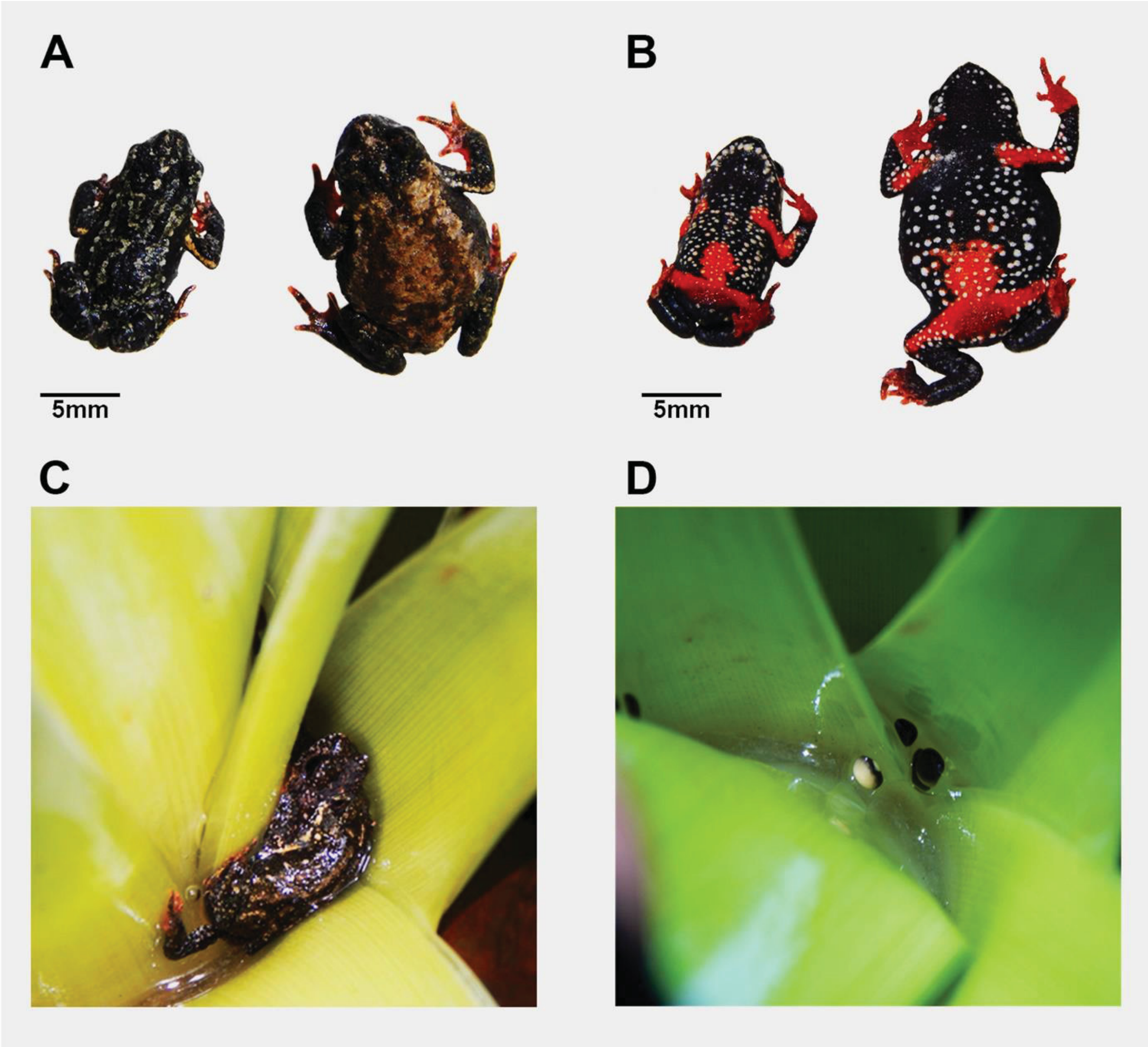
2 MATERIAL AND METHODS

2.1 STUDY ORGANISMS

The genus *Melanophryniscus* Gallardo, 1961 comprises 31 recognized species, occurring from the coastal lowlands of southern Brazil to northern Argentina, including Paraguay, Uruguay, and the interandean valleys of southern Bolivia (Frost, 2023). *Melanophryniscus* is a basal and monophyletic genus within the Bufonidae (Graybeal & Cannatella, 1995; Frost, 2023). Until 2008, all species of the genus *Melanophryniscus* were supposed to exhibit explosive breeding behavior, with reproductive events occurring for a few days after heavy rainfall, during which they would lay their eggs in temporary ponds or streams (Caorsi, 2011). However, in 2008, a new reproductive mode was described for the genus. *Melanophryniscus alipioi* Langone, Segalla, Bornschein, and de Sá, 2008 was the first species within the genus to reproduce in the water accumulated within the leaf axils of bromeliads, a behavior also reported in six other genera of the Bufonidae (Langone *et al.*, 2008). Since the description of *M. alipioi*, four additional species have been described as phytotelm-breeding: *M. vilavelhensis, M. biancae, M. milanoi,* and *M. xanthostomus* (Steinbach-Padilha, 2008; Bornschein *et al.*, 2015).

The model species for this study, *M. vilavelhensis* (Figure 01), is characterized by its small size (SVL males 12.8-14mm), nocturnal behavior, and its exclusive use of a unique breeding site (Steinbach-Padilha, 2008). *Melanophryniscus vilavelhensis* reproduces in the water accumulated in the leaf axils of *Eryngium* sp. (Apiaceae) and *Eriocaulon* sp. (Eriocaulaceae) (Steinbach-Padilha, 2008) and is restricted to two populations. The first population is located at the type locality, Vila Velha State Park (25° 13′ S, 50° 01′ W; 824 m a.s.l.), in the municipality of Ponta Grossa, Paraná State. The second population is found in a nearby rural private property (L.B. Crivellari, pers. comm., 2020). The Park consists of a mosaic ecosystem composed of patches of Araucaria Forest and natural grassland, while the private property contains a small area of phytotelms located between a soy plantation and a highway. The limited abundance, distribution, and behavioral information regarding phytotelm-breeding *Melanophryniscus*, including *M. vilavelhensis*, highlight their high conservation value and the need to protect their respective endemic areas (Steinbach-Padilha, 2008; Bornschein *et al.*, 2015) .

Figure 01 –*Melanophryniscus vilavelhensis*. Panels A and B show the dorsal and ventral views of the male and female, respectively. Panel C represents an amplexus in the phytotelm, and panel D shows eggs in the axils of the leaves of a phytotelm.



2.2 STUDY SITES/ DATA COLLECTION

This study was conducted at Vila Velha State Park (VVSP), situated on the second plateau of Paraná in Ponta Grossa, Paraná State, Brazil. Fieldwork was carried out from November 2021 to April 2022 and August 2022 to January 2023, totaling 15 field expeditions. Sampling was conducted from 18:00 to 23:00, involving two researchers, resulting in 32 field nights and 80 hours of work per researcher. The fields expeditions were conducted preferably after heavy or moderate rainfall.

The Vila Velha State Park is a fully protected conservation unit with the primary objective of preserving the natural ecosystem (Brasil, 2000). The Park covers an area of 3,122 hectares and reaches a maximum altitude of 1,068 meters. It is located between coordinates 25°12'34" and 25°15'35" S latitude, and 49°58'04" and 50°03'37" W longitude (Gobbi, 2004). According to Köppen's climate classification, the region experiences a Marine West Coast climate with warm summers (Cfb). This climate is characterized by an average temperature in the hottest month below 22°C, the absence of a defined dry season, and an average temperature in the coldest month below 18°C. The annual precipitation ranges between 1,400 and 1,600 mm, and the average temperature is 17.1-18°C (Nitsche *et al.*, 2019). VVSP is situated within the Atlantic Forest domain and encompasses mosaic vegetation consisting of Araucaria Forest and South Brazilian Grasslands (Cervi *et al.*, 2007).

For this study, we selected two areas within the humid grassland region (Figure 02). These areas are characterized by waterlogged soil due to their location in a drainage valley on the left bank of the Quebra Perna river (Gobbi, 2004) (Figure 03). 'Site 1' was divided into five transects measuring 30 meters each, covering the entire width of the natural grassland. These transects were spaced 15 meters apart from each other. In contrast, due to its small size, 'Site 2' was not divided into transects.

A manual inspection was performed to verify all phytotelms in both areas. Additionally, a combination of visual encounter techniques and audio strip transects was employed to search for vocalization activity (Heyer, 1994). Any individuals found in the occupied plots were captured by hand and placed in plastic bags for subsequent analysis. The phytotelm occupied by the anuran was labeled with an identification. Male individuals were identified based on their vocalizations and the presence of nuptial pads (Jeckel *et al.*, 2019).

Figure 02 – Map of the study area showing the location of Vila Velha State Park (VVSP) in Paraná State, Brazil. Site 1 (blue) and Site 2 (red) are marked on the right side of the figure. The red spots indicate the phytotelms inhabited by *Melanophryniscus vilavelhensis*, and the white lines represent the study transects.

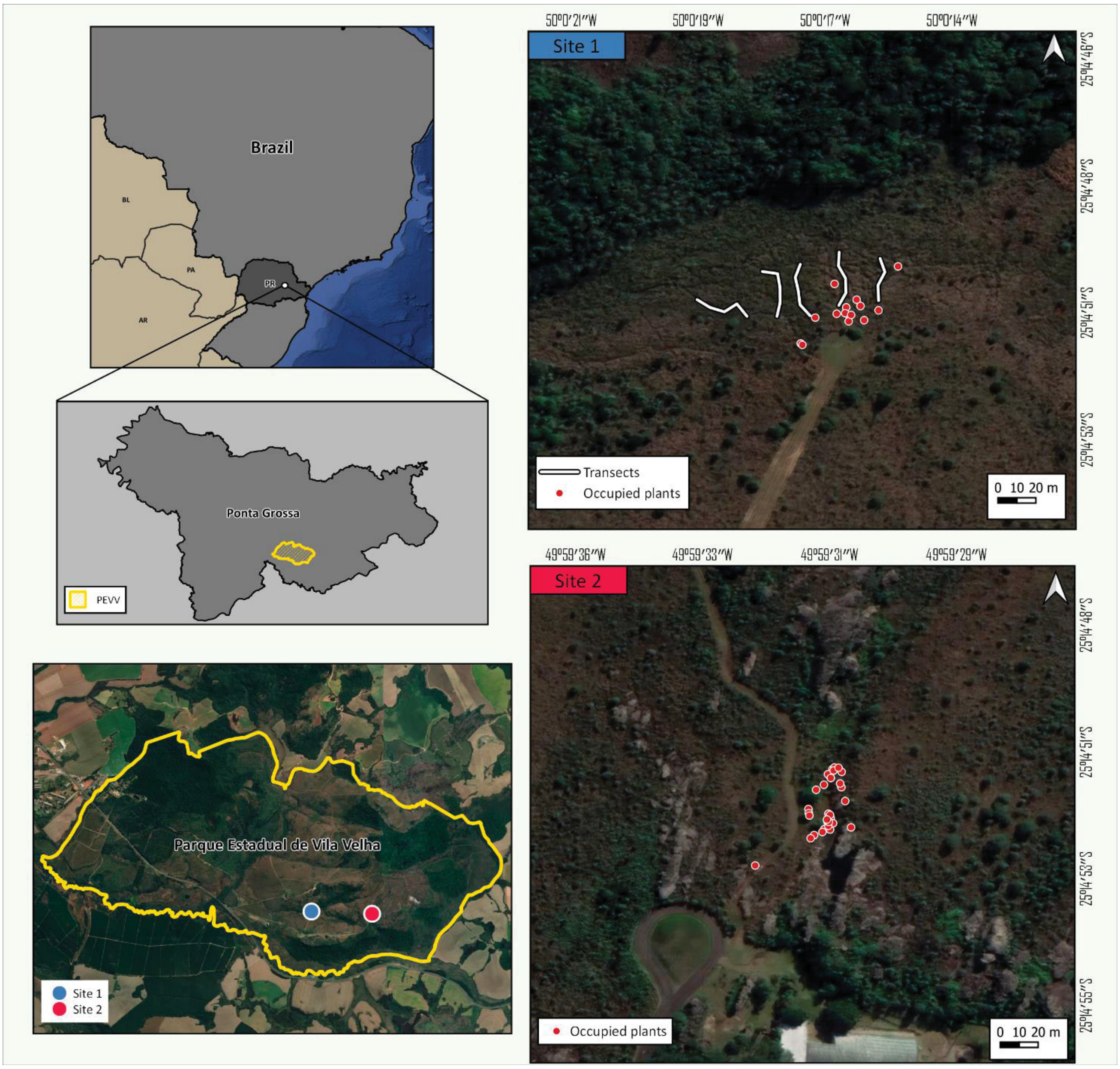
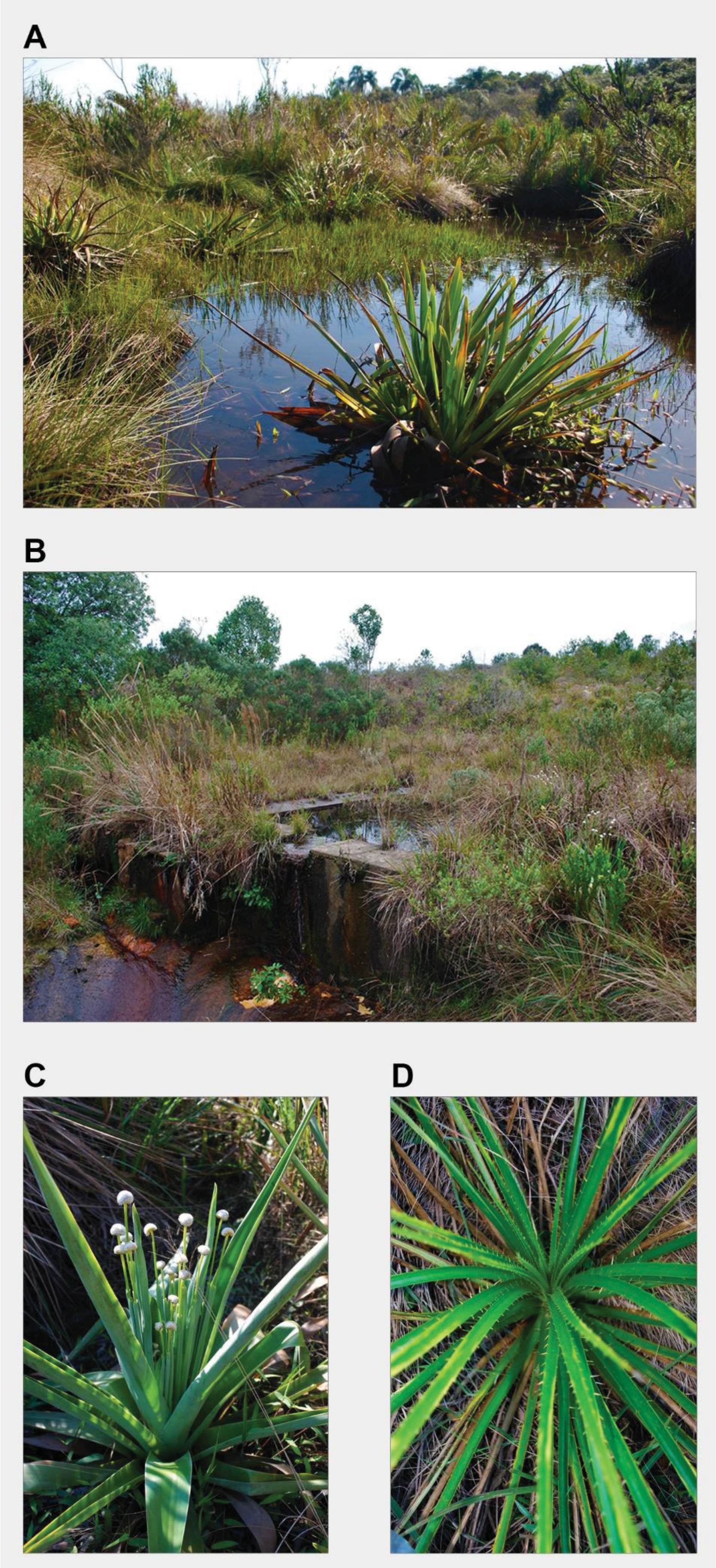


Figure 03 - Study area of Vila Velha State Park (VVSP) in Paraná State, Brazil. Site 1 (A) and Site 2 (B). Phytotelms species used by the anuran *M. vilavelhensis*. *Eriocaulon* sp. (Eriocaulaceae) (C) and *Eryngium* sp. (Apiaceae) (D).



At the Park's lodging, we measured the snout-vent length (SVL) of all *M. vilavelhensis* individuals using a Digimess digital caliper with a precision of 0.01 mm. Photo-identification was employed for individualization in the capture/recapture method. Each individual of *M. vilavelhensis* was photographed in a ventral position from a 15 cm distance using a Nikon Coolpix L820 camera. To identify individuals, the patterns of ventral spots were carefully examined, and individuals with matching patterns were considered the same.

The following morning, we released all individuals back into their original phytotelm with the corresponding label and proceeded to measure nine predictor variables associated with each plant: (1) tank height, (2) tank radius, (3) number of leaf axils, (4) number of phytotelms within a one-meter radius (density), (5) total volume of water in the tank, (6) pH, (7) conductivity, (8) temperature, and (9) total dissolved solids (TDS). Variables 1 to 4 represent the structural characteristics of the plants, while variables 5 to 9 represent the physical-chemical characteristics of the water accumulated in the tank. To measure the tank's height, we considered the distance from the ground to the tip of the highest leaf. To determine the radius, we measured the distance between the center of the tank and the edge of the outermost leaf, using a millimeter ruler. The tank's volume was determined by removing all accumulated water with a pipette, a pear, and a graduated beaker. We utilized a Multi-Parameter PCSTester™ 35 to measure the parameters 6 to 9, with an accuracy of 0.01 pH, 0.5°C, and 1% full scale for conductivity and TDS measurements, alongside a scale ruler. To ensure a standardized sample size and conduct comprehensive analyses we mensured all nine variables for a new phytotelm without previous Anura presence. To select the phytotelm without the anuran, we employed a random sampling approach. We drew a number from 0 to 30, representing the length of the transect, and then selected 1 to the right and 2 to the left of a new drawn number. For example, if the numbers drawn were 15 and 2, we measured the first phytotelm located at right from 15 meters. For Site 2, where no transects were present, the selection process involved drawing a right or left direction and then measure the phytotelm corresponding to the number select. In addition, we recorded the coordinates of all plants (both occupied and unoccupied) using the AlpineQuest Pro GPS app.

The sampling was conducted under the authorization for research in the conservation unit of Paraná, granted by the “Instituto de Água e Terra” (IAT) under number 32.21.

2.3 DATA ANALYSIS

2.3.1 Site selection

Exploratory ecological studies often employ multiple models as potential hypotheses to explain a phenomenon or behavior (Chamberlin, 1897). Consequently, various authors propose the selection of a single model as the "true" hypothesis (Rencher & Pun, 1980; Hurvich & Tsai, 1990; George & McCulloch, 1993). However, alternative approaches suggest the use of a weighted average of parameters of all models, known as "model averaging approach" (Burnham & Anderson, 2002; Lukacs, Burnham & Anderson, 2010; Symonds & Moussalli, 2011; Dormann *et al.*, 2018). This approach considers all models or hypotheses, reducing uncertainty associated with parameter estimates in each model, and is particularly recommended for exploratory analyses with smaller sample sizes (Lukacs *et al.*, 2010; Dormann *et al.*, 2018).

In this study, we selected morphometric variables of the plant (tank height, tank radius, number of leaves, and density of phytotelms) and physical-chemical variables of the water (volume, temperature, pH, and total dissolved solids - TDS) as predictor variables for the response variables of presence/absence of anurans, eggs, and tadpoles, following similar approaches (De Oliveira & Navas, 2004; Domingos *et al.*, 2015; Cunha & Napoli, 2016; Mageski *et al.*, 2016). However, conductivity was not included in the analyses due to its high correlation with TDS ($r=0.945$; $p=2.2e-16$).

To calculate the probability of anuran occurrence, egg presence, and tadpole presence as functions of morphometric and physicochemical variables, we fit generalized linear models (GLMs) with a binomial distribution and a logit link function. We considered the biological relevance of the candidate hypotheses in to construct the set of candidate models, not using all possibilities of model formulation. We fit a full model to assess the probability of occurrence of the adult anurans, eggs, and tadpoles of *M. vilavelhensis,* incorporating additive variables (sampling site, temperature, tank height and radius, number of leaves, and density), two-variable interactions (density and site; volume and radius), and a three-variable interaction (volume, pH, and TDS). We then selected the best models based on Akaike criteria (delta AICc less than or equal to 2). Additionally, we performed a model averaging of the parameters from all candidate models to determine which structural and water quality variables of the phytotelm are associated with habitat selection by *M. vilavelhensis*. For these analyses, we utilized the MuMIn package (Bartoń, 2022). This package allowed us to perform model selection and model averaging. To visualize the effects of the selected predictor variables from all possible models, we generated effects plots using the effects package (Fox & Weisberg, 2018). These plots provided a graphical representation of the relationships between the predictor variables and the response variables, aiding in the interpretation of the results.

Among the selected pool of plants, we focused on those where tadpoles/eggs were present to determinate the factors associated with effective breeding of *M. vilavelhensis*, Our full model included the occurrence of eggs and tadpoles as a function of additive variables (site, temperature, height and radius of the tank, number of leaves, and density), interactions between two variables (density and site; volume and radius), and an interaction involving three physicochemical variables (volume, pH, and TDS). We proceed with the identical process outlined in the preceding analysis.

Additionally, we explored the influence of environmental variables on the effective reproduction of the anurans. The environmental variables, including average temperature (°C), total solar radiation (W/m²), accumulated precipitation (mm), and relative humidity (%), were obtained from the SIMEPAR (Paraná Meteorologic System) at the Ponta Grossa weather station in Vila Velha State Park. To incorporate these environmental variables into our analysis, we calculated the average values for the three previous days as well as for all the days of each field expedition. This allowed us to account for the climatic conditions leading up to and during the sampling period. Similar to the previous analysis, we employed a generalized linear model (GLM) with errors following a binomial distribution and a logit link. To test for an influence of the environmental variables on the effective reproduction of *M. vilavelhensis*, our full model was fit with the probability of the presence of eggs or tadpoles as a function of additive variables including temperature, humidity, and precipitation. Radiation was not included in the analyses due to its correlation with temperature (r= 0.706; p= 0.003). The same model selection and averaging procedures were applied.

Finally, we examined the relationship between the body size of male *M. vilavelhensis*, measured as snout-to-vent length (SVL), and the selected predictor variables using simple linear regressions. This analysis allowed us to investigate the potential influence of these variables on the body size of the individuals.

All statistical analyses were conducted using R within the R Studio software (R Core Team, 2022).

2.3.2 Population dynamics

For the population dynamics analysis, we extended our investigation by utilizing the same environmental variables obtained from SIMEPAR. The goal was to assess demographic parameters, the apparent survival (ϕ) and capturability (p), of *M. vilavelhensis* population using a Cormack-Jolly-Seber (CJS) model for a closed population (White & Burnham, 1999).To fit the model, we incorporated the time intervals between the fifteen collection expeditions. In order to evaluate the goodness-of-fit (GOF), we employed adherence tests using the "release.gof" from the RMark package in R (Laake, 2013). Our models were developed with constant ϕ and p and considering both temporal factors and environmental climatic variables such as temperature, precipitation, and humidity. To ensure comparability, we standardized all climatic variables using z-scores. To determine the most suitable models, we employed a selection criterion where models with a delta AICc less than or equal to 2 were considered of equal evidence (Burnham & Anderson, 2002). All statistical analyses were conducted using R within the R Studio software (R Core Team, 2022).

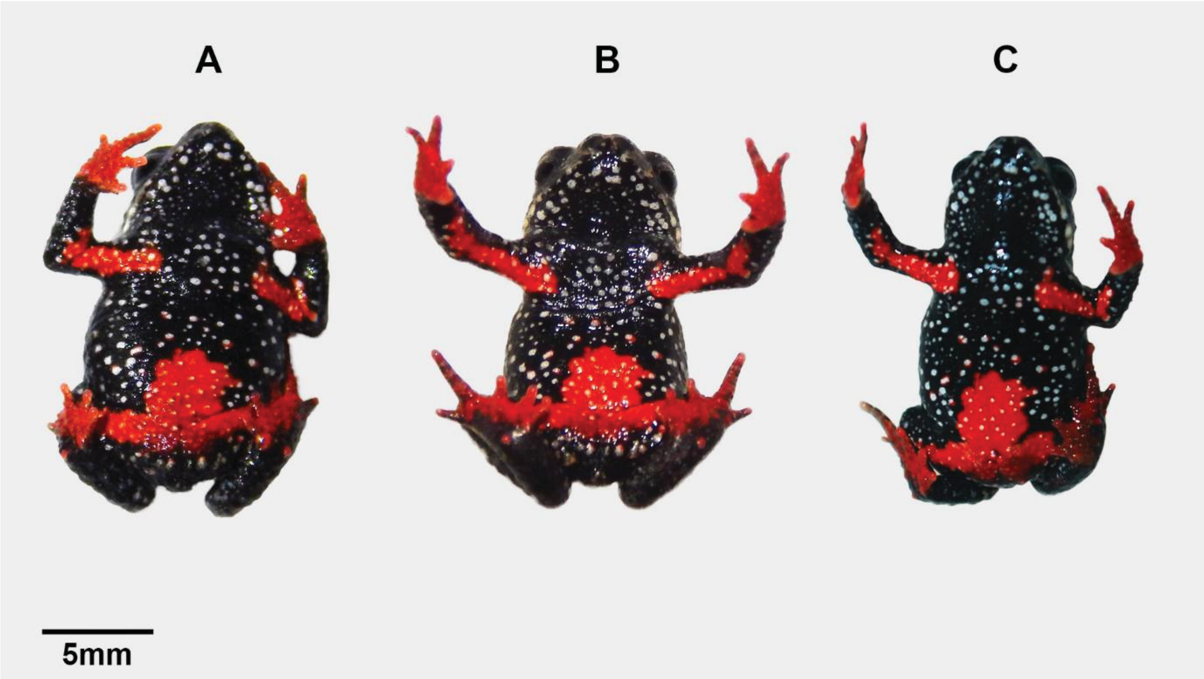
3 RESULTS

3.1 NATURAL HISTORY AND SPATIAL DISTRIBUTION

Males of *M. vilavelhensis* call from within phytotelms filled with water (N=83) with only one exception of a vocalizing male within a waterless phytotelm. When males are in amplexus, they don't emit any call. In total 62 individuals were identified, 55 males (SVL 12.08 - 15.73) and 7 females (SVL 14.71 - 18.33). Two males were observed in a single phytotelm on one occasion, with one of them in amplexus with a female. A total of five amplexus were recorded, involving different male and female pairs. Tadpoles or eggs were found in 47 (55.95%) out of 84 phytotelms where calling males were present. Additionally, 10 phytotelms (10.64%) contained only tadpoles/eggs without any sampling of a calling male before. Overall, a total of 94 phytotelms were utilized by *M. vilavelhensis*. The months of August, October, and November had the highest number of individuals sampled, which also corresponded to the months with the highest abundance of phytotelms containing tadpoles and/or eggs.

A total of 20 males (36.36%) were recaptured during the study period, while only 1 female (14.28%) was recaptured. Only one individual was recaptured three times during the study (Figure 04). The maximum time between capture and recapture was 276 days, with two individuals reaching this time interval. The distances traveled by the individuals varied from 0 to 32 meters, with a mean distance of 6 ± 9.93 meters. Among the thirty recapture events, thirteen (43.33%) occurred on the same plant where the anuran was previously found.

Figure 04 – An individual of *M. vilavelhensis* recaptured three times during the study. In A and B, the individual was captured on two occasions in August 2022, and in panel C, it was captured in January 2023.



Regarding the utilization of phytotelms, most anurans (74.2%) were observed using only one phytotelm, while a smaller proportion (25.8%) utilized two phytotelms. A similar pattern was observed for the number of anurans occupying the same plant (although not simultaneously). Specifically, the majority of phytotelms (74.5%) harbored only one individual, while some phytotelms (23.7%) were occupied by two to three individuals. Notably, there was one particular phytotelm that accommodated four distinct individuals of *M. vilavelhensis*.

3.2 SITE SELECTION

A total of 94 plants with and without (control) *M. vilavelhensis* were measured (Table 01). From the full model to assess the probability of occurrence of the adult anurans, we derived 20 candidate models (hypotheses), and we selected the top 8 models with equal evidence (Table 02). The eight best models collectively explain 71.5% of the relative likelihood of being the best model and all showed the importance of plant architecture and plant density (Table 02). No one had variables associated with physico-chemical properties.

Table 01 – Mean trait values (±SD) and range of all variables measured from phytotelms (N) with and without use by *M. vilavelhensis*. Phytotelms measured in the two sampling site from VVSP between the years 2021 and 2023. N: number of phytotelms; SD: standard deviation; TDS: Total dissolved solids; *number of phytotelms within a one-meter radius.

	With *M. vilavelhensis*				Without *M. vilavelhensis*			
Variables	**N**	**Mean**	**SD**	**Range**	**N**	**Mean**	**SD**	**Range**
Height (cm)	94	53.89	17.22	(23-117)	94	47.5	15.85	(12-80)
Radius (cm)	94	5.80	3.11	(2-22)	94	4.915	2.55	(1-13)
Number of leaf axils	93	23.59	8.15	(11-53)	94	23.78	13.16	(9-69)
Density*	94	3	2.96	(0-16)	94	7.074	3.57	(0-25)
Volume (ml)	93	36.72	33.66	(0-200)	88	23.53	31.51	(0-140)
pH	86	6.29	0.8	(4.15-8.19)	53	5.82	0.95	(4.06-7.92)
Conductivity (	87	90.82	84.7	(0.20-406)	53	63.5	56.1	(0.4-270)
Temperature (°C)	87	22	4.76	(14.8-33)	54	24.02	4.89	(14.6-35.1)
TDS (ppm)	87	69.19	68.1	(0.20-288)	53	44.7	40.82	(0.3-206)

TABELA 02 – Model selection and parameter estimates for the occurrence of *M. vilavelhensis* with eight best models select by delta AICc less or equal than 2. Global model: occurrence of *M. vilavelhensis* ~ site + temperature + height + radius + number of leaves + density + density*site + volume*radius + volume*pH*TDS. Abbreviations: Int: Intercept; df: Degrees of freedom; AICc: Akaike Information Criterion.

Model	(Int)	Height	Site	Density	Radius	df	AICc	delta	weight
~ Height + Density + Radius	0.96	-0.015		0.084	-0.08865	4	257.1	0.00	0.130
~ Height + Density	0.78	-0.020		0.076		3	257.3	0.12	0.122
~ Density + Radius	0.28			0.100	-0.12130	3	257.5	0.37	0.108
~ Height	1.20	-0.023				2	257.7	0.58	0.097
~ Height + Radius	1.38	-0.019			-0.07506	3	258.1	0.98	0.080
~ Height + Site + Density + Radius	0.94	-0.015	+	0.087	-0.10530	5	258.4	1.23	0.070
~ Site + Density + Radius	0.27		+	0.104	-0.13810	4	258.7	1.56	0.060
~ Height + Site + Density	0.76	-0.021	+	0.077		4	259.1	1.98	0.048

Using a multimodel averaging approach leads to the same conclusion, all selected variables belong to plant architecture (Table 03)

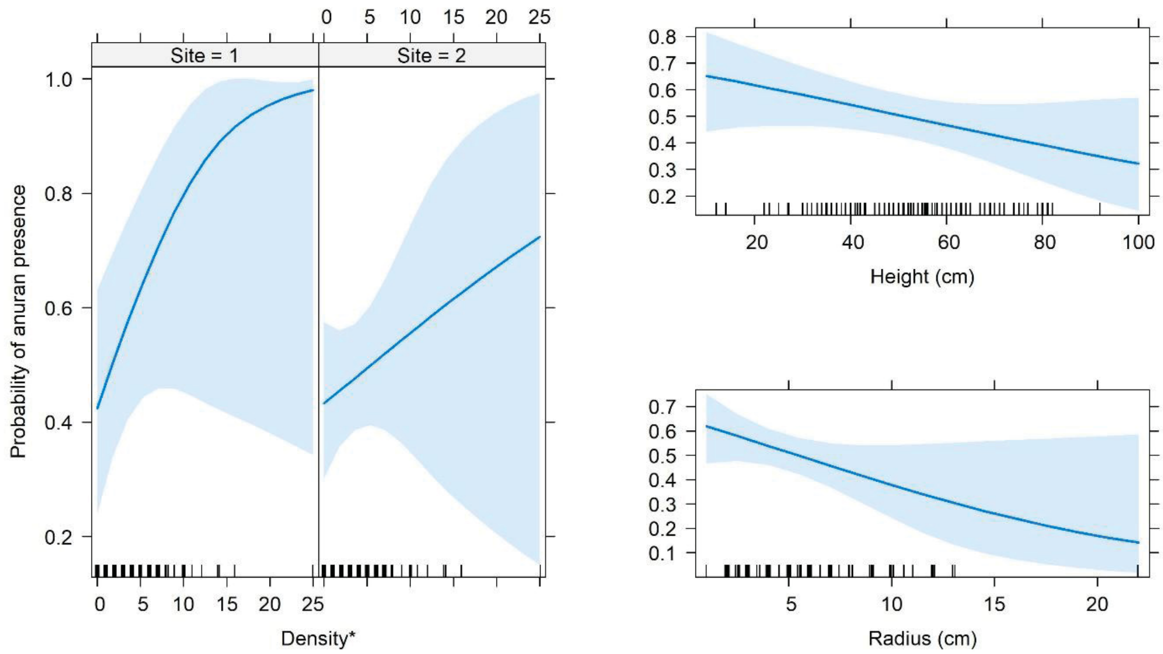
Table 03: Conditional model-averaged estimates from all candidate models for the occurrence of *M. vilavelhensis*. The table presents coefficient estimates for the selected parameters. Abbreviations: Std. Error: standard error; N containing models: number of containing models.

Parameters	Estimate	Std. Error	Sum of weights	N containing models
Intercept	0.87	0.71		
Height	-0.02	0.01	0.70	10
Density	0.10	0.07	0.69	12
Radius	-0.11	0.06	0.61	10
Site	-0.17	0.45	0.40	12

Parameters	Estimate	Std. Error	Sum of weights	N containing models
Site: Density	-0.12	0.12	0.11	4

Overall, the size of the tank, indicated by its radius and height, along with the density of phytotelm occupants (based on the sampling site), are key variables that influence the probability of occurrence for *M. vilavelhensis* individuals or life stages within a phytotelm. In summary, *M. vilavelhensis* tends to select phytotelms with smaller dimensions (lower height and radius) and a higher density of individuals within clustered plants (Figure 05).

Figure 05 – Effects of selected variables on the presence of *M. vilavelhensis* response variable. Probability of anuran presence (0 – 1) depending on density*, height and radius of the tank. Density is divided between the two sampling sites, whereas height and radius encompass both locations. *number of phytotelms within a one-meter radius.



From the full model to determine which variables influence reproductive site choice, we generated 20 candidate hypotheses, of which 3 were selected based on the AICc (see Table 04).

Table 04 – Model selection and parameter estimates for the occurrence of effective reproduction with tree best models select by delta AICc less or equal than 2. Global model: occurrence of tadpoles or/and eggs ~ site + temperature + height + radius + number of leaves + density + density*site + volume*radius + volume*pH*TDS. Abbreviations: Int: Intercept; df: Degrees of freedom; AICc: Akaike Information Criterion.

Model	(Int)	Height	Site	Density	Radius	df	AICc	delta	weight
~ Height + Radius	-0.65	0.04			-0.22	3	126.0	0.00	0.29

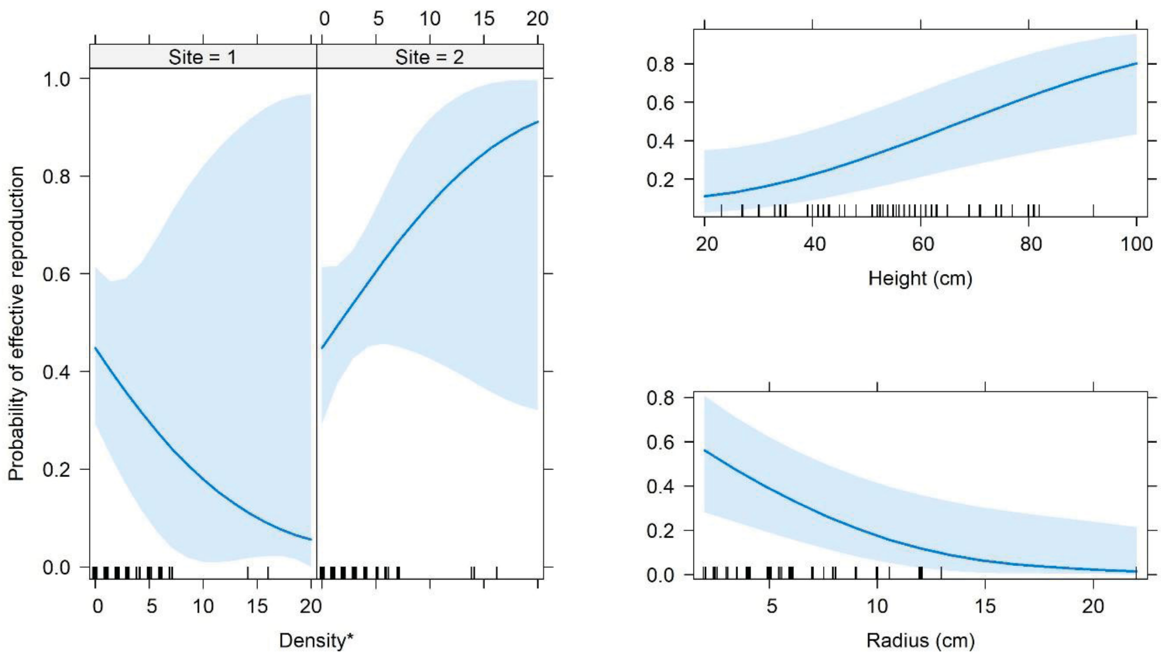
Model	(Int)	Height	Site	Density	Radius	df	AICc	delta	weight
~ Height + Site + Radius	-1.45	0.04	+		-0.19	4	126.5	0.56	0.22
~ Height + Density + Radius	-1.00	0.04		0.06	-0.22	4	127.6	1.60	0.13

Model averaging (see Table 05) with all candidate models indicates that the radius and the height of the tank, as well as density and sampling site, are the variables that significantly influence the probability of effective reproduction (presence of tadpoles or eggs) occurring in a given phytotelm. However, these variables have different effects on the occurrence (Figure 06).

Table 05 – Conditional model-averaged estimates from all candidate models for the effective reproduction of *M. vilavelhensis*. The table presents coefficient estimates for the selected parameters. Abbreviations: Std. Error: standard error; N containing models: number of containing models.

Parameters	Estimate	Std. Error	Sum of weights	N containing models
Intercept	-1.035	1.09		
Height	0.039	0.01	0.91	10
Density	-0.003	0.14	0.36	12
Radius	-0.21	0.09	0.87	10
Site	0.58	0.67	0.50	12
Site: Density	0.26	0.21	0.10	4

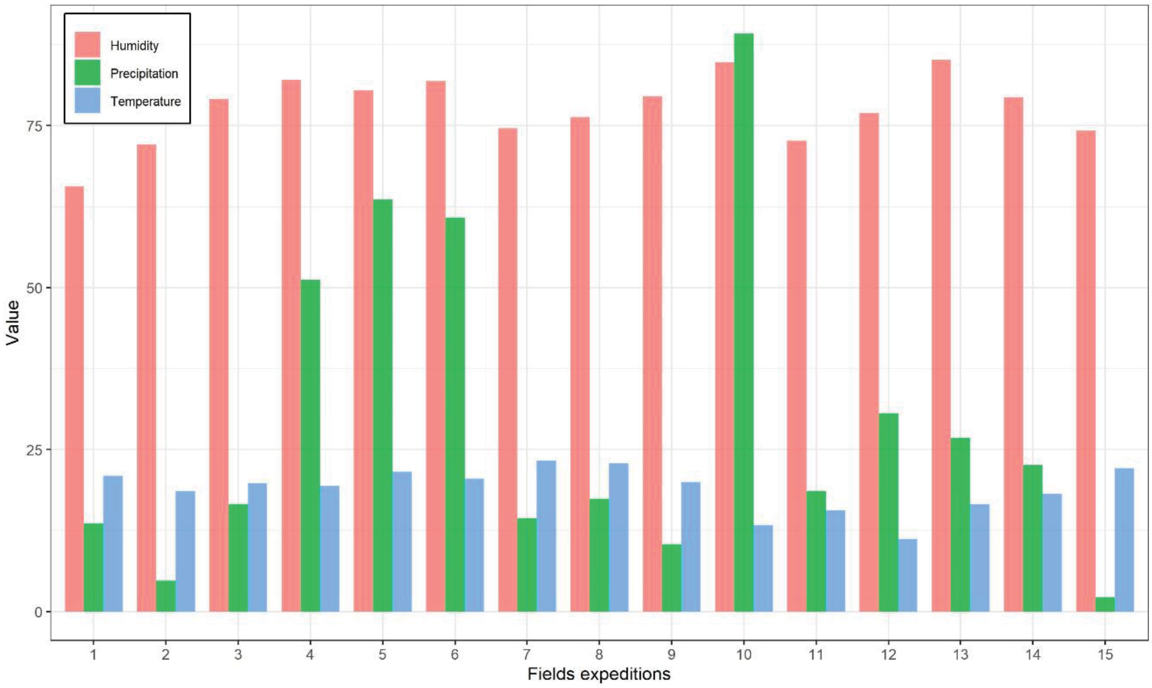
Figure 06 – Effects of selected variables on the effective reproduction response variable. Probability of tadpoles or egg presence (0 – 1) depending on density*, height and radius of the tank. Density is divided between the two sampling sites, whereas height and radius encompass both locations. *number of phytotelms within a one-meter radius.



During the study period, rainfall varied from 2mm to 89.2mm, with an average of 18.6mm. The temperature ranged from 11.24°C to 23.28°C, with an average of

18.94°C, and the humidity ranged from 65.65% to 85.13%, with an average of 77.64% (Figure 07).

Figure 07 – Variation of climatic parameters during the field expeditions. Field 1 was conducted in November 2021; Field 2 in December 2021; Fields 3, 4, 5, and 6 in January 2022; Field 7 in February 2022; Field 8 in March 2022; Field 9 in April 2022; Fields 10, 11, and 12 in August 2022; Field 13 in October 2022; Field 14 in November 2022; and Field 15 in January 2023. Colors represent different climatic variables: relative humidity (%) in pink, accumulated precipitation (mm) in green, and average temperature (°C) in blue.



From the full model to access the influence of environmental variables on effective reproduction, we fit 8 candidate models (hypotheses) and selected the 2 best models based on their similar evidence (delta AICc less than or equal to 2). Temperature is a shared variable in both models, indicating its importanceto the effective reproduction of anurans. Additionally, the second model incorporates the influence of precipitation on reproductive success. The two models differ by 0.9 in terms of delta AICC, indicating a similar level of information (Table 06).

Table 06 – Model selection and parameter estimates for the occurrence of effective reproduction with two best models select by delta AICc less or equal than 2. Global model: occurrence of tadpole and/or eggs ~ temperature + humidity + precipitation. Abbreviations: Int: Intercept; df: Degrees of freedom; AICc: Akaike Information Criterion.

Model	(Int)	Precipitation	Temperature	df	AICc	delta	weight
~ Temperature	9.26		-0.47	2	20.8	0.00	0.396
~ Precipitation + Temperature	14.03	-0.047	-0.65	3	21.7	0.9	0.252

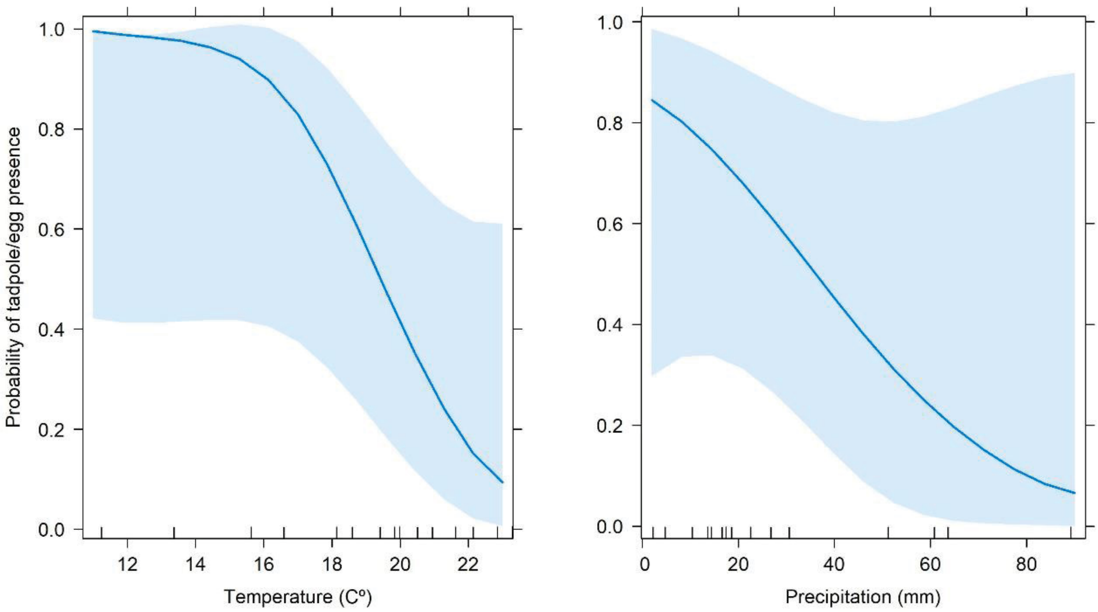
The two best models together have a relative likelihood of 64.8% for being the best model. To estimate the parameters, we performed a model averaging with all candidate models (Table 07).

Table 07 – Conditional model-averaged estimates from all candidate models for the effective reproduction of *M. vilavelhensis*. The table presents coefficient estimates for the selected parameters. Abbreviations: Std. Error: standard error; N containing models: number of containing models.

Parameters	Estimate	Std. Error	Sum of weights	N containing models
Intercept	9.47242	9.34924		
Temperature	-0.54504	0.31556	0.79	4
Precipitation	-0.04262	0.03631	0.34	4
Humidity	-0.04212	0.14239	0.18	4

Temperature and precipitation are the variables that influence the probability of effective reproduction of *M. vilavelhensis*. Although relative humidity was included in the model averaging it was not in the best models because its small effect. In summary, *M. vilavelhensis* tends to reproduce in days with moderate temperatures (a significantly higher chance at temperatures below 20C°) and lower rainfall (below 30mm), based on the climatic data from samplings (Figure 8).

Figure 8 – Effects of selected variables on the effective reproduction response variable. Probability of tadpoles or egg presence (0 – 1) depending on temperature and precipitation.

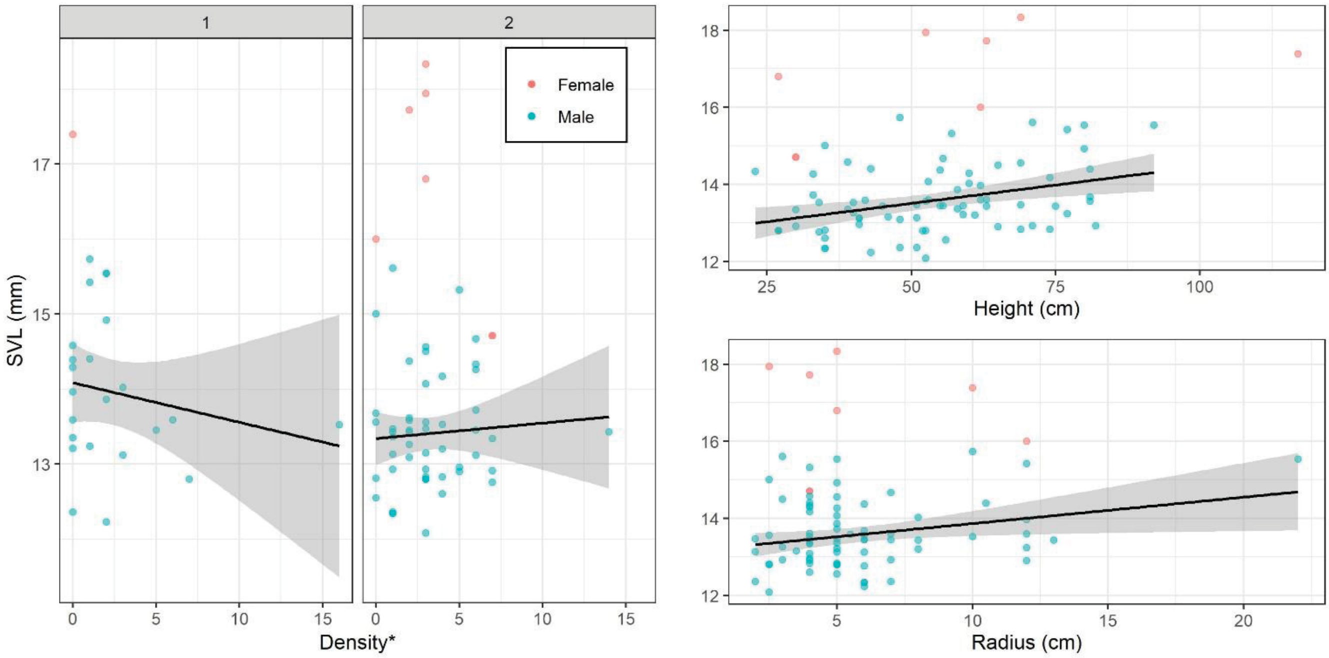


Plant morphological characteristics play a significant role in determining occupancy patterns based on male size. Our findings indicate that larger individuals tend to inhabit taller plants (β = 0.01 ± 0.005) with larger tanks (β = 0.06 ± 0.03) (Table 08). However, we did not find a significant linear relationship between plant density and male size. Females, on the other hand, do not show any relationship with the morphological characteristics of the phytotelms (Figure 9).

Table 08 – Linear regression analysis of the selected parameters with male size (snout to vent length - SVL) of *M. vilavelhensis* from the two sampling sites. * ($P \leq 0.05$)

Parameters	Estimate	Std. Error	P value
Height	0.019	0.005	**0.0017***
Density	-0.02	0.03	0.469
Radius	0.07	0.03	**0.0271***

Figure 9 – – Linear regression analysis of the selected parameters with male size (snout to vent length - SVL) of *M. vilavelhensis* from the two sampling sites. *number of phytotelms within a one-meter radius. Females of *M. vilavelhensis* represented in pink dots.



3.3 POPULATION DYNAMICS

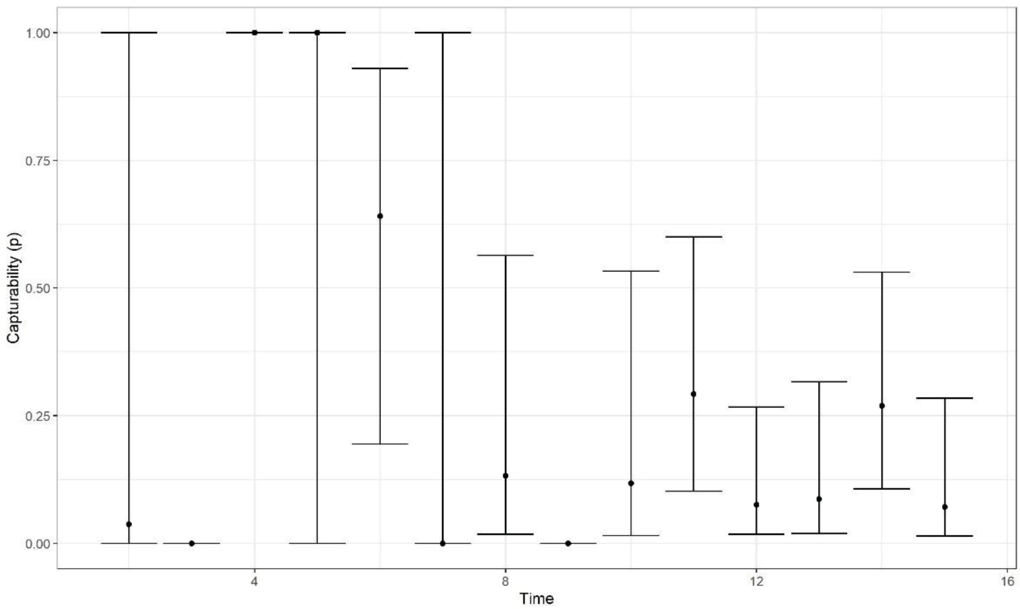
From the analysis of the Cormack Jolly–Seber (CJS) model, 16 candidate hypotheses were generated, and only one model was selected based on the AICc (see Table 09). The selected model has 61.75% of the relative likelihood of being the best model. According to the best model, the apparent survival (ϕ) remained constant across time, while capturability (p) was dependent on time.

Based on the best model, the survival rate between occasions was 0.996 (SE = 0.002, lower 95% CI = 0.988, upper 95% CI = 0.998), and the average survival rate over the course of this study was 0.86. The capturability, in turn, varied from 0 to 1 with a mean of (0.265 ± 0.354) across occasions (Figure 10). Overall, capturability shows stability after the tenth sampling occasion. Before it, there is no clear pattern (Figure 10).

Table 09 – Model selection table for apparent survival (ϕ) and capturability (p) based on CJS analysis of *M. vilavelhensis* from the two sampling sites. The best model was selected by delta AICc less or equal than 2. The table presents the intercept and slope values for each model. Abbreviations: npar: Number of parameters; AICc: Akaike Information Criterion; Prec: precipitation; Temp: Temperature.

Model	npar	AICc	delta	weight
Φ (~1) + p (Time)	9	157.94	0.00	0.62

Figure 10 – Variation of capture rate (p) of *M. vilavelhensis* from the two sampling sites across collection occasions with associated error bars.



4 DISCUSSION

4.1 NATURAL HISTORY AND SPATIAL DISTRIBUTION

Movement patterns can vary among species and environments, as it is correlated with multiple factors. Movement in *M. vilavelhensis* appears to be limited, likely due to the individuals' high site fidelity.

In our study, we found that thirteen (43.33%) of the recapture events occurred on the same plant where the anuran was initially found, demonstrating a high site fidelity by *M. vilavelhensis*. For the *M. vilavelhensis* anurans that dispersed, the distance traveled was less than 35 meters, with an average of 6 ± 9.93 meters. This strategy of remaining in the same location or even returning to it may be advantageous for anurans as it ensures suitable habitat conditions and avoids the risks associated with dispersal, such as desiccation and predation(Semlitsch, 2008). In a study on movement patterns of the *Melanophryniscus montevidensis* population, Pereira and Maneyro (2016) observed that 81.2% of the recaptured individuals were recaptured at the same capture site, indicating a high site fidelity as well and distances of up to 746 meters were observed, although most individuals (71.8%) covered distances less than 100 meters. However, in a study with the phytotelm-breeding anuran *Phyllodytes melanomystax*, a larger species from the Hylidae family (20.0-25.4 mm SVL in males) (Caramaschi, da Silva & de Britto-Pereira, 1992) out of 27 recaptured males, only three were found on the same plant (Cunha & Napoli, 2016). Cunha & Napoli, (2016) reported an even shorter distance traveled, less than 11.5 meters which had an average of 4.30 ± 3.4 meters.

In the present study, although our sampling design was not specifically aimed at evaluating the demographic variation of the anuran throughout the year, we observed a concentration of activity during the months of August, October, and November. These findings are consistent with previous studies conducted in 2010 to 2014 (see supplementary material- Figure 11). The reproductive period of these species is usually associated with the rainy seasons, such as spring and summer in the southern hemisphere, spanning from September to March (Baldo & Basso, 2004; Peloso *et al.*, 2012). However, some species exhibit reproductive events throughout the year, with heavy rainfall as the trigger (Santos *et al.*, 2010; Santos & Grant, 2011; Pereira & Maneyro, 2016). For instance, Pereira and Maneyro (2016) documented reproductive events of *M. montevidensis* in August, October, December, and February, indicating breeding throughout winter, spring, and summer.

As the temporal pattern of reproduction changed from highly concentrated to seasonal, the behavior of males and females may change. Overall, it seems that the reproductive pattern changed from intraspecific competition to female choice because males of *M. vilavelhensis* exhibited chorus behavior, consistently vocalizing in the axils of phytotelm. So, the scramble competition behavior with antagonistic interactions commonly observed in explosive breeding frogs was not observed in *M. vilavelhensis*.

4.2 SITE SELECTION

Site selection in *M. vilavelhensis* exhibits a non-random pattern, with a preference for smaller plants in terms of radius and height, as well as a tendency to choose more clustered plants. Previous studies have also highlighted the significance of phytothelm size in anuran site selection (De Oliveira & Navas, 2004; Pederassi *et al.*, 2012; Ruano-Fajardo, Rovito & Ladle, 2014; Mageski *et al.*, 2016). While phytotelm size is consistently linked to water storage capacity, we hypothesize that the selection of smaller plants by *M. vilavelhensis* is influenced by a morphological constraint. Given its small size, the anuran likely chooses plants that can be accessed with minimal energy expenditure (Alves-Silva & da Silva, 2009). The positive correlation between the snout-to-vent length (SVL) of male *M. vilavelhensis* and the height and radius of the plant supports the role of this morphological constraint on site selection. Mageski et al. (2017) also reported a preference for smaller plants in the phytotelmata anuran *Phyllodytes luteolus*, which facilitates adult movement during escape situations and provides quick and easy access to the plant. Additionally, other studies have found that anurans prefer plants closer to the ground (De Oliveira & Navas, 2004; Poelman, van Wijngaarden & Raaijmakers, 2013). Regarding the density of neighboring conspecifics in phytotelm, Cunha & Napoli (2016) found that the anuran *Phyllodytes melanomystax* also displays a preference for clustered plants. The density was influenced by the sampling site in this study, as there was a difference in phytotelm availability between site 1 and site 2. Site 1 had a higher number of available plants compared to site 2. Selecting clustered plants allows males to expand their habitat beyond a single phytotelm to a cluster of plants, increasing their access to potential breeding sites (Cunha & Napoli, 2016).

The physical-chemical parameters of the water did not influence in the selection of phytotelm by *M. vilavelhensis*. However, other studies have suggested that factors such as pH (De Oliveira & Navas, 2004; Domingos *et al.*, 2015), temperature (Ruano-

Fajardo *et al.*, 2014), and the amount of debris inside the tank (Cunha & Napoli, 2016)may influence site selection by anurans. Nevertheless, these authors also emphasize the challenge of determining whether these variables were initially chosen by the anuran or if the presence and activity of the animal subsequently modify the physicochemical parameters of the habitat. We lean towards the latter possibility. Field studies involving multiple and uncontrollable variables can often lead to confusion regarding cause-and-effect processes. Additionally, these factors often exhibit substantial variation over short time periods due to environmental fluctuations such as rainfall, desiccation, and radiation. Therefore, it is unlikely that the anuran's selection is primarily based on such variablefactors, and it is more plausible that *M. vilavelhensis* primarily selects based on fixed variables related to the phytotelm’s structure.

Among the plants chosen by *M. vilavelhensis* as vocalization sites, there were also instances of effective reproduction, indicated by the presence of eggs and/or tadpoles. The probability of effective reproduction occurring in these phytotelms is influenced by the same factors that influence site selection, namely density, height, and radius of the pond. The differences in density observed between sites are likely due to variations in the availability of phytothelms. Additionally, larger plants have a higher probability of supporting effective reproduction among the chosen plants. This suggests that, among the accessible plants, those with larger ponds are more likely to facilitate reproduction in *M. vilavelhensis*.

Regarding the influence of environmental variables on the effective reproduction of anurans, the model reveals a significant relationship between the presence of tadpoles and eggs and temperature and rainfall. This finding aligns with expectations for amphibian anurans, as their reproductive activity is known to be influenced by external factors and environmental variables such as temperature, humidity, and rainfall (Wells, 2007). The model suggests that the likelihood of finding tadpoles and eggs significantly decreases at temperatures exceeding 20°C, which may be attributed to the increased risk of desiccation under higher temperatures. Surprisingly, the model also indicates that the probability of finding tadpoles and eggs is higher during periods of lower rainfall, specifically up to 30 mm. This finding contrasts with the common assumption that anura reproduction is linked to rainfall. We hypothesize that excessive rainfall may result in the overflowing of water in the phytotelm tanks, potentially displacing or harming the anuran offspring.

4.3 POPULATION DYNAMICS

In this study, *M. vilavelhensis* exhibited high and consistent survival rates across collection occasions and the capturability of individuals showed considerable variation over time. It is important to highlight that constructing individual recapture histories for population dynamics analysis requires a well-designed sampling design spanning a sufficient time interval. In our study, the field occasions were not pre-established but rather conducted after rains to increase the chances of encountering the target species. This approach, while increasing encounter probability, may potentially mask long-term demographic patterns (Vasconcellos & Colli, 2009).

Comparisons with other species within the genus reveal that survival rates can vary significantly depending on factors such as sex and season. For instance, a study on *M. montevidensis* demonstrated distinct survival rates among males, females, and juveniles, which were influenced by season (Bardier *et al.*, 2019). Male survival rates in the cold season exceeded 80%, while in the warm season, survival dropped to 60%. In contrast, our findings showed a remarkably high survival rate of 99% for *M. vilavelhensis*. It is important to consider that this high survival rate is partly influenced by the recapture of individuals between large time intervals, such as between the first and last collections.

Regarding capturability, our study observed a higher value (0.265 ± 0.354). Notably, the variation in capturability followed the sampling events, as evidenced by the absence of recapture between certain occasions and the recapture of all individuals in other occasions. Therefore, to obtain more reliable capture and survival rates for *M. vilavelhensis*, it is crucial to implement an appropriate sampling design that allows for a comprehensive analysis of population dynamics.

5 CONCLUDING REMARKS

In conclusion, our findings suggest that the phytotelm-breeding anuran, *M. vilavelhensis*, exhibits strong site fidelity and selective microhabitat occupancy based on morphological constraints. Additionally, these anurans demonstrate a close association with environmental factors, particularly temperature and rainfall accumulation. It is important to highlight that amphibians are currently facing significant threats, primarily due to habitat loss and disease, making them one of the most endangered groups of vertebrates (IUCN, 2022). Studies that delve into the natural history and demographics of these species provide valuable knowledge and insights for establishing effective conservation strategies. By contributing new data on this previously understudied phytotelm-breeding species, we aim to contribute to the conservation efforts of the fauna within the campos gerais region.

REFERENCES

Alves-Silva, R. & da Silva, H.R. (2009). Life in bromeliads: reproductive behaviour and the monophyly of the Scinax perpusillus species group (Anura: Hylidae). *J Nat Hist* **43**, 205–217.

Baldo, D. & Basso, N.G. (2004). A new species of Melanophryniscus Gallardo, 1961 (Anura: Bufonidae), with comments on the species of the genus reported for Misiones, northeastern Argentina. *J Herpetol* **38**, 393–403.

Bardier, C., Martínez-Latorraca, N., Porley, J.L., Bortolini, S.V., Cabrera Alonzo, N., Maneyro, R. & Toledo Bardier, L.F. (2019). Seasonal demography of the threatened Redbelly Toad Melanophryniscus montevidensis in a protected area of Uruguay Canadian Journal of Zoology. *Can J Zool* **97**, 131–147.

Bartoń, K. (2022). Model selection and multimodel inference: a practical information-theoretic approach.

Beebee, T.J.C. & Griffiths, R.A. (2005). The amphibian decline crisis: A watershed for conservation biology? *Biol Conserv* **125**, 271–285.

Bornschein, M.R., Firkowski, C.R., Baldo, D., Ribeiro, L.F., Belmonte-Lopes, R., Corrêa, L., Morato, S.A.A. & Pie, M.R. (2015). Three new species of phytotelm-breeding melanophryniscus from the atlantic rainforest of southern Brazil (Anura: Bufonidae). *PLoS One* **10**.

Burnham, K.P. & Anderson, D.R. (2002). *Model selection and multimodel inference: a practical information-theoretic approach. A practical information-theoretic approach*. 2nd ed. New York: Springer-Verlag.

Caorsi, V.Z. (2011). Comportamento Reprodutivo de Melanophryniscus cambaraensis (Anura: Bufonidae) na Floresta Nacional de São Francisco de Paula, Rio Grande do Sul, Brasil.

Caramaschi, U., da Silva, H.R. & de Britto-Pereira, M.C. (1992). A new species of Phyllodytes (Anura, Hylidae) from southern Bahia, Brazil. *Copeia* 187–191.

Castro, J.C. & Pinto, A.T. (2000). Qualidade da Água em Tanques de Girinos de Rã-Touro, Rana catesbeiana Shaw, 1802, Cultivados em Diferentes Densidades de Estocagem. *Revista Brasileira de Zootecnia* **29**, 1903–1911.

Céréghino, R., Pillar, V.D., Srivastava, D.S., de Omena, P.M., MacDonald, A.A.M., Barberis, I.M., Corbara, B., Guzman, L.M., Leroy, C., Bautista, F.O., Romero, G.Q., Trzcinski, M.K., Kratina, P., Debastiani, V.J., Gonçalves, A.Z., Marino, N.A.C., Farjalla, V.F., Richardson, B.A., Richardson, M.J., Dézerald, O., Gilbert, B., Petermann, J., Talaga, S., Piccoli, G.C.O., Jocqué, M. & Montero, G. (2018). Constraints on the functional trait space of aquatic invertebrates in bromeliads. *Funct Ecol* **32**, 2435–2447.

Cervi, A.C., Von Linsingen, L., Hatschbach, G. & Ribas, O.S. (2007). *A vegetação do Parque Estadual de Vila Velha, Município de Ponta Grossa, Paraná, Brasil*. Curitiba.

Chamberlin, T.C. (1897). The Method of Multiple Working Hypotheses. *J Geol* **5**, 837–848.

Cunha, M.S. & Napoli, M.F. (2016). Calling site selection by the bromeliad-dwelling treefrog Phyllodytes melanomystax (Amphibia: Anura: Hylidae) in a coastal sand dune habitat. *Stud Neotrop Fauna Environ* **51**, 144–151.

Domingos, F.M.C.B., Arantes, Í.C., Cavalcanti, D.R. & Jotta, P.A.C. V. (2015). Shelter from the sand: microhabitat selection by the bromelicolous tree frog Scinax cuspidatus (Anura, Hylidae) in a Brazilian restinga. *North West J Zool* **11**, 27–33.

Dormann, C.F., Calabrese, J.M., Guillera-Arroita, G., Matechou, E., Bahn, V., Barton, K., Beale, C.M., Ciuti, S., Elith, J., Gerstner, K., Guelat, J., Keil, P., Lahoz-Monfort, J.J., Pollock, L.J., Reineking, B., Roberts, D.R., Schroder, B., Thuiller, W., Warton, D.I., Wintle, B.A., Wood, S.N., Wuest, R.O. & Hartig, F. (2018). Model averaging in ecology: a review of Bayesian, information-theoretic, and tactical approaches for predictive inference. *Ecol Monogr* **88**, 485–504.

Duellman, W.E. & Trueb, L. (1994). *Biology of amphibians*. JHU press.

Fox, J. & Weisberg, S. (2018). Visualizing Fit and Lack of Fit in Complex Regression Models with Predictor Effect Plots and Partial Residuals. *J Stat Softw* **87**, 1–27.

Fretwell, S.D. & Calver, J.S. (1969). On territorial behavior and other factors influencing habitat distribution in birds: II. Sex ratio variation in the Dickcissel (Spiza americana Gmel). *Acta Biotheor* **19**, 37–44.

Frost, D.R. (2023). Amphibian Species of the World: an Online Reference. [WWW Document]. *Version 6.2*. URL https://amphibiansoftheworld.amnh.org/index.php

George, E.I. & McCulloch, R.E. (1993). Variable selection via Gibbs sampling. *J Am Stat Assoc* **88**, 881–889.

Gobbi, E.F. (2004). Plano de Manejo Parque Estadual de Vila Velha.

Graybeal, A. & Cannatella, D.C. (1995). A New Taxon of Bufonidae from Peru, with Descriptions of Two New Species and a Review of the Phylogenetic Status of Supraspecific Bufonid Taxa. *Herpetologica* **51**, 105–131.

Haddad, C.F.B., Toledo, L.F., Prado, C.P.A., Loebmann, D., Gasparini, J.L. & Sazima, I. (2013). *Guia dos anfíbios da Mata Atlântica: diversidade e biologia*. Anolis Books.

Heyer, W.R. (1994). Standard Techniques for Inventory and Monitoring. In *Measuring and monitoring biological diversity*: 75–141. Smithsonian Institution Press.

Hiert, C. & Moura, M.O. (2010). Abiotic correlates of temporal variation of Hypsiboas leptolineatus (Amphibia: Hylidae). *Zoologia* **27**, 703–708.

Hurvich, C.M. & Tsai, C.-L. (1990). The impact of model selection on inference in linear regression. *Am Stat* **44**, 214–217.

IUCN. (2022). The IUCN Red List of Threatened Species [WWW Document]. *Version 2022-2*. URL https://www.iucnredlist.org

Jaenike, J. & Holt, R.D. (1991). *GENETIC VARIATION FOR HABITAT PREFERENCE: EVIDENCE AND EXPLANATIONS*.

Jeckel, A.M., Caorsi, V.Z., Grant, T. & Borges-Martins, M. (2019). The nuptial pads of melanophryniscus (anura: Bufonidae), with the unexpected occurrence of nuptial-pad-like structures in females of two species. *J Herpetol* **53**, 53–61.

Jorge, J.S., Sales, R.F.D., Santos, R.L. & Freire, E.M.X. (2020). Living among thorns: herpetofaunal community (Anura and Squamata) associated to the rupicolous bromeliad Encholirium spectabile (Pitcairnioideae) in the Brazilian semi-arid Caatinga. *Zoologia* **37**, 1–12.

Kitching, R.L. (2000). Food webs in phytotelmata:“bottom-up” and “top-down” explanations for community structure. *Annu Rev Entomol* **46**, 729–760.

Laake, J.L. (2013). *RMark: An R Interface for analysis of capture-recapture data with MARK*. Seattle, WA.

Langone, J.A., Segalla, M. V., Bornschein, M. & de Sá, R.O. (2008). A new reproductive mode in the genus Melanzophryniscus Gallardo, 1961 (Anura: Bufonidae) with description of a new species from the state of Paraná, Brazil. *South Am J Herpetol* **3**, 1–9.

Lukacs, P.M., Burnham, K.P. & Anderson, D.R. (2010). Model selection bias and Freedman’s paradox. *Ann Inst Stat Math* **62**, 117–125.

Mageski, M.M., Ferreira, R.B., Beard, K.H., Costa, L.C., Jesus, P.R., Medeiros, C.C. & Ferreira, P.D. (2016). Bromeliad Selection by Phyllodytes luteolus (Anura, Hylidae): The Influence of Plant Structure and Water Quality Factors. *J Herpetol* **50**, 108–112.

Matthiopoulos, J., Fieberg, J., Aarts, G., Beyer, H.L., Morales, J.M. & Haydon, D.T. (2015). Establishing the link between habitat selection and animal population dynamics. *Ecol Monogr* **85**, 413–436.

Von May, R., Medina-Müller, M., Donnelly, M.A. & Summers, K. (2009). Breeding-site selection by the poison frog ranitomeya biolat in amazonian bamboo forests: An experimental approach. *Can J Zool* **87**, 453–463.

Morris, D.W. (2003). Toward an ecological synthesis: A case for habitat selection. *Oecologia* **136**, 1–13.

Nitsche, P.R., Camori, P.H., Ricce, W. da S. & Pinto, D.F.L. (2019). *Atlas Climático do Estado do Paraná*. Londrina.

Nunes-de-Almeida, C.H.L., Haddad, C.F.B. & Toledo, L.F. (2021). A revised classification of the amphibian reproductive modes. *Salamandra* **57**, 413–427.

De Oliveira, F.B. & Navas, C.A. (2004). Plant selection and seasonal patterns of vocal activity in two populations of the bromeligen treefrog Scinax perpusillus (Anura, Hylidae). *J Herpetol* **38**, 331–339.

Pederassi, J., Lima, MSCS., Peixoto, OL. & Souza, CAS. (2012). The choice of bromeliads as a microhabitat by Scinax argyreornatus (Anura, Hylidae). *Brazilian Journal of Biology* **72**, 229–233.

Peixoto, O.L. (1975). Associação de anuros a bromeliáceas na Mata Atlântica. *Revista Universidade Rural, Série Ciências da Vida* **17**, 75–83.

Peloso, P.L.V., Faivovich, J., Grant, T., Gasparini, J.L. & Haddad, C.F.B. (2012). An extraordinary new species of melanophryniscus (anura, bufonidae) from Southeastern Brazil. *Am Mus Novit* 1–32.

Pereira, G. & Maneyro, R. (2016). Use of Reproductive Microhabitat by Melanophryniscus montevidensis (Anura: Bufonidae) from Uruguay. *Zoolog Sci* **33**, 337–344.

Poelman, E.H., van Wijngaarden, R.P.A. & Raaijmakers, C.E. (2013). Amazon poison frogs (Ranitomeya amazonica) use different phytotelm characteristics to determine their suitability for egg and tadpole deposition. *Evol Ecol* **27**, 661–674.

Protázio, A. dos S., Protázio, A. dos S., Ribeiro, E. de S., Nogueira, E.M. de S. & de Moura, G.J.B. (2013). The role of bromeliad architecture and abiotic factors in occupation by anurans. *Neotropical Biology and Conservation* **8**, 88–95.

R Core Team. (2022). R: A Language and Environment for Statistical Computing.

Rencher, A.C. & Pun, F.C. (1980). Inflation of R2 in best subset regression. *Technometrics* **22**, 49–53.

Ruano-Fajardo, G., Rovito, S.M. & Ladle, R.J. (2014). Bromeliad Selection by Two Salamander Species in a Harsh Environment. *PLoS One* **9**, e98474.

Rudolf, V.H.W. & Rödel, M.-O. (2005). Oviposition site selection in a complex and variable environment: the role of habitat quality and conspecific cues. *Oecologia* **142**, 316–325.

Sabagh, L.T., Ferreira, R.B. & Rocha, C.F.D. (2017). Host bromeliads and their associated frog species: Further considerations on the importance of species interactions for conservation. *Symbiosis* **73**, 201–211.

Sanches, P.R., Santos, F.P., Gonçalves Corrêa, J., França, P.F., Sousa, J.C., Cantuária, P.C. & Costa-Campos, C.E. (2019). Anurans associated with the bromeliad Bromelia goeldiana on a floodplain in northern Brazil. *Herpetol Notes* **12**, 431–433.

Santos, R.R. & Grant, T. (2011). Diel pattern of migration in a poisonous toad from Brazil and the evolution of chemical defenses in diurnal amphibians. *Evol Ecol* **25**, 249–258.

Santos, R.R., Leonardi, S.B., Caorsi, V.Z. & Grant, T. (2010). Directional orientation of migration in an aseasonal explosive-breeding toad from Brazil. *J Trop Ecol* **26**, 415–421.

Semlitsch, R.D. (2008). Differentiating migration and dispersal processes for pond-breeding amphibians. *J Wildl Manage* **72**, 260–267.

Steinbach-Padilha, G.C. (2008). A new species of Melanophryniscus (Anura, Bufonidae) from the Campos Gerais region of Southern Brazil. *Phyllomedusa* **7**, 99–108.

Symonds, M.R.E. & Moussalli, A. (2011). A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behav Ecol Sociobiol* **65**, 13–21.

Vasconcellos, M.M. & Colli, G.R. (2009). Factors affecting the population dynamics of two toads (Anura: Bufonidae) in a seasonal neotropical savanna. *Copeia* **2009**, 266–276.

Wells, K.D. (2007). *The ecology and behavior of amphibians*. University of Chicago press.

White, G.C. & Burnham, K.P. (1999). Program mark: Survival estimation from populations of marked animals. *Bird Study* **46**, S120-139.

SUPPLEMENTARY MATERIAL

Figure 11 – Temporal variation in abundance of *M. vilavelhensis* during the years of 2010; 2011; 2013 and 2014. Data from unpublished study (Crivellari, 2014)